AQUATIC HABITAT SHIFT ASSESSMENT IN A GROUNDWATER-FED SEMI-ARID STREAM

An investigation into the response of Karoo hydroecology to system variability

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By

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CHAPTER 1: INTRODUCTION

1.1 Introduction to the study

The subject of biological response to changes in aquatic habitat is one which has been well explored in many regions of the world. Examples include work in south east Spain by Mellado Diaz *et al.* (2008) and Oliva-Paterna *et al.* (2003), in western United States of America by Hauer and Lorang (2004), and in West Germany by Meyer *et al.* (2003). Similarly, a number of studies have been conducted in semi-arid regions, exploring elements such as erosion, climate, lithology and landscape formations (e.g. Boardman *et al.*, 2013; Le Maitre *et al.*, 2007; Meyer *et al.*, 2003). However, apart from the study by Uys (1997), and Uys and O'Keeffe (1997), there is a noticeable lack of literature on aquatic habitat shifts in semi-arid stream systems, despite these systems being recognised for their high natural variability. This study provides a base-level approach to conducting habitat shift assessments in a semi-arid stream system and monitoring the hydroecological responses to system variability.

The aim of this study is to investigate the effects of flow, water quality variables and sediment processes on habitat availability and the richness of biota in a groundwater-fed system in the Karoo, South Africa. This research also provides information relevant to other arid and semi-arid regions of the world. These areas are globally characterised by low precipitation and high evapotranspiration rates, and are broadly considered to be comparable in terms of climate, geomorphological processes and hydrology (Tooth, 2013). Current literature often refers to findings from and draws comparisons between arid and semi-arid regions from different areas of a continent (e.g. parts of Africa or North America) (see Patten, 1998; Dean & McDonald, 1994) or different countries (e.g. Australia, South Africa and the USA) (see Boardman *et al.*, 2013; Dickie & Parsons, 2012). Furthermore, frameworks which model predicted ecological responses, such as the Ecological Limits of Hydrological Alteration (ELOHA), rely on generalizable models and data which can be extrapolated to multiple areas and systems (Poff *et al.*, 2009). While extrapolation is always limited by site-specific conditions, the value of data which can be broadly used and applied to understand the basic functions of other systems is evident. However, this study represents a semi-arid geographic environment, and while the results could be applied to many semi-arid river systems, it is limited by and has particular relevance to groundwater-fed streams as they are less rapidly and severely affected by seasonal fluctuation and changes in climate than other semi-arid systems.

There are two key objectives for this study:

- Describe the spatial and temporal variability of habitat conditions and biotic community composition for a groundwater–fed semi-arid river
- Investigate the relationship between system drivers (flow and sediment), habitat quality and biotic response.

A secondary objective is to assess the usefulness of the MiniSASS methodology and scoring system as a measure of habitat quality as well as water quality, in an attempt to streamline the process of habitat assessment for citizen science.

1.2 Study area

The study is based in the eastern central Karoo region of the Eastern Cape, South Africa. A general description of the region has been given by Boardman *et al.* (2013). The Karoo lies between 28°S and 33°S and covers around one third of South Africa's land surface.

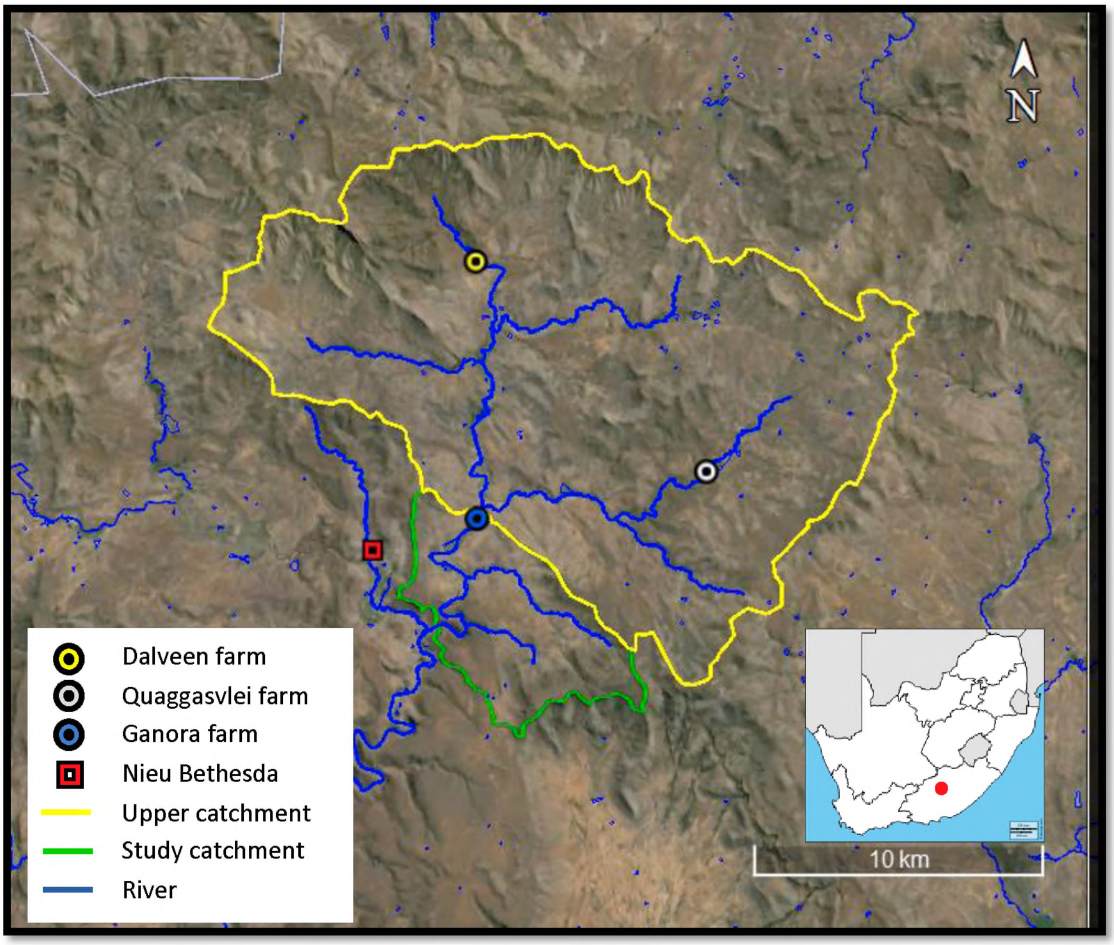

Figure 1: Catchment boundaries and main tributaries of the Wilgerbos River. Also shown: Dalveen farm and Quaggasvlei farm rainfall stations, Ganora farm and the town of Nieu Bethesda.

The landscape is a combination of plains, hills and an east-west oriented mountain range, the Sneeuberg Mountains. The hills and mountains make up an important part of river recharge in the wet season as many of the rivers are fed by overland flow from surrounding slopes (Le Maitre *et al.*, 2007). River systems in the study area are comprised of a number of tributaries draining from the steep slopes of the upper catchment. The Wilgerbos River (the focus of this study) has its headwaters in the Compassberg mountain catchments and feeds into the Sunday's River which flows to the south coast of South Africa.

The Karoo falls within the Warm Temperate Zone and the eastern central region has a distinctly continental climate (Boardman *et al.*, 2003; Grenfell *et al.*, 2012). The average maximum summer temperature, as given by Grenfell *et al.* (2012), is 36°C, and the average minimum temperature in winter is -7°C. Annual rainfall is highly variable, and average rainfall figures vary between 346 mm/a (measured at Graaff Reinet) and 422 mm/a (measured at Gordonville); however, as Figure 2 shows, annual rainfall is also temporally highly variable, ranging from 173 mm in 1966 to 866 mm in 2011.

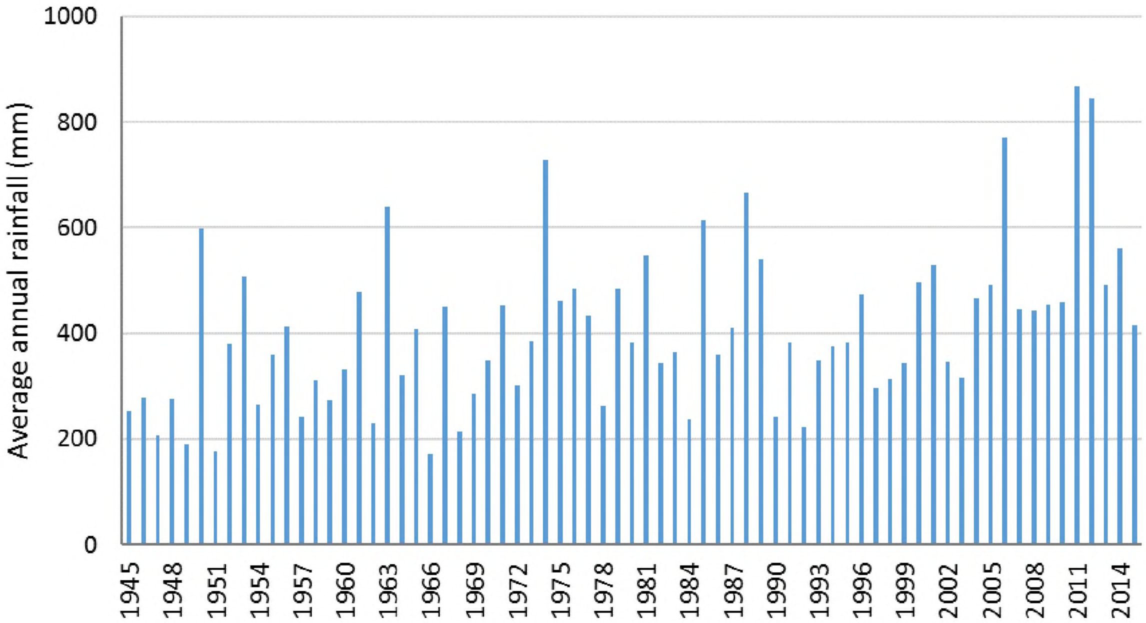

Figure 2: Average annual rainfall for Quaggasvlei farm and Dalveen farm, from 1945 to 2015. Source: Quaggasvlei and Dalveen private farm records.

Grenfell *et al.* (2012) calculate a 37% inter-annual rainfall variance co-efficient, which they claim is among the highest in the world. High seasonal variability of rainfall is also consistently noted in literature, with approximately 30% of rain falling in the summer months and peaking in March (Sami, 1992; Boardman *et al.*, 2003; Grenfell *et al.*, 2012; Boardman *et al.*, 2013). Annual potential evapotranspiration is estimated at 2287 mm/a, creating a water deficit of approximately 1865 mm/a (depending on where rainfall measurements are taken) (Grenfell *et al.*, 2012). Figure 3 shows long term average monthly rainfall recorded at Quaggasvlei (location shown in Figure 1), which illustrates the seasonality of rainfall in the region.

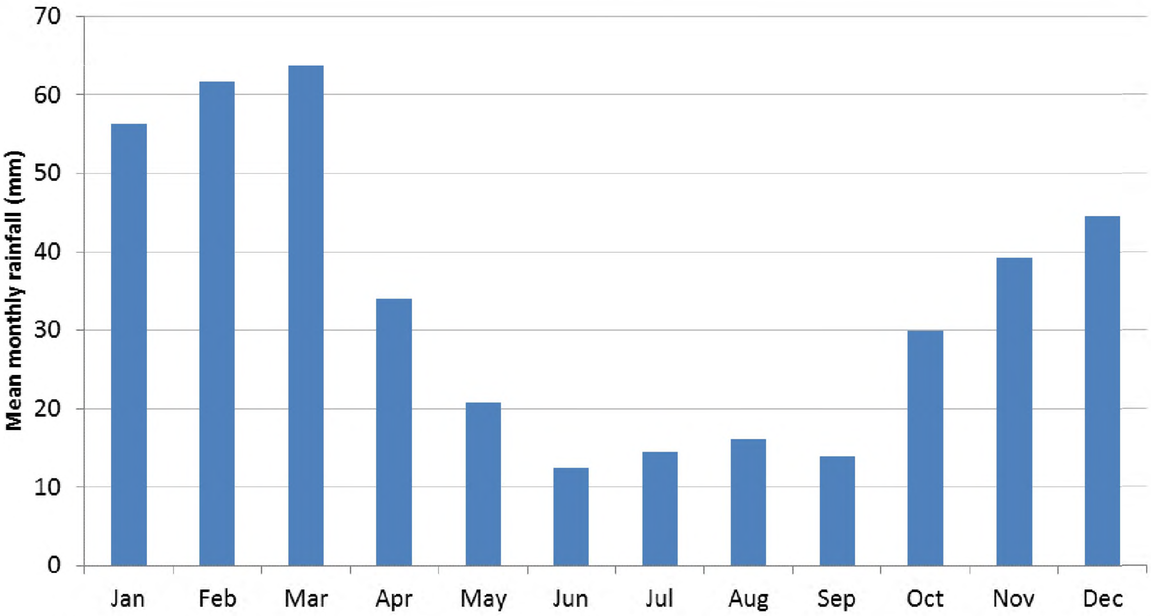

Figure 3: Mean monthly rainfall for Quaggasvlei farm, from 1945 to 2015. Source: Quaggasvlei private farm records.

Table 1 provides the standard deviation and coefficient of variation for this data, to highlight the variability of rainfall from year to year. It is also important to note that rainfall variability for winter and spring months (May to October) is far higher than the variability in summer months.

Table 1: Mean monthly rainfall for Quaggasvlei farm, from 1945 to 2015, with standard deviation and coefficient of variation.

	Mean (mm)	**St Dev (mm)**	**CV (mm)**
January	56	48	85
February	62	52	85
March	64	51	79
April	34	26	76
May	21	23	111
June	12	16	128
July	15	18	123
August	16	26	161
September	14	18	131
October	30	32	106
November	39	32	81
December	44	43	97

Convectional thunderstorms often occur during summer, but the region seldom receives more than 10 mm of rainfall a day, even in peak season (Boardman *et al.*, 2003). Frost is common in winter, occurring approximately 80 days a year, and snow falls in the upper mountain areas when cold fronts pass over the Karoo (Boardman *et al.*, 2003; Grenfell *et al.*, 2012). Long term daily rainfall data for Cranemere Farm, approximately 50 km away from the study site examined in this thesis, was analysed by Foster and Rowntree (2012). The data, reaching back to 1888, showed that March and November were the wettest months, with mean rainfall of 46 mm and 45 mm respectively, while July was the driest month, with mean rainfall of only 11 mm (Foster & Rowntree, 2012).

The Wilgerbos River flows through the foothills of the Sneeuberg Mountain range, which lies within the Beaufort Group of the Karoo Basin and has a geology comprising mainly Mezo-Palaeozoic sandstones and mudstones, with dolerite caps and numerous intrusions (Turner, 1981; Boardman *et al.*, 2003; Schlüter, 2008). Vegetation in the Sneeuberg Mountain region is a mix of the Grassland and Nama Karoo Biomes, comprising Karroid scrub on the plains and sourveld in the highlands (Boardman *et al.*, 2003; Rutherford *et al.*, 2006). The upper slopes are typically used for cattle grazing and the plains are used for small stock, such as sheep and goats (Boardman *et al.*, 2003). Some farmers use valley floors to grow small patches of fodder crops, wheat and potatoes (Boardman *et al.*, 2003). The total area of the upper and lower catchment (as shown in Figure 1) is approximately 306 km^2. Relief ranges from 1300 m to 2000 m, with a relief ratio of 0,04 for the catchment (Foster *et al.*, 2012). The rounded shape of the upper catchment, combined with the sparse vegetation and shallow soils, creates a system with 'flashy' hydrographs due to shorter lag times following rainfall events and relatively high peak discharges. As the area is sparsely vegetated with Karoo scrub, soil infiltration rates are low during intense rainfall, surface run-off is high.

Relatively steep slope profiles, compounded by shallow soil and prevailing scrub-type vegetation, have led to the formation of erosion features, namely dongas and extensive badlands, which are a major source of sediment transport to the river systems (Boardman *et al.*, 2003; Foster *et al.*, 2012; Foster & Rowntree, 2012). Erosion rates in the Karoo badlands are significantly higher than other catchment regions, and evidence from the Ganora Farm dam suggests that changes in connectivity have occurred over time (Foster *et al.*, 2012). The steep gradient of the catchment yields large quantities of sediment. Rowntree and Foster (2012) measured a sediment yield of 654 t/km^2/a, far higher than the 207 t/km^2/a estimated by Msadala *et al.* (2010) from a 3666 km^2 catchment in the nearby town of Graaff Reinet. A number of farm dams within the catchment have become filled with sediment due to the high sediment load transported by the rivers.

The Wilgerbos River itself is a combination of straight and braided channels, with a large variety of water column depths and flow speeds. Erosional and depositional processes vary as flow fluctuates throughout the year, but the presence of gravel islands and deep accumulations of fine sediment in pools suggests that deposition is the more dominant of these two. Some reaches of the stream are fed by rainfall and surface flow, but stretches of the lower reaches are maintained by groundwater reserves, and are therefore less susceptible to droughts and often able to maintain perennial, albeit low flow even in dry seasons. Channel confinement varies along the course of the river, as some reaches flow through a gorge and are therefore confined by impervious dolerite intrusions, while others are unconfined.

The section of the river used for this study runs through two farms, Ganora Farm and De Toren. As with most of the Karoo, the land is used for stock farming. Much of the land has not been cultivated, other than river terraces, and primarily sheep are kept. The river has been modified for extraction at a number of points along its course, including dams, diversions and channelization, which is typical of river systems in water stressed regions. Due to the high seasonal variability of Karoo rainfall it is essential for farmers to make use of dams to ensure water supply throughout the year. The Wilgerbos River is also frequently disturbed by livestock, (predominantly sheep and cows) which use it as a source of water or to cross the channel to reach new grazing lands, and farm vehicles. Despite this, large stretches of the river are unmodified and for the most part undisturbed.

This study uses three sections of the river (shown in Figure 4), of varying degrees of modification and disturbance. In total the length of the reach in approximately 8,8 km. The first site (146 m long) is fed by the larger upper catchment, and could be considered largely modified as it is situated directly below Ganora farm and is an important source of water for farm use. Upstream of the site is a weir for water storage which traps and stores sediment, and a small off-channel dam. This site also separates the sheep from the farm and their grazing lands and is immediately upstream of a crossing point for the livestock, herders and farm vehicles. The second site (214 m long) is situated 5 km downstream of the first, fed by the lower catchment, and is at the start of a gorge. Three km upstream of this site is a third farm dam, a weir and a small diversion channel. No evidence of major disturbance is visible in the site itself. The third and final site (137 m long) is situated a further 3 km downstream of the second, at the upper boundary of De Toren farm. This site is also fed by the lower catchment and is considered the least disturbed, with no activity evident in the site itself. As it is situated at the end of the gorge, little farming activity occurs in the surrounds.

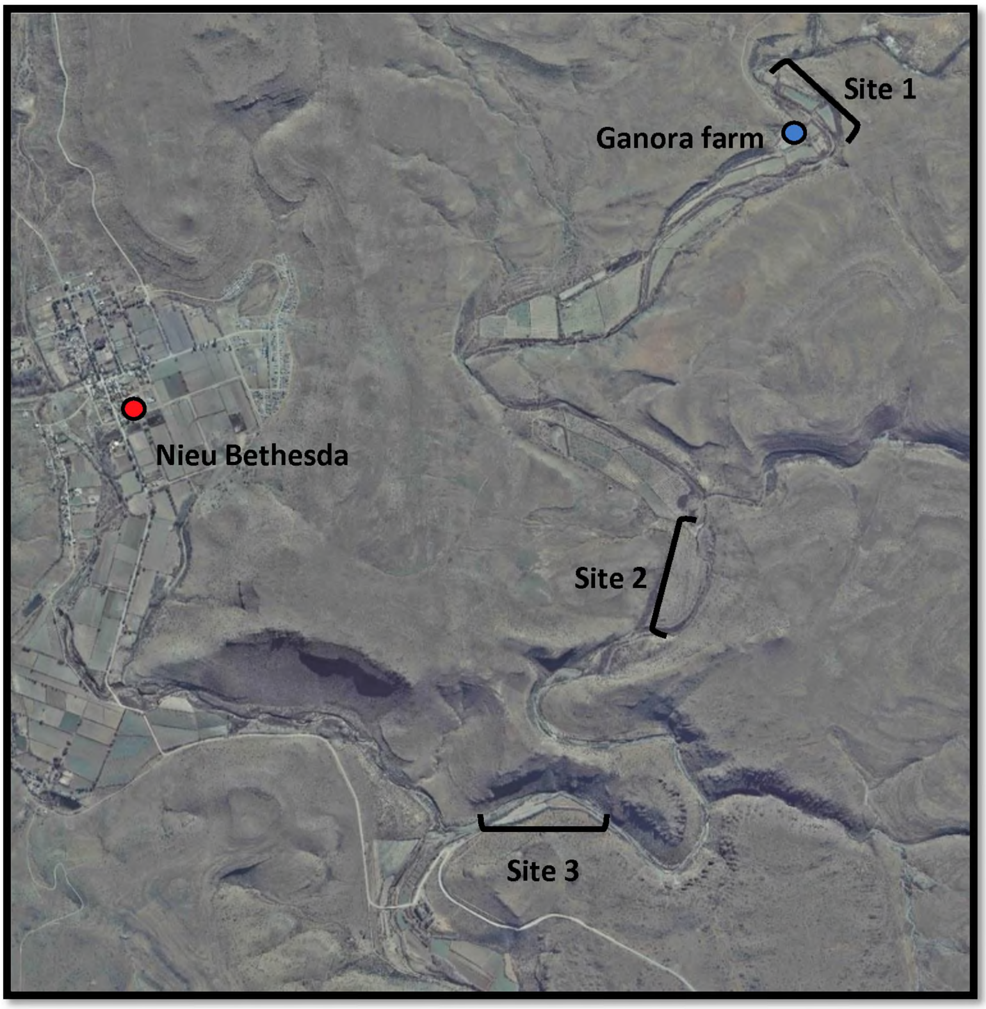

Figure 4: Location of study sites (Site 1, 2 and 3) in relation to Ganora farm and Nieu Bethesda.

The three sites used in this study represent a variety of physical properties. Each site has distinctive characteristics and the physical properties of the river vary noticeably within and between sites (see Photo 1 – 6). Site 1 is the simplest of the three sites in terms of channel type, having the straightest channel and the fewest islands, bars and flow diversions. As a result, this site also has comparatively little rooted instream vegetation. The channel is wide and only semi-constrained, with a row of poplar trees and hardy sedge growing on the left bank (looking downstream), and dense shrubs on the right bank. Site 2 has the greatest variety of habitat types, ranging from shallow riffles, to deep sediment rich pools. The top of the site contains a number of vegetated islands, made up predominantly of sedges and grasses, the right bank is covered with a dense growth of sedges and reeds, while the left bank supports Karoo scrub. The bottom of the site is constrained by a dolerite intrusion and the wall of the gorge, and contains no instream vegetation. Site 3 has far more braiding and flow diversions than either of the others. Again, a number of the islands are vegetated with sedges and grasses. The site is also constrained by the gorge, but less so than Site 2 as there is no dolerite sill. The left bank is bordered by a row of dense bush, and the right bank, a steep slope due to the gorge, has fairly patchy Karoo scrub growth.

All three sites also contain diverse bed and bank material. There are a number of large boulders in Site 1 (Photo 1), many of which were deposited from a weir wall, which was broken in a flood in 2011 (Steynberg, J. P., pers. comm., 2015), but the typical flow in the site would not be strong enough to move large material on a regular basis. As a result, the site is dominated by gravel and small cobbles, with large areas of sand and other fine materials on the banks and bars (shown in Photo 2).

Photo 1: Site 1: large boulders at top of site, transported by flood water.

Photo 2: Site 1: gravel and sand bars at bottom of site.

Site 2 also has the greatest variety of clast arrangement of the three. The top and bottom sections of the site are low flow straight channels with large amounts of fine sediment and gravel in the bed and on the banks. The top section has fast flowing shallow riffles containing large gravels, cobbles and small boulders, with very low amounts of fine sediments. The middle section of the site is dominated by a large, deep pool with low flow, where the bed is made up almost exclusively of fine sediment and sand (Photo 3). Below the pool a second riffle section flows over bed rock, and has a higher proportion of cobbles and boulders, with gravels and fine material occurring in low amounts. Site 3 contains patches of gravels, but is predominantly made up of fast flowing shallow riffles and therefore contains higher proportions of larger clasts and lower amounts of fine material (Photo 5). However, like Site 2, the top and bottom sections of the site are made up of low gradient, low flow straight channels, which have high fine sediment content both in the channel and on the banks and bars (Photo 6). Low flow sections such as these were able to support large amounts of algae at times, and mats of filamentous green algae were found in all three sites at least once during the study.

Photo 3: Site 2: mid-section pool with dolerite sill and gorge on right

Photo 4: Site 2: thick algal growth caught on rocks in flow, July 2014.

Photo 5: Site 3: rocky channel section, confined by gorge on left and right. (Credit: R. Fox)

Photo 6: Site 3: thick algal growth growing in fine sediment, July 2014.

1.3 Rationale

This study provides an examination of changes in physical and chemical habitat properties over one year and the response of instream biota to these changes. It addresses the knowledge gap related to semi-arid stream systems and changes in biotic community composition and creates a specific baseline for future studies in this field. This thesis discusses established and current literature on the topics of river systems in general, semi-arid systems, habitat quality and assessment methods, and drivers and limiters of biotic communities. The knowledge gained through the execution of this study largely affirms current knowledge, but it also puts forward the use of the MiniSASS assessment method as a measure of habitat quality as well as water quality.

The weather conditions prevailing through the study period enabled an assessment of biotic response to and recovery from drought systems and highlights the importance of groundwater streams in a semi-arid environment in terms of their ability to resist drought. It is important that semi-arid systems be studied as separate from perennial systems, as the processes which occur and the way in which the systems respond to them are not comparable (Uys & O'Keeffe, 1997; Hansen, 2001; Buffington & Montgomery, 2013). The importance of effective water management in these water-stressed regions cannot be overlooked. The Karoo is a water scarce area, which relies heavily on groundwater and dams for its water supply, for domestic, commercial and agricultural purposes (Uys & O'Keeffe, 1997). This places pressure on the hydrological system of the area and, if not properly managed, could lead to long-term damage through over-use and extraction (Uys & O'Keeffe, 1997). With the issue of potential hydraulic fracturing in the future, many people are increasingly concerned about impacts on the quality of groundwater systems in the Karoo, which feed into the rivers.

Baseline studies provide a point-of-reference for any future assessments, and can be conducted at a small scale, such as the impact of a farm dam on the river downstream, or at a larger scale, such as a catchment in which a large-scale disturbance has occurred. Natural and anthropogenic disturbance can equally affect instream habitats, and habitat assessments can be as useful for measuring recovery time after a drought or flood as they can be for long term measurement of anthropogenic disturbance such as water extraction. Therefore, greater focus needs to be placed on gaining the same level of knowledge about semi-arid systems as exists about perennial systems, in order to ensure that suitable and specific management can be put in place. It is especially useful in producing management that is aware of the complexity of these systems, rather than implementing generalized models and practices which are not fully equipped or designed to respond adaptively to the highly variable nature of a semi-arid environment.

CHAPTER 2: LITERATURE REVIEW

In order to investigate the effects of physical and chemical variables on habitat quality, it is necessary to establish a basic understanding of what variables to consider and what is known about their effects on habitat and biotic communities. A number of studies have investigated individual elements of the aquatic habitat, such as flow, or substrate, or vegetation, but a synthesis of these investigations is difficult to find, and while terms like “heterogeneity” and “habitat health” are commonly used, there is disagreement over definitions, measurements and assessment techniques. The field of habitat assessment is an increasingly interdisciplinary one and as a result there are still gaps in knowledge and areas of uncertainty, which creates a need for research. This chapter will begin by reviewing studies on river categories and definitions of the word “habitat”, followed by a discussion of key habitat drivers, especially flow and sediment dynamics, for habitat assessment. The features of semi-arid systems will be briefly described, and the impact of physical drivers on streams in these environments. Findings on the effects of physical habitat on biotic communities will then be reviewed, after which knowledge gaps will be identified and potential uses of and future projections for this study in terms of river management are suggested.

There are a number of features used to characterise and classify rivers at a variety of scales (Hauer & Lorang, 2004). These features fall into the broad categories of physical, chemical and biological properties and are largely defined by the catchments from which the rivers originate (Hauer & Lorang, 2004). It is understood that rivers are dynamic ecosystems which are longitudinally, laterally and vertically connected with their surroundings, creating a complex network of systems and feedbacks at all temporal and spatial scales (Hauer & Lorang, 2004; van Wyk *et al.*, 2012). As a result, the study of river ecology has become a multidisciplinary field, incorporating physical sciences with chemistry and biological studies, among others, to achieve a more comprehensive understanding of the way in which river systems function (Hauer & Lorang, 2004; Collins & Owen, 2006; Barbour *et al.*, 2008). In order to study and assess rivers and how they function, it is important that scientists are able to characterise the features of these systems in space and time, which can also provide useful information when assessing river impairment and degradation and evaluating management strategies (Hauer & Lorang, 2004). River systems are often described as being hierarchical in arrangement and a number of different hierarchical models have been suggested (Poff, 1997). Poff (1997) describes rivers in terms of four simple but effective levels of hierarchy, which can be used to determine the structure and functionality of a system. These levels are (from coarsest to finest): watershed, stream reach, channel unit, microhabitat (hereafter referred to as the habitat patch). A similar, three-stage hierarchical system is proposed by Thoms and Sheldon (2002), which simply excludes the watershed as part of the direct hydraulic habitat in which biota survive.

2.1 Instream habitat

The habitat patch is commonly considered the smallest unit making up a river system. Habitat units are multidimensional features, which create a mosaic of diverse components that vary over relatively short distances (Newson & Newson, 2000) and are dynamic in time as disturbances "alter hydrology, sediment transport and distributions of vegetation and other biota on daily, seasonal and inter-annual time scales" (Hauer & Lorang, 2004: 396). There are a number of physical indicators of channel condition which are strongly linked to instream habitat quality, including channel width to depth ratios, channel sinuosity, gradient, bank stability and pool to riffle ratios (Bunn & Arthington, 2002; Apitz, 2012). Habitat availability within a system is dictated by the size of the channel, while the shape of the channel affects the hydraulic properties of the habitats, and habitat quality (the ability of the habitat to support biotic communities) is largely determined by the size of the sediment being deposited by the stream i.e. the relationship between deposition and erosion (van Wyk *et al.*, 2012). Heterogeneity of habitats is an essential aspect of in-stream ecosystem functioning and the natural complexity of a habitat patch is necessary for ecosystem health (Apitz, 2012). Tews *et al.* (2004) describe the "habitat heterogeneity hypothesis" which posits that the more heterogeneous (or structurally complex) a habitat is, the more niches and environmental resources it provides, which in turn supports a greater diversity of biotic life. Results of previous heterogeneity studies have yielded contradictory results, as it can be difficult to define a habitat, especially at a small scale, and what may seem to be habitat heterogeneity by one taxonomic group can also be perceived as habitat fragmentation for other groups (Tews *et al.*, 2004). The size of the habitat patch is also dependent on the size of the organism being studied, and other organisms which are larger or smaller can either be excluded from the patch or be overlooked. However, the hypothesis has been a corner stone of ecology for over 60 years and there is as much evidence to support it as there is to disprove it (see Simpson, 1949; MacArthur & Wilson, 1967; Lack, 1969, cited in Tews *et al.*, 2004).

Habitats are determined by a channel's spatial situation, form or structure, and supporting processes, and in this way the physical structure of the river dictates the structure of habitats, which in turn dictates the structure of the biotic communities (Rigsbee *et al.*, 2013). Wadeson and Rowntree (1998) draw a distinction between a habitat, the abiotic environment of a species, and a biotope, the abiotic environment of a community or species assemblages. The hydraulic biotope is a spatially distinct in-stream environment, comprised of flow related abiotic factors and resulting from interactions among flow, substrate and channel morphology (Wadeson & Rowntree, 1998; Rowntree & Wadeson, 1999). Although biotope may be more accurate for describing the living space of a biotic community, the term habitat is more widely used, so for the purpose of simplicity in this study, 'habitat' will be used as a catch-all for both these concepts.

2.2 The role of flow and sediment dynamics in habitat structure

Newson and Newson (2000) state that, until the late 1990s, work at the habitat scale of a river system was conducted almost exclusively by ecologists in an empirical, top-down manner, retroactively applying theoretical geomorphological principles to explain habitat distribution. Little attention was given to the physical properties of the habitat itself, except in so far as they related to the requirements of the biota. However there has been a growing awareness of the importance of multi-disciplinary work, so as to make use of a wider range of methods and to incorporate more diverse elements, such as geomorphology (Newson & Newson, 2000). This has led to growth of the school of habitat hydraulics, or 'eco-hydraulics', a bottom-up approach to habitat studies which assumes that there is an inherent link between flow processes, the physical environment, and biotic patterns and habitat needs (Newson & Newson, 2000). This school of thought, as explained by Newson and Newson (2000), also states that stream hydraulics determine the assemblage of species along the longitudinal profile of the river system, and that a complete physical description of any site under study is essential. Furthermore, they explain that studies in the UK, New Zealand and South Africa have provided substantial evidence to show that eco-hydraulics are strongly regulated by the morphology and substrate material of the stream. Furthermore, eco-hydraulic patterns vary with stream flow, making it essential that ecosystem health be studied at both a spatial and temporal scale to account for a range of flows (Newson & Newson, 2000).

Bunn and Arthington (2002) summarise the role of flow dynamics using four key concepts, which highlight the link between hydrology and other facets of a river system: 1) flow is a key factor in determining physical habitat structure; 2) riverine biota evolve life histories in response to flow regimes of the system; 3) connectivity in both the longitudinal and lateral dimension affects the ability of biota to move freely through the river system; 4) the establishment of invasive and alien species communities can be facilitated by irregular or unnatural flow regimes. The first three of these four principles inform the key theory of this study. Poff and Zimmerman (2010) expand the discussion of stream flow as a key factor in habitat condition, referring to it as a 'master variable' of ecological characteristics. Flow is the main driver of geomorphological processes and channel morphology, which in turn is key to habitat structure and ecological processes (Arthington & Zaluki 1998; Bunn and Arthington 2002). Similarly, Hughes and Louw (2010) point out that ecological responses occur as a result of habitat variations caused by variations in flow. Extreme flow fluctuations determine the success of stream biota populations and regulate ecosystem processes, and the variation of flows over time determines evolutionary adaptations (Poff & Zimmerman, 2010). Extreme fluctuations in flow are often the result of disturbances, which can be natural, such as floods and droughts, or man-made, such as the construction of dams and canals and diversions of flow (Hauer & Lorang, 2004; Poff & Zimmerman, 2010). Up to a point, disturbances can benefit river systems by encouraging biodiversity and habitat complexity, and maintaining environmental gradients (Hauer & Lorang, 2004). However, disturbance can also reduce habitat complexity, and cause physical and chemical changes in habitat and water quality which hamper the functioning of that habitat (Apitz, 2012).

Sediment is also an essential but currently inadequately understood component of aquatic ecosystems as it creates the foundation for many habitat types, provides a substrate for many organisms, and contributes to the sustainability of a number of important ecosystem services and functions (Apitz, 2012; van Wyk *et al.*, 2012). A number of biota have specific sediment requirements and the extent to which the sediment meets these requirements determines the influence it has on habitat quality, and the role it plays in ecosystem service provision (Apitz, 2012). Fine sediment is fundamentally soil which has been deposited in an aquatic setting, but a more accurate definition would include suspended, as well as deposited solids, which are transported by water (Apitz, 2012). The dynamic movement of water creates a cycle of erosion, suspension and deposition, which moves and stores sediment through the hydrological system over short and long temporal and spatial timescales, resulting in river channels that are continuously adapting (Apitz, 2012). Changes in sediment inputs (either increases or decreases) can result in habitat change or even loss (Berry *et al.*, 2003; Apitz, 2012). However, the effects of sediment on an ecosystem are complex and, as there is no agreed-upon method for measuring sediment loads, the impacts of changing sediment inputs cannot be quantitatively represented in their entirety (Berry *et al.*, 2003). Although limited, it is possible to study the impacts of sediment load through key habitat quality indicators and processes. Increased quantities of suspended and deposited sediments affect habitat quality in four main areas: 1) reduced light penetration to lower levels of the water column, inhibiting photosynthesis, 2) physical damage to biota through abrasion, 3) absorption and transport of toxicants and heavy metals, and 4) change to substrates on the river bed and associated habitats (Griffith & Walton, 1978; Apitz, 2012). Any of these changes can result in a loss of biodiversity, and reduced resilience and ecosystem functioning (Apitz, 2012).

According to Apitz (2012), the status of instream sediment is determined by four attributes – quality, quantity, transport and location – and is controlled by biophysical condition at a range of spatial scales. She highlights that these attributes are interrelated; transport cannot be considered without location and quality, and quantity cannot be considered without transport; however, they must be independently studied so that their relationships can be understood. Furthermore, Apitz (2012) states that sediment quality is the composition of the substrate, in terms of grain size, organic matter, nutrient content, contaminants, pollutants and pathogens, which she discusses in detail. Grain size determines the types of habitats the substrate will create, e.g. large rocks provide refugia for fish, while fine sediment accommodates organisms which prefer to bury themselves. Benthic organisms also have sediment requirements in terms of chemical, mineral and nutrient concentrations, and therefore these are also important determinants of what biota can inhabit the substrate of a river bed. Sediment quantity is the amount of sediment delivered to the river system from the landscape, and is affected by channel morphology and flow dynamics. The consistency of sediment input determines the stability of habitats (too much sediment can cause smothering, while too little can cause a habitat to gradually be washed away by flow). Sediment transport depends on how effectively sediment is moved from the catchment, into and through the river system. Transport is a factor of discharge and the carrying capacity of the stream, and for the most part sediment supply remains in balance with the stream's ability to transport it. Finally, sediment location is the end point at which the sediment finds itself after transportation, which is dependent on the sediment and flow dynamics of the system.

Similarly, Owens *et al.* (1999) discuss fine sediment processes, explaining that during transport large quantities of sediment enter the system from the drainage basin, but are lost along the course of the stream channel, resulting in a far smaller quantity of sediment leaving a channel than what entered it. This is represented visually in Figure 5. In many cases, the 'lost' sediment will be stored within the channel or floodplains, and it is therefore also important to consider the residence times of sediment in these stores as this determines the lifespan of habitats created by deposited fine sediment. They add that this also has implications from a management perspective in order to design and implement effective sediment control strategies, and adaptive strategies which are able to cope with the natural fluctuations in sediment loads over time.

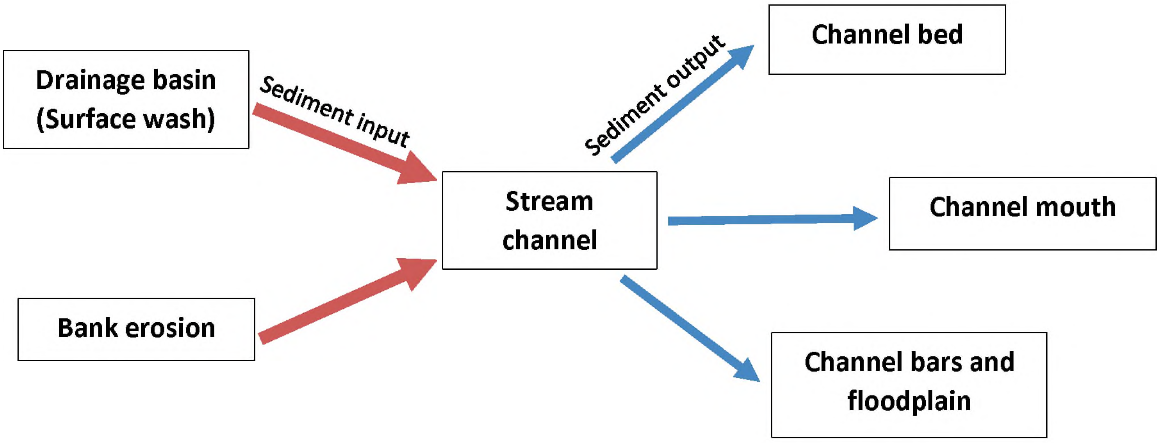

Figure 5: Movement of sediment through a stream system, adapted from Owens et al. (1999).

2.3 Factors for habitat assessment

Though it is largely agreed upon in literature that flow and flow disturbance, and substrate are the main factors influencing riverine habitat, ecosystem functioning and community composition are controlled by a number of different sub-factors, operating at different scales, including instream and out-of-stream variables (Mellado Diaz *et al.*, 2008). The variety of variables listed in literature is large and can include catchment scale factors such as land use/land cover, underlying geology, catchment area and altitude, and instream factors such as substrate particle size, conductivity, nitrate content, water temperature, disturbance frequency, timing and duration of disturbance, and river discharge, among many others, which act at different spatial and temporal scales (Pennak, 1971; Puckridge *et al.*, 1998; Mellado Diaz *et al.*, 2008). Standard foci of habitat classification are "flow, temperature, vegetation, substrate, width, depth, water hardness, and bottom fauna" (Pennak, 1971: 321). However, Pennak (1971: 321) calls classification systems for lotic system habitats "varied, uncertain and unreliable", because not enough variables are taken into account during assessments. Puckridge *et al.*

(1998) suggest that between 8 and 11 variables should be considered when conducting habitat assessments, so as to ensure some degree of understanding of the system. Mellado Diaz *et al.* (2008) used a total of 39 variables in a habitat assessment, which included 12 physicochemical variables, 7 geomorphic variables, 5 land use/land cover variables, and 8 in-stream variable metrics, among others. In stark contrast, Jowett (2003) states that in order to assess physical habitat and its influence on benthic organisms, the primary factors are simply water depth, flow velocity, and substrate size and stability. Pennak (1971) states that during his research 13 variables were identified as crucial for an accurate habitat assessment. He emphasises that physical and chemical variables can be objectively measured, are universally common to all streams, are indicative of the biological features one could expect to find in a habitat patch, and are therefore the most useful criteria for habitat assessment (Pennak, 1971). A more recent study by Rivers-Moore *et al.* (2008) strongly emphasises the importance of measuring water temperature in conjunction with flow rates when assessing habitat stability and biotic community structure.

The list of variables discussed by Pennak (1971) includes a number which are relevant to this study and therefore this will become an important reference. While Jowett's (2003) list of four variables may be more contemporary and simpler to assess, given the lack of knowledge on the study area, the author felt that Pennak's (1971) criteria would provide a more comprehensive dataset for future researchers to build on. Although the list is designed for habitat classification, it also provides useful criteria for a habitat inventory as it takes into account seasonal variability and allows for potential physical changes to the channel over time. Elements from this list were used as selection criteria for sites for this study. Diversity between sites in factors such as type of substrate and flow velocity was important for representative sampling. Eight factors were later used as a basis for the structure of the sampling technique used. The variables are listed below, with (*) used to indicate those used in this study:

1. Width of the stream channel*
2. Nature of flow (temporary or permanent) *
3. Flow velocity*
4. Type of substrate*
5. Summer water temperatures*
6. Winter water temperatures*
7. Turbidity*
8. Total dissolved inorganic matter

9. Total dissolved organic matter
10. Water hardness
11. Dissolved oxygen*
12. Rooted aquatics
13. Streamside vegetation

However, this list is not fully comprehensive and a number of important variables, such as depth, have not been included. As a result, the list needs to be augmented with variables from the far longer list described by Mellado Diaz *et al.* (2008), to include pH and depth, and uses EC as a measure for total dissolved inorganic matter.

2.4 Habitat assessments in semi-arid environments

Semi-arid regions are particularly susceptible to changes in flow and sediment dynamics, and the effects of such changes are often easily observed. Oliva-Paterna *et al.* (2003) define 'semi-arid' as regions which have a negative water balance, leading to non-permanent and often unpredictable periods of environmental stress. Further, they are temporary lotic systems which frequently experience dry periods and have a high hydrological variability (Uys & O'Keeffe, 1997; Meyer *et al.*, 2003). Semi-arid regions receive strongly seasonal rainfall and are naturally drought prone, but with a propensity for flash floods during heavy rains (Goodrich *et al.*, 2000; Foster *et al.*, 2012; van Wyk *et al.*, 2012; Boardman *et al.*, 2013). Streams in such areas are therefore typically non-perennial or ephemeral and subject to an irregular hydrological regime where natural disturbances can occur any number of times within a year (Uys, 1997; Oliva-Paterna *et al.*, 2003; Mellado Diaz *et al.*, 2008). Much of the literature reviewed in this chapter is derived from studies of perennial rivers, which, while they are well understood, are uncommon in semi-arid regions, and the knowledge gained from studies of these systems is of limited use when studying non-perennial or ephemeral streams (Shaw and Cooper, 2008). A similar challenge is discussed by Uys (1997) and Uys and O'Keeffe (1997) for South African semi-arid rivers; however, there is still a conspicuous lack of historical and contemporary data on these systems, particularly in South Africa. The magnitude, extent and duration of floods and droughts in semi-arid river systems is extremely variable between basins, streams and even stream sections, as well as on a temporal scale (Meyer *et al.*, 2003). Such variability of streamflow is greatest in ephemeral basins, which can remain dry for over 10 months of a year (Meyer *et al.*, 2003; Shaw & Cooper, 2008). Meyer *et al.* (2003: 271), therefore, emphasise that defining drought hydrologically in semi-arid regions is challenging, because it is essential to consider spatial and temporal specificity, as well as to distinguish between regular, seasonal drought and "supraseasonal drought", which is less predictable and spans a number of seasons. Episodic streamflow is also subject to high transmission losses downstream, a major component of groundwater recharge, and irregular tributary inputs (Meyer *et al.*, 2003; Shaw & Cooper, 2008). Consequently, the existing definitions of streams as perennial, intermittent or ephemeral are inadequate for hydro-ecological assessments in semi-arid environments, as there is a limited understanding of the ecological processes which take place (Uys & O'Keeffe, 1997; Goodrich *et al.*, 2000; Meyer *et al.*, 2003).

The Karoo region of South Africa is typical of a semi-arid landscape which is largely dependent on rainfall for river recharge. Le Maitre *et al.* (2007) conducted a study on landscape-scale hydrology and ecosystem services of the Little Karoo, which lies to the south-west of the study area. Their research showed that while a large proportion of rainwater reaches rivers by surface runoff, some water infiltrates the soil, recharging ground water which can continue to feed the rivers through subsurface pathways over a longer period of time. They add that groundwater can help to smooth the fluctuations in flow throughout the year, creating less extreme flow variability in some river systems. It is clear that lateral connectivity is as vital as longitudinal connectivity to structure and functioning of ecosystems in semi-arid streams (Le Maitre *et al.*, 2007).

Some river systems in semi-arid regions are maintained by groundwater as the primary source of base flow in the channel, allowing them to maintain flow all year round (Hynes, 1983; van Wyk *et al.*, 2012). The continued flow measured in the Wilgerbos River in the Karoo throughout the dry season supports the statement by the owners of Ganora farm that it is an example of a groundwater fed system (Steynberg, J.P, Pers. Comm. 2015). Due to the scarce and unreliable nature of surface water sources, biota in semi-arid stream systems are heavily dependent on groundwater for habitat stability (Sami, 1992; Scanlon *et al.*, 2006). Climate change is expected to decrease run-off by 10 – 30% in dry regions by 2050, which will make groundwater an increasingly essential source for dryland systems (Kingsford, 2011). In conjunction with high variability, semi-arid regions generally experience high evapotranspiration which leads to an almost perennial state of soil moisture deficit (van Wyk *et al.*, 2012). Groundwater can be recharged either by precipitation or melt water, which is quite common during Karoo winters, and can be stored for very long periods of time, or until it is discharged into a stream or other body of water (Hynes, 1983). Groundwater stored at a shallow depth (presumed to be 'newer' water) will enter a stream nearer its bank, while deeper groundwater (presumed to be older) will enter towards the middle of the channel (Hynes, 1983). Groundwater provides a relatively constant supply of water to the river channels, but is also sensitive to precipitation and infiltration rates, and is susceptible to depletion by drought and transpiration by vegetation (Hynes, 1983; Kirchner & van Tonder, 1990; Lamontagne *et al.*, 2005; Maréchal *et al.*, 2006).

A number of early studies reviewed by Hynes (1983) have recognised that groundwater inputs are not only important for stream flow recharge, but can also influence the oxygen concentrations, temperature and pH of the water (Hynes, 1983). While oxygen has been found in groundwater at most depths, the amount of dissolved oxygen stored in groundwater decreases with depth, and so only shallow stores can be considered a notable source of oxygen (Hynes, 1983). Similarly, Hynes (1983) reported a decrease in pH with depth, so groundwater from deeper sources may increase the acidity of the stream flow, while water temperature can be moderated year-round, as groundwater is cooler in summer and warmer in winter than the water column itself (Rivers-Moore *et al.*, 2008). Sophocleous (2002) also discusses a moderating effect on temperature by groundwater, based on a thermal displacement system between the surface water and interstitial groundwater. He also implies that groundwater inputs near channel banks may carry more chemicals and dissolved organic matter from the land surface into the channel, as the water is less filtered due to its shorter percolation process. However, deep groundwater generally has a higher dissolved particle load as a result of longer residence times.

Studies have also found that groundwater is important for maintaining biotic diversity. Hynes (1983) describes cases of instream fauna inhabiting the hyporheal zone, where groundwater and stream flow meet, which is suggestive of downward mixing. This evidence indicates that biota are able to move freely between the water column and the water table via the hyporheal zone, and that hospitable conditions exist within the water table. Reviews by Brunke and Gonser (1997) and Sophocleous (2002) provide evidence to support this, explaining that groundwater ecosystems are more stable than river systems, have laminar flow, and a relatively constant sediment structure. Groundwater systems are often less affected by floods than the surface water, while subsurface flow paths continually transfer water from the surface into deeper layers, thus delivering resources from the surface to support a high diversity of subterranean life (Gilbert *et al.*, 1994; Sophocleous, 2002). Brunke and Gonser (1997) also comment on hyporheobiont species, which live exclusively in the hyporheic zone and hyporheophile species, which begin life in the hyporheic zone and advance to the benthic zone as they develop. These species further suggest that the hyporheic zone provides a habitable environment, and may account for the presence of biota described by Hynes (1983). Groundwater has also been found to contain high volumes of dissolved organic carbon, far higher than those of the stream channel (Hynes, 1983). This is most likely due to filtering by the stream bed as the groundwater mixes with stream flow, as well as metabolism by biota in the substrate (Hynes, 1983). It can therefore be assumed that groundwater also serves a role as a food source for some biota, though the extent to which it fulfils this role is not mentioned in literature.

2.5 Instream biota as habitat indicators

Riverine biota are directly controlled by the hydrologic and geomorphic regimes of the river system, and spatial variations in biotic arrangements and ecological complexity are the product of water and habitat availability, and disturbances, such as drought and flood cycles (Goodrich *et al.*, 2000; Shaw & Cooper, 2008). Biota adapt to specific niches in the ecosystem, which are predominantly determined by patterning of physical conditions, with the creation of refugia playing a particularly notable role in biotic community establishment (Newson & Newson, 2000). In the case of non-perennial river systems, Uys (1997: 6) states that the "running water phase adds to the diversity of niches available" to biota. The sensitivity of biota to their habitat conditions makes them valuable indicators of water quality, but their ability to indicate physical habitat conditions has not been as comprehensively explored.

Primary producers, such as algae, the base of the food pyramid, can provide a useful indication of sediment loads, as they are affected by the amount of sediment in the river system. Although their productivity is controlled by many factors, including light, nutrients, oxygen levels, temperature, flow dynamics and predation, all of these can be reduced by increased sediment loads, leading to reduced primary production, which in turn affects all other levels of the trophic system (Griffith & Walton, 1978). The growth of benthic algae can therefore be an indicator of in-stream conditions. Algal growth depends on a wide variety of biotic and abiotic factors including hydrology, substrate, water temperature, herbivory and organic matter (OM) (Artmann *et al.*, 2003). This creates a large number of limiting factors and therefore algal growth depends on a specific combination of variables (Artmann *et al.*, 2003). Algae produces oxygen as a waste product of photosynthesis when exposed to light (Pinder & Friet, 1994) but also require oxygen for respiration. Studies into oxygen requirements for amphibians have suggested that at 25 °C, oxygen consumption by algae is higher than its output, suggesting that in warm water the presence of algae may cause anoxia, but in lower water temperatures algae can create hyperoxic conditions (Pinder & Friet, 1994). Pinder and Friet (1994) used 10 °C to produce hyperoxic conditions in their study, but Choo *et al.* (2004) used 16 °C, also noting that their control temperature was 20 °C to replicate the natural conditions in which the algae had been growing and producing high oxygen content. However, different species of algae have different temperature and light exposure preferences (Choo *et al.*, 2004). As algae can provide a habitat, breeding grounds and resources for a number of biota, factors which limit algal growth will also limit biotic communities in general.

Aquatic organisms respond to the overall condition of their environment, and thus provide a better indication of long term water quality than other chemical and physical factors (Griffith & Walton, 1978). For example, Mellado-Diaz *et al.* (2008) found that macro-invertebrates in semi-arid environments have evolved to have smaller body sizes than their humid environment counterparts, in order to cope better with smaller pools, lower flow and inconsistent supply of food. Uys (1997) emphasises that the harshness of non-perennial river systems does not detract from biotic diversity and that most of the biotic groups found in perennial systems have also been found in temporary waters.

Aquatic macroinvertebrates are recognised in South Africa as ideal biomonitoring candidates for a number of reasons: they are abundantly present across South Africa, they tend to be sedentary, different taxa have different sensitivities to water quality variables, they are easy to collect and fairly easy to identify, and they provide continuous data of past, present and episodic water pollution (Graham *et al.*, 2004). The South African Scoring System (SASS) is a rigorous biomonitoring method which relies on the identification of up to 90 aquatic macroinvertebrate families, and was developed to provide a 'low technology', reliable and inexpensive method of water quality assessment (Graham *et al.*, 2004). Graham *et al.* (2004) acknowledge the usefulness of the SASS method in terms of assigning quality scores to macroinvertebrate taxa based on their sensitivity to pollution, but while this method is simple for trained professionals, the required background knowledge made it impractical for lay users. As a result, the MiniSASS methodology was derived to be used as both a monitoring tool and an educational resource for communities and schools, using a simplified subset of 13 more easily identified macroinvertebrate taxonomic groups (with 3 subgroups for groups with distinctive stages of development), producing a total of 16 identifiable groups of biota. While the results of MiniSASS vary slightly from those of a full SASS assessment, Graham *et al.* (2004) contend that it is generally not a significant variation. The results have proven to be very valuable as the simplified system has broadened the scope for water quality assessment and much of the river health data used by the government and NGOs alike can be sourced through citizen science.

2.6 Habitat drivers of biota

MiniSASS is typically used to assess water quality, but may have the potential to also assess habitat health and the impact of changes in habitat variables on biotic diversity. Seasonal water fluctuation has been found to have a significant impact on the growth patterns of in-stream biota, especially for fish species (Bunn & Arthington, 2002; Oliva-Paterna *et al.*, 2003; Mellado Diaz *et al.*, 2008). Flow regimes vary in time and space according to climate, particularly temperature and precipitation (Poff & Zimmerman, 2010). Water management strategies typically assume that rainfall fluctuates predictably throughout the year based on historical records, but this is increasingly proving inadequate due to the impacts of climate change (Kingsford, 2011). Semi-arid regions are already subject to extreme variability of seasonal climatic conditions, which are likely to become more variable still in the future (Oliva-Paterna *et al.*, 2003; Kingsford, 2011). This highly variable climate produces even more variable water fluctuation which creates a diverse arrangement of habitat patches and, subsequently, diverse assortment and distribution of biota (Oliva-Paterna *et al.*, 2003). Oliva-Paterna *et al.* (2003) state the importance of small pool refugia during drought periods for allowing some fish species to survive year-round in certain parts of a river channel. This can cause large temporal and spatial variations in species distribution, in response to the variations in the physical and chemical characteristics of the rivers themselves. These refugia, although spatially small, are also important for ensuring long-term aquatic biodiversity as they provide organisms with a starting point for recolonisation after a period of drought (Meyer *et al.*, 2003). The hydrological gradient created by high transmission losses downstream (as described above) also create longitudinal variability in species richness (Pennak, 1971; Meyer *et al.*, 2003).

Habitat factors act as filters for biological traits of instream biota. By precluding species which are not suited to cope with environmental conditions, they essentially dictate the distribution patterns of these traits through the spatial variability of habitats within a stream network (Poff, 1997; Mellado Diaz *et al.*, 2008). For example, in arid and semi-arid environments the temporary nature of the majority of streams means they are characterised by a chemistry which is highly biologically restrictive (Pennak, 1971). Abiotic factors connected to habitat heterogeneity and disturbances are, universally, major driving factors of community composition, especially for invertebrates (Poff, 1997; Mellado Diaz *et al.*, 2008). The properties of a habitat impose physical restrictions that constrain the organisms living within it, often limiting the range of ecological and evolutionary strategies they can adopt (Ribera & Vogler, 2000).

Macroinvertebrates are highly sensitive to the impacts of both suspended and deposited sediments (Griffith & Walton, 1978). The greatest impacts of increased sediment load on macroinvertebrates are loss of food supply and shelter, caused by infilling of spaces between larger substrate materials by fine sediment, as well as physical damage through abrasion and clogging of gills and feeding apparatus (Griffith & Walton, 1978; Berry *et al.*, 2003). The knock-on effect of the damaging effect of sediment on macroinvertebrates is that many of them are important sources of food for larger biota, such as fish (Griffith & Walton, 1978). Heavily disturbed habitats often experience a reduction in density and diversity of macroinvertebrates, particularly with more sensitive species, which are often entirely replaced by those that are more tolerant (Griffith & Walton, 1978). While the effects of local-scale environmental factors on community composition have long been recognised, more recent studies have shown that biotic communities are products of multiple interacting and overlapping factors, at multiple scales (Poff, 1997).

Southwood (1977), cited in Mellado Diaz *et al.* (2008: 2), stated that the spatio-temporal variations in habitat provide a “templet” which allows species traits to be correlated to survival and reproduction techniques employed by the species. This concept has been well explored in literature since. It is similar to Poff’s (1997: 392) concept of habitat “filters” that influence the probability that an organism with specific characteristics or traits will be able to survive in a specific habitat. In other words, the physical and life history traits of a species allow them to survive in certain habitats and they must employ certain survival methods to ensure the continuation of the community in these habitats. Based on this idea, Mellado Diaz *et al.* (2008) take temporal variability of a habitat to be the frequency of disturbances, and spatial variability refers to the prevalence of refugia which act as buffers against disturbance. Southwood’s (1977) (cited in Mellado Diaz *et al.*, 2008: 2) templet also comprised a third axis, the “adversity axis”, which indicated the harshness or richness of a habitat in which a species was surviving.

Lake (1982, cited in Rivers-Moore *et al.*, 2008) hypothesized that highly variable in-stream conditions will favour biota with flexible life histories, and are likely to have less clearly-defined community structures. Vanote and Sweeney (1980, cited in Rivers-Moore *et al.*, 2008: 4) add that "variable seasonal river temperature patterns are the critical factor in maintaining temporal segregation in aquatic invertebrate communities, thus allowing for resource partitioning, and preventing competitive exclusions." Species which are able to survive in very harsh conditions will experience less competition for resources, while rich habitats which support many species will display very high levels of interspecies competition (Mellado Diaz *et al.*, 2008). This concept is supported by findings from Meyer *et al.* (2003) and Poff (1997) who discovered that areas with high variability (harsh habitats) had low species richness, but high stability in community structure and an even distribution of species. However, this approach requires that habitat features be identified and quantified by their capacity to induce mortality, and then to identify the adaptive features which allow some organisms to inhabit patches which others cannot (Poff, 1997). It can also be difficult to relate these categories, as adaptive features in an organism can occur in response to multiple habitat features and mechanistic interpretations cannot always be made (Poff, 1997). This approach has proved to be challenging and somewhat controversial, but is also very valuable as it can provide a strong biological tool for facilitating studies of complex assemblages of species in a system (Poff, 1997). Furthermore, the use of multiple traits and factors improves scientific understanding of how biotic abundance and distribution can vary in response to changes across the system, from habitat patch to catchment scale (Poff, 1997).

Apitz (2012) similarly argues that a comprehensive measurement of ecosystem health should include three key components: 1) vigour, measured by the productivity of the ecosystem, 2) organisation, measured by diversity of species and complexity of biotic interactions, and 3) resilience, measured by the ability of the ecosystem to maintain its structure and function under stressful conditions. However, she adds that the term "ecosystem health", while commonly used in scientific and non-scientific community, is a difficult term to define quantitatively and that current definitions are broad and include: integrity of biodiversity and the ecosystem, lack of stress on the habitat, stability over time and resilience or resistance to change. Other schools have proposed that the term 'ecosystem health' should not be used at all as it is loaded and implies a value system for levels of ecosystem functioning, and often leads to the development of inadequate tools and poor decision making (Apitz, 2012). Alternate terms, such as 'environmental sustainability' or 'habitat quality', as will be used in this study, have been suggested by Apitz (2012). The Department of Water Affairs and Forestry (DWAF) (1996) states that water quality variables can be used to create water quality objectives for rivers, but note the limitations of doing so. Knowledge of water quality criteria allows predictions to be made about likely outcomes of changes in water quality variables; however, there are a number of inherent uncertainties and often generalisations are made based on a select few organisms' response to such changes (DWAF, 1996). They state that the direct impact of water quality on biota is the primary focus of water quality criteria and objective setting, while secondary effects, such as changes in ecosystem structure, can arise as a result of toxic effects, or as a result of changes in the behaviour of aquatic organisms, making these effects more difficult to study. The inherent limitations of water quality objectives stem from the focus on only the water column, often not considering other parts of the aquatic ecosystem, and the focus on single-species response data, rather than multi-species or community response data (DWAF, 1996). Therefore, DWAF (1996) emphasises that water quality criteria often need to be modified to accommodate site-specific conditions, such as catchment vegetation, land use, geology and rainfall patterns, which requires good knowledge of the site in which the work is being conducted.

2.7 The value of habitat assessments

This study provides a baseline for management and monitoring of the Wilgerbos River and can be taken further as an input into an environmental flow assessment (EFA) for this and similar rivers. Environmental flows can be simply defined as "water that is left in a river system… for the specific purpose of maintain the condition of that ecosystem" (King *et al.*, 2003: 620). Allocating water for the environment is necessary to "sustain and … restore ecological processes and biodiversity of water dependent systems" (Arthington & Zaluki, 1998: 1), as well as to maintain the provision of ecosystem goods and services (Kingsford, 2011; Arthington *et al.*, 2006). The South African National Water Act of 1998 stipulates a certain volume and quality of water available in a system must be dedicated to maintaining environmental health at a predetermined level, this is known at the ecological Reserve (NWA, 1998). In order to determine the Reserve, EFAs must be conducted to establish the flow characteristics for all parts of the system from source to mouth (Brown *et al.*, 2010). Water requirements from within the basin are then accounted for and subtracted from the total calculated flow, and the result is used to predict various future scenarios for the system until an acceptable future condition is agreed upon (Brown *et al.*, 2010). The ecological Reserve is then set based on this acceptable future condition (Brown *et al.* 2010). According to Poff and

Zimmerman (2010) and Bunn and Arthington (2002), alteration of the natural flow regime poses one of the greatest threats to ecological stability of rivers, with different types of alterations producing different ecological responses

Stressors such as climate change, acidification, eutrophication, pollution, introduction of invasive species and habitat loss can cause a shift in the baseline functioning of an ecosystem and the structure of biotic communities, and in many cases these habitats cannot be fully restored after the fact (Apitz, 2012). In these cases, the complexity of habitats also becomes a hindrance, as it makes understanding and managing them effectively difficult, if not impossible, and often certain properties will be selected for restoration because they are easier to measure or are more tangibly important, rather than those which provide the most valuable services to system functioning (Apitz, 2012). The effects of climate change are expected to not only alter rainfall trends, but also to change hydrologic regimes, reduce flow and increase water temperature, all of which could be fatal for many riverine biota, making maintenance of environmental flow essential for the survival of these ecosystems (Kingsford, 2011). Studies of habitat properties and indices of habitat quality factors can facilitate more valuable management decisions, and may allow for improved restoration efforts at a later stage (Apitz, 2012).

Rivers and their associated wetland systems across the world are degrading more rapidly than most other ecosystems (Kingsford, 2011). Ecosystem management strategies often are in competition with resource development strategies, which currently dominate decision making processes (Kingsford, 2011). However, as human populations and their demand for water continue to grow, concern about the impact of human activity on water resources, such as dams and flow alteration, has been growing in the scientific community for some time, as modified river basins are expected to be more vulnerable to climate change (Arthington *et al.*, 2006; King & Brown, 2010; Kingsford, 2011). In response to this a number of EFA methodologies have been developed (Arthington *et al.*, 2006). Because water resources are under such great demand for human development, a limited amount of water is left for the environment, making it more feasible for EFAs to focus on key ecosystems and processes rather than a whole river system (Kingsford, 2011). This 'protected area' approach has been commonly accepted for many years, but it is becoming apparent that this does not provide enough control over the flow regime, and that resource management needs to take place at a catchment scale (Kingsford, 2011). Therefore, a priority of resource management should be to establish protected area networks, which incorporate refugia, key habitats and corridors as a comprehensive representation for conservation of the catchment as a whole (Kingsford, 2011). While essential, there are many challenges to assessing the environmental requirements of a system and to develop a conceptual model which is suitable for all river systems (Arthington & Zaluki, 1998). The cost involved in developing site-specific habitat quality assessment criteria would most likely make it unachievable, and so methods which are transferable between systems are therefore prioritised (Jowett, 2003). A number of techniques can be used for a flow assessment, ranging from simply using hydrological records to spatio-temporal modelling of geomorphological and ecological responses to fluctuations in discharge (Arthington & Zaluki, 1998). There are also a variety of approaches based on the primary focus of the assessment, such as the hydrological approach which equates environmental flow to desired ecological condition, the hydraulic approach which is more concerned with physical condition, or the habitat rating approach which quantitatively assesses changes in habitat condition in response to simulated changes in flow dynamics (King *et al.*, 2003; Hughes & Louw, 2010).

There are four basic steps to conducting an effective ecologically based EFA: 1) determine the physical characteristics of the channel which dictate habitat properties and availability, 2) identify the ecological processes of each habitat, 3) identify hydrological drivers and potential impacts of hydrological change on the physical habitat, and 4) decide on key management objectives and needs (Thoms & Sheldon, 2002). As this methodology shows, an EFA cannot be conducted in isolation from other stream properties, and an ecological approach requires knowledge of geomorphology and hydrology. An accurate EFA needs to mimic the various components of natural flow and account for variability of “magnitude, frequency, timing, duration, rate of change and predictability of flow events”, as well as the relationship between flow and ecology, and must account for all biotic and abiotic components which constitute the ecosystem (King *et al.*, 2003; Arthington *et al.*, 2006: 1311; Hughes & Louw, 2010). Therefore, a combination of methods is considered to be a “holistic” approach, and provides a broader and more comprehensive set of results, which can account for more variables and be used for more specific and valuable recommendations for water use and allocation (Arthington & Zaluki, 1998; King *et al.*, 2003; Hughes & Louw, 2010). Holistic approaches are often by definition multi-disciplinary and allow for the integration of data and knowledge from a group of specialists (King *et al.*, 2003). Arthington *et al.* (2006) emphasise the need for site-specific benchmarks in the absence of comprehensive scientific understanding of river systems as a whole, as this allows for monitoring river health over time, comparing each system with itself, rather than with broad and vague rules of thumb.

Management of river ecology is often considered too complicated, expensive and time consuming and is therefore often overlooked (Poff *et al.*, 2011). However, effective resource management requires decisions to be guided by scientific knowledge, making it crucial that all shareholders and interested parties have access to correct, comprehensive and accessible information (Poff & Zimmerman, 2010; Barbour *et al.*, 2008). In many developing countries, lack of data and understanding are major hindrances to management programmes (King *et al.*, 2003). Scientists therefore have an obligation to communicate their findings to anyone who has the potential to use, protect or affect water resources (Barbour *et al.*, 2008). Based on this assumption, Poff *et al.* (2011: 1) put forward the Ecological Limits of Hydrologic Alteration (ELOHA) framework, as a "flexible, scientifically defensible" method for broad-based environmental flow requirements, which is both time and cost effective. The authors propose that the wealth of information on individual rivers that has accumulated over time could be synthesised into a database of system types, properties and variables, thus reducing the need for site specific information for every EFA in the future (Poff *et al.*, 2011). This method would require regional databases to be built from existing literature and frequently updated streamflow data (i.e. hydrographs), classification of river segments by flow regime, records of past hydrological modification and flow alterations, and calculations of the ecological impacts of these alterations (Poff *et al.*, 2011). From this data future EFAs could quickly and effectively be conducted based on data from similar sites, and ecological responses could be predicted based on past experience in other sites (Poff *et al.*, 2011). Somewhat in contrast, Hughes and Louw (2010) raise the issue of implicit assumption and question whether any method can be applied with certainty in a system where the relationship between flow and biological response is not fully understood. They discuss the usefulness of holistic approaches such as the Flow Stressor-Response model and Downstream Response to Imposed Flow Transformations (DRIFT), stating that while both methods incorporate hydrology, hydraulics, water quality and ecology, and are theoretically considered to produce high confidence results, this may not always be true in practise (Hughes and Louw, 2010). However, there will always be sites which have not been previously studied, and for which a site-specific study would be too difficult, expensive and/or time consuming (Hughes & Louw, 2010). Therefore, there is a need for methods such as ELOHA, Flow Stressor-Response and DRIFT, but in order for them to be successful, a broad and comprehensive database must first be established for a variety of rivers in a variety of climates and physical environments, so that reliable conclusions can be drawn for future flow assessments.

As previously established, data on semi-arid river systems in regions such as the Karoo is currently far from comprehensive. This study does not attempt to conduct an EFA, but can be used to inform EFAs by providing information on the relationships among flow, sediment and habitat quality, as a lack of this information is a hindrance to all EFAs (Rowntree, K., pers. comm., 2015)

2.8 Knowledge gaps

As mentioned previously, there are a number of knowledge gaps in the field of habitat quality assessment. Much of the existing literature on the subject focuses on either particular aspects or broad theories. For example, authors such as Griffith and Walton (1978), Berry *et al.* (2003), Apitz (2012) and Foster and Rowntree (2012) among others provide insightful discussions on sediment dynamics, while Bunn and Arthington (2002), King *et al.* (2003), Arthington *et al.* (2006) and Poff and Zimmerman (2010) discuss flow assessment and the impact of flow on river ecosystems. Discussions on the impact of habitat on specific biota can also be found in studies such as those by Ribera and Vogler (2000), who investigate species range size in aquatic *Coleoptera*, Oliva-Paterna *et al.* (2003), who discuss *Barbus sclateri* communities, and Artmann *et al.* (2003) and in Choo *et al.*'s (2004) papers on algal biomass growth in response to coarse particulate input and oxidative stress respectively. Research was conducted by Dufrêne and Legendre (1997) to identify species of ground beetles as indicator species for soil moisture content in a variety of habitats in Belgium, using abundance and community composition in each habitat patch. However, there is a lack of synthesis in such research, which creates challenges when trying to develop a representative model or theory, particularly for highly variable and dynamic systems such as rivers. Similarly, Meyer *et al.* (2003) discuss the impacts of highly variable flow conditions on lotic communities, and Mellado-Diaz *et al.* (2008) present findings on the response of macroinvertebrates to physical, chemical and landscape factors in semi-arid regions, but neither have conducted studies in groundwater-fed streams, and so their findings, while useful, cannot be generalized to all semi-arid systems. Uys (1997: 8 & 9) describes how biotic communities in variable environments, such as semi-arid riverine systems, are often referred to as "unstructured" or "fluid", as the community is "in a state of perpetual recovery from frequent disturbances" to which individual biota respond differently. In contrast, communities in perennial systems are viewed as stable and well organised (Uys, 1997). While a large portion of South Africa's rivers are non-perennial (Uys, 1997), these systems remain poorly understood as research into riverine habitats and biotic communities has been directed mainly at perennial rivers. Therefore, there is a gap in knowledge in terms of studying macroinvertebrate communities in semi-arid rivers, and the specific tolerances of biota to changes water chemistry and sediment dynamics in such highly variable systems. Furthermore, many of the studies mentioned above were conducted in European semi-arid areas, and many more have been conducted in North America and Australia. Rivers-Moore *et al.* (2008: 11) highlight that southern African Rivers "have extreme flow regimes, displaying twice the world average of flow variability", making it very difficult to apply theories and models based findings on the flow regimes of their northern hemisphere counterparts. Limited literature could be found on either biotic communities or habitat condition in the Karoo region, particularly for groundwater-fed systems in this region, which is an area of concern for South African water management as agriculture and stock keeping (both of which have an impact on water resources) are primary sources of income for many who inhabit the region.

CHAPTER 3: METHODS

According to literature (see Mellado Diaz *et al.*, 2008; Hauer & Lorang, 2004; Puckridge *et al.*, 1998; Pennak, 1971), habitat assessment involves a number of distinct factors that contribute to the overall condition of a river system. While these factors must be measured and assessed individually, the end result of data collection and analysis should produce a multi-layered representation of the system and a dynamic understanding of habitat condition over the given time frame. Due to the time constraints of this study a limited number of factors could be tested, and it was therefore necessary to identify key factors which would provide the most useful results. Those used in this study were selected based on their prevalence in relevant literature, measurability, and necessity for specialist knowledge. Eleven factors were identified as being commonly included in literature, easily measurable and not requiring specialist training, making them time and cost efficient. They were grouped into three broad categories: water chemistry, physical variables, and biota. Water chemistry variables include nitrate and phosphate concentrations, electro-conductivity, pH, dissolved oxygen concentration, turbidity, and water temperature. Physical variables include channel bed substrate, fine sediment storage, water depth, flow speed and discharge. Biota were represented by macroinvertebrates that inhabit the channel bed.

In order to meet with the objectives laid out for this study, the methodology was designed as a step-by-step series of measurements which could be easily replicated in each data collection trip. The two primary objectives presented in the Introduction chapter can be disaggregated into four parts with corresponding methods and measures, as follows:

1. Describe the spatial and temporal instream habitat condition and availability
    a. Site length and width measurements
    b. Long profiles of each site
    c. Discharge transects
    d. Continuous flow records from level loggers
    e. Water chemistry measurements
    f. Visual assessment of substrate composition
    g. Visual assessment of fine sediment (as embeddedness)
    h. Stored bed sediment
2. Describe the biotic community composition
    a. MiniSASS sampling methodology of macroinvertebrates

    b. Frequency of occurrence of macroinvertebrates
3. Investigate the influence of flow and sediment, as system drivers, on habitat quality
    a. Analysis of data collected for objective 1
    b. Comparison of data sets to identify correlations in fluctuations over time and space (addressed in Results and Discussion chapters)
4. Investigate the influence of flow and sediment, as system drivers, on biotic response to habitat changes.
    a. MiniSASS sensitivity scores
    b. Chi-square dependency testing
    c. CANOCO redundancy analysis (RDA)
    d. Hydromorphological Index of Diversity (HMID)
    e. Shannon Index of substrate diversity (H')

It was also important to identify study sites which were representative of the stream channel condition, in terms of physical properties and biological activity. The geomorphological characteristics of rivers vary along their course, making it necessary to use more than one sample site to improve the representivity of findings. A number of field observations were made prior to the start of this study to identify stream sections with variable physical properties and visible biological activity (e.g. riparian vegetation, evidence of living organisms, presence of predatory species like birds and otters), as well as differing levels of disturbance. Within the Wilgerbos River, three stream reaches were selected through a process of elimination as being representative of the upper, middle and lower sections of the channel, hereafter referred to as Site 1, 2 and 3 respectively. The sites were chosen based on levels of activity and disturbance, variability in conditions, arrangement of habitats and ease of access. Each of the sites is accessible by farm road, which was important for safely transporting equipment between them and to reduce the amount of travel time required each day. Site 1 represents a heavily disturbed habitat. As described previously, this site is directly below Ganora farmhouse and is disturbed downstream on a daily basis by vehicles, farm workers and sheep flocks, as well as being directly below a weir. Site 2 represents a slightly disturbed habitat. This site runs along the lower part of Ganora farm property for part of the reach, making it susceptible to run-off from the farmland but with little to no physical disturbance, and is situated above a gorge. Site 3 represents a low disturbance habitat. It is 3 km downstream of Site 2, at the end of a gorge, and lies upstream of De Toren farm, leaving it largely undisturbed. Finally, each site contains a variety of flow types (e.g. riffles, pools), flow speeds, water column depths and substrate types (e.g. sand, cobble, bedrock). This allows for a variety of habitats to form and dynamic habitat arrangement within the sites, and thus allows the sites to support a range of biota.

In each site, data for water quality, hydraulic and substrate condition and biota were collected. Data were collected over a three-day period, approximately every three to four months to allow seasonal changes in habitat quality to be monitored, and to generate data which reflected the timeframe in which changes can occur in the system. The pilot study was conducted in November 2013. Thereafter, the main study comprised five field trips in July 2014, October 2014, March 2015, May 2015 (shortly after a large rainfall event) and August 2015. Between November 2013 and July 2014, the author was overseas on a student exchange and so this period was mainly used for gathering literature and refining the list of variables to measure. Over the course of this study, the methodology for field data collection was altered and improved between the pilot study and the second set of data collection. As a result, some early data sets are incomplete as new methods were introduced later in the study. However, the methodology was kept constant wherever possible and incomplete data sets were treated as such in the data analysis stage.

Assessment of the habitat quality of each site involved a number of methods. For the most part methods were chosen based on availability of equipment and how easily sampling could be done. Methods which could be done in the field and did not require samples to be returned to a laboratory were prioritised in order to avoid contamination or changes in the sample due to biological activity. Delta Environmental Consulting (2014) and The World Health Organisation (WHO) (1997) outline a number of challenges facing scientists collecting data in the field. While it is impossible for field data to be fully accurate, taking laboratory analysis of water samples should ideally be done within a couple of hours of the samples being collected (WHO, 1997). This would not be feasible in the case of this study given the distance, the quality of roads and the number of days required to complete data collection. Other potential risks associated with collecting samples for laboratory analysis can include poorly washed sample bottles, which may contain remnants of previous samples or chemicals from the cleaning process (Delta Environmental Consulting, 2014). Certain cleaning agents react differently with various water quality variables, and the preservation of each variable requires specific conditions to be met, in terms of cleaning agent used, material of the sample bottle and exposure to light (Delta Environmental Consulting, 2014). Furthermore, temperature changes (an inevitable factor when samples are kept overnight, or transported long distances) can alter solute concentrations, creating inaccurate results if samples cannot be stored at the ideal temperature, or tested within the ideal time (Delta Environmental Consulting, 2014). WHO (1997) recommends that samples be tested as they are collected, wherever possible.

3.1 Pilot study

Having selected the three sites, a pilot study was conducted in November 2013, during which time basic physical and water quality data were collected. This included testing for nitrate and soluble phosphate concentrations, electrical conductivity, pH, turbidity and water temperature. Discharge was also measured along a transect in each site, and a long profile of the channel was surveyed to establish slope gradient. During this initial site visit, Solinst level loggers were installed at each site to record continuous data on depth (as water pressure) and water temperature. A barometric logger was kept at Ganora farm, which was used to correct water level data for variations in air pressure. These data were used to gain an initial understanding of the similarities and differences among the three sites, and provided an opportunity to consider why these similarities or differences existed. The pilot study also provided an opportunity to refine the basic methodology, which could then be used for the main surveys.

Following the pilot study, the sampling strategy for the study was planned. Figure 6 provides an illustration of the general sample design used.

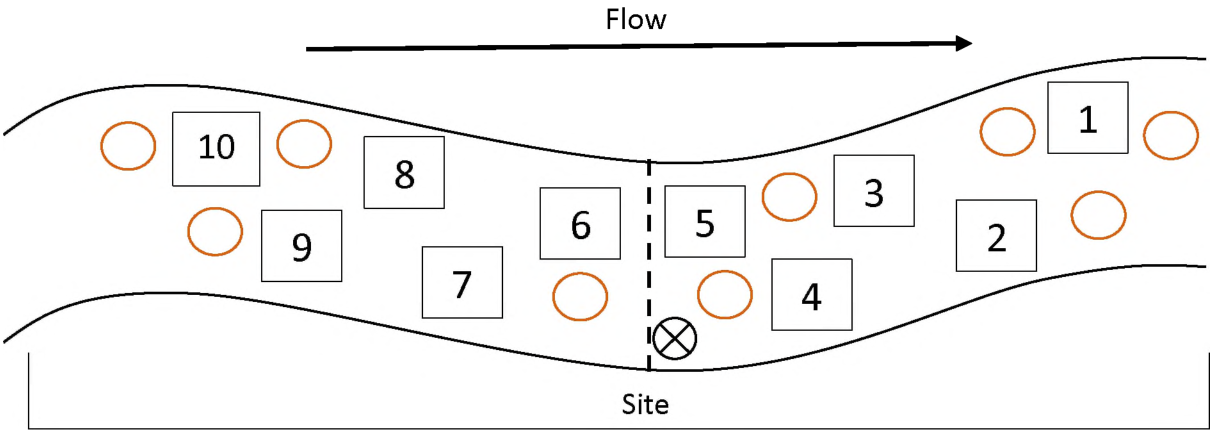

Figure 6: Sample design used in each site. Squares represent bed substrate and biological sampling points. Circles represent stored sediment sampling points. X represents water chemistry sampling point. Dashed line represents discharge sampling transect.

Ten quadrats for biological and habitat condition sampling were spaced out in order to cover as much of each site as possible, while stored sediment was clustered in groups of three at the top, bottom and middle of each site. As stored sediment sampling depended on finding patches of fine sediment on the river bed, the arrangement of sample points varied over the study, but the grouping was similar each time. Discharge sampling and water chemistry sampling were measured in a suitable part of the stream, close to the middle of the study site, which had been marked during the pilot study using metal poles and white paint. The position of the level logger was different in each site as it was necessary to place them where accurate measurements could be taken and where the device could be firmly anchored to a rock. As a result, the level loggers are not depicted on Figure 6.

3.2 Site properties

Habitat quality assessments also included the geomorphological and physical variables of the stream channel in each site. Geomorphology refers to the tangible physical properties which can act as drivers or inhibitors of aquatic habitat quality (Newson & Newson, 2000). In order to compare the three sites, it was necessary to first have an idea of their different physical properties which can influence interpretation of data later on. In this case, the physical properties measured in each site were:

- Site length
- Channel width
- Site gradient

Channel width was calculated using mean values from a number of cross-channel measurements in each site (Gostner, 2012). At points where the channel was not a single thread, the width of each section of the river was noted, as well as the total width. The wetted width was calculated by subtracting the width of the bars and other obstructions from the total width of each transect. As the measurements were taken in August 2015, following a number of high rainfall events, the wetted widths represent high baseflow conditions, similar to those observed in July 2014 and May 2015, but higher than those observed in October 2014 and March 2015. The mean value was calculated for bank-to-bank width, as well as the width of the wetted area.

The site gradient was measured using the average of readings taken above and below each riffle and pool. At points where the stream diverged, readings were taken in both channels, recorded separately as “Channel 1”, “Channel 2” etc.

The presence of algae was also noted in each site, both as field notes and as part of biological and habitat sampling, which is described in *Section 3.6*. While algae was never quantitatively measured, it’s presence was noted as important factor affecting flow, water chemistry and biotic community.

3.3 Hydrology

Hydrological variability has a direct effect on the habitat variables water depth and velocity; aspects of habitat that can be easily observed. As a result, these were a large contributing factor in site selection. The site-scale hydrological properties measured in this study were:

- Discharge
- Water level and temperature (continuous)
- Rainfall

Discharge was calculated based on measurements taken along a transect across each site. While discharge itself is not a habitat variable, it provided a means of monitoring channel condition as a result of flow changes over time, by comparing the data collected at each visit. The total width of the channel at each transect was measured, and the transect divided into approximately 20 equal units, with more or fewer measurements taken in high flow or dead water respectively. At each unit, the depth of the channel was measured and a Flow Mate 2000 portable flow meter was used to measure velocity in the water column (m/s) at 0.4 of the total depth above the stream bed. Discharge per unit was then determined with the formula

$$\mathrm{Q} = (\mathrm{d} \times \mathrm{l}) \times \mathrm{v}$$

where Q is discharge, d is depth of the unit, l is length of the unit, and v is flow velocity. The sum of all the units was calculated to determine the total discharge for each site.

A Solinst Model 3001 level logger was also installed at each site to collect continuous data, at 20-minute intervals, on water level (as water pressure above the logger, presented as depth above the logger in meters) and temperature. The data from the level logger was downloaded during each field work trip and processed using Levelogger 4.0 software and Microsoft Excel to create line graphs showing fluctuations in each variable. This provided information on flow variation at the three sites in between sampling visits. Continuous barometric readings were taken in order to correct water level readings for atmospheric pressure, which was also performed using the Levelogger 4.0 software. During each data collection trip, the depth of water above the level logger was measured manually in each site. These measurements were used to correct for inconsistencies or errors in the level logger data. For example, due to a technical issue, readings in Site 3 were given as negative values, and in this case, the measured depth was used to create a correction factor to make the data comparable with the other two sites. Data from the level loggers were exported from the Levelogger programme as graphs and spreadsheets for analysis. As data was recorded for water pressure, water temperature and conductivity every 20 minutes, with approximately three-month intervals between data retrieval, this provided a large number of values. The data were simplified by grouping recordings by day and month, and calculating averages and totals for each data set, to allow for easier representation in graphs and data interpretation.

Water level, rather than discharge, was used to describe habitat variability as it determines bed shear at high flows for sediment transport and relates directly to variations in water depth, which is a key variable in biotic community composition. Attempts to establish rating curves of discharge against water level did not provide a useable relationship, possibly due to changes in the bed profile as a result of erosion or deposition between measurements.

Daily rainfall data for the period 1 January 2013 to 31 November 2015 were acquired from neighbouring farms, and were used to create monthly records for the months before the first data sampling and between each subsequent trip. The longer data sets allowed the study period to be situated in a broader climatic setting. These were compared with the data from the level loggers to explain water level fluctuations, and were later compared with the long-term rainfall records presented in Figure 3 to observe irregular rainfall patterns experienced in 2015.

Velocity and depth were also measured at a patch scale, as part of the habitat descriptions for biological sampling. Velocity was again measured using the Flow Mate 2000 portable flow meter, but readings were taken directly above the stream bed to measure the velocity felt by organisms living in the substrate. As the sample quadrat was only 1 m^2, generally only one velocity measurement was taken in each patch, but in some patches, it was necessary to take two or three measurements and calculate the average velocity to account for flow diversity or obstructions such as large boulders. Similarly, depth measurements were taken in the centre and, where applicable, at each edge of the quadrat, and the average depth for the patch was calculated. As far as possible, patches which contained highly variable velocity and depth were avoided to create as clear a relationship as possible. These velocity and depth measurements were used to create a hydromorphological index of diversity (HMID) for each site and perform redundancy analyses using the CANOCO programme, both of which will be discussed in *Section 3.7.*

3.4 Water chemistry

Water chemistry can be an important driver of aquatic habitat, as the concentrations of dissolved constituents are regulators of primary and secondary productivity, and will determine what organisms are able to survive in a stream (Rigsbee *et al.*, 2013). Dissolved solutes make up a large proportion of the total transported load of a stream, and can vary greatly according to regional climate and underlying geology (Rigsbee *et al.*, 2013). The water quality variables tested in this study are presented in Table 2.

Turbidity was added to the study at the end of the first set of data collection, as field observations raised questions about water clarity. Although not strictly a measure of turbidity, the reading given in ppm was used, rather than a reading given in NTU, as this better reflects sediment concentration in the water column. As sediment dynamics were a primary focus of this study, it was considered more valuable to measure suspended sediment than dissolved solids. The reading given in ppm is calculated by the turbidity meter from turbidity measured in NTU.

Table 2: Water quality variables measured in each site, including units, and measurement range and detection limit of equipment. Equipment used: 1. Hanna Combo pH and EC meter; 2. AZ8403 handheld DO probe; 3. Visocolor Alpha colorimetric test kit; 4. Visocolor ECO colorimetric test kit; 5. Partech 740 handheld turbidity meter.

Variable	Units	Meas. Range	Detection limit
Electrical conductivity^1 (EC)	µS/cm	0 – 3999	1
pH^1	__	0,00 – 14,00	0,05
Water temperature^2	°C	0 – 50	1,5
Dissolved oxygen^2 (DO)	% saturation	0 – 199,9	0,3
Nitrate^3 (N)	mg/L	2 – 50	2
Soluble Phosphate^4 (P)	mg/L	0,2 – 5	0,2
Turbidity^5	ppm (from NTU)	0 – 200	Unknown

Water quality variables were measured at a single point near the middle of each site as they would not vary significantly over the scale of a site. These variables were measured first to avoid any contamination by activity in the stream or disturbance of the stream bed during the data collection process. Readings were taken between 09:30 and 10:00 on each day of data collection. When measuring turbidity, temperature and DO, the probes were held at a constant depth, in the middle of the water column, in order to avoid disturbing bed sediments or causing mixing of surface water with bottom water, which may have altered the readings. Point measurements of temperature were superseded by the continuous temperature readings taken by the level logger, but were recorded in case any unusual trends were noticed in biological sampling, which may have relied on specific temperature measurements.

3.5 Bed sediment and substrate

Substrate is referred to many times in literature as an important driver of habitat type as many organisms are limited to one type of substrate during each stage of their lifecycle, because of feeding, breeding or anchoring requirements (see Griffith & Walton, 1978; Artmann *et al.*, 2003; Apitz, 2012). In this study, substrate was assessed visually and measured quantitatively using three methods. Visual assessments were conducted using a stratified random approach throughout each site, in ten 1 m x 1 m quadrats (hereafter referred to as patches), trying as far as possible to include a range of different flow speeds, water column depths and substrate types. The GPS co-ordinates of each patch were marked to enable sampling of similar locations, within the margin of error of a handheld GPS device, each time field data were collected.

At each patch a set of tables on description of sediment properties, taken from Gordon *et al.* (2004), was used to assess sorting values and embeddedness. In accordance with this method, the size class of the largest and second largest clast types were recorded (as in Table 3), and the percentage cover of fine sediment in each patch was estimated (as in Table 4).

Table 3: Clast size and descriptions, adapted from Gordon et al. (2004)

Code	**Description**	**Particle size (mm)**
1	Fines	Sand and smaller
2	Small gravel	4 – 25
3	Medium gravel	25 – 50
4	Large gravel	50 – 75
5	Small cobble	75 – 150
6	Medium cobble	150 – 225
7	Large cobble	225 – 300
8	Small boulder	300 – 600
9	Large boulder	>600
10	Bedrock	

In each patch substrate was further visually assessed as percentage cover of each clast category to assess material size distribution and the percentage of fine sediment was given a score between 1 and 5 according to Minshall's (1983, cited in Sylte & Fischenich, 2002) embeddedness rating table (Table 4).

Table 4: Embeddedness ratings, adapted from Minshall (1983, cited in Sylte & Fischenich, 2002).

Rating	Description
5	>75 % of surface covered by fine sediment
4	50 – 75 % of surface covered by fine sediment
3	25 – 50 % of surface covered by fine sediment
2	5 – 25 % of surface covered by fine sediment
1	<5 % of surface covered by fine sediment

These scores were used as descriptors of substrate in each patch and as a measure of changes in sediment quantity in each site over time. Box-and-whisker plots and modal scores were also calculated to assess the embeddedness preferences of instream biota.

From this, the level of embeddedness was determined and the sorting class of the substrate was selected based on the diversity of clast sizes within the patch area (Figure 7). Sorting data were used in conjunction with biological sampling data to create a Shannon index (see *Section 3.7*) to assess the correlation between biotic diversity and substrate composition (Boyero, 2003).

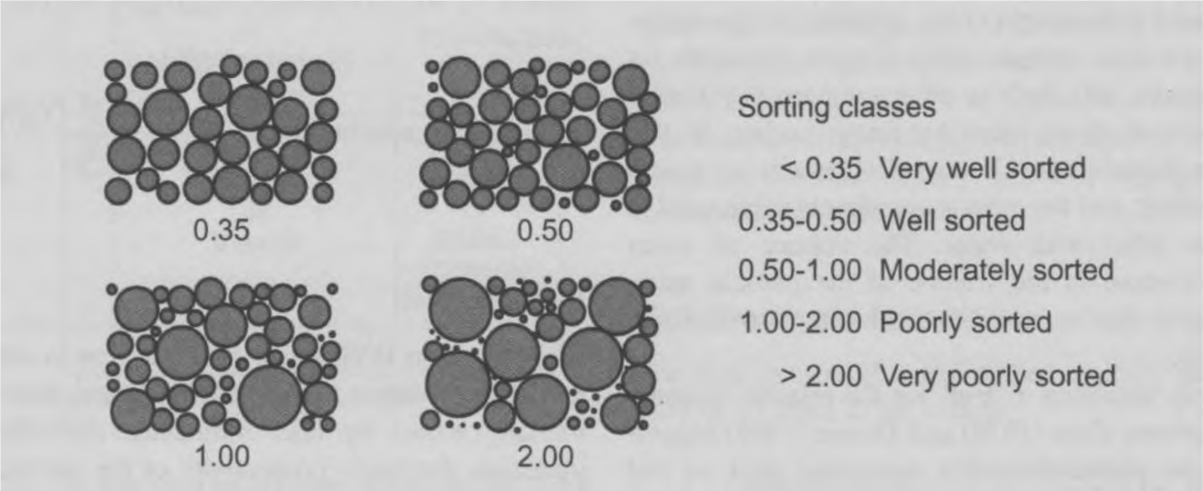

Figure 7: Sorting classes as presented by Gordon et al. (2004).

These visual assessments were reinforced by a simple but effective quantitative measure of fine sediment storage in the stream bed, devised by Lambert and Walling (1988). A large bucket with the bottom removed was firmly pressed into the substrate of the channel bed to create a standardised area of bed to be sampled. The depth of the water in the bucket was measured with a tape measure. The bed was disturbed with a sturdy stick to a depth of approximately 5 cm, stirring in a clockwise direction for approximately 20 seconds. Once the bed had been sufficiently disturbed, a 250 ml water sample was taken, containing the disturbed bed sediments. This method was used to assess the variation in fine sediment accumulation over time. This was originally done three times in each site (at the top, middle and bottom of each site). However, to improve accuracy of data analysis, the sample size was increased to nine in each site. This method is limited by the substrate of the channel bed, as samples can only be taken where the bucket can be pressed into fine sediment or gravels, and where the bed material is loose enough to be stirred. Further, each patch can only be sampled once per field visit, as the substrate must be undisturbed for the samples to adequately represent sediment storage. It is not possible to use this method to test fine sediment storage in areas of the channel bed that are dominated by large gravels, cobbles or boulders, or areas with densely packed material or armouring that cannot be dug into. In such areas, the embeddedness score was used to assess fine sediment storage.

The lab analysis of these samples involved a simple gravimetric process to determine the concentration of sediment in each bottle. Each sample was decanted into a beaker and weighed in its entirety (the weight of the empty beaker being recorded separately). The samples were then left to stand to allow the sediment to settle, after which the clear water was drained off. The remaining sediment residue was dried in an oven at 50°C for at least 24 hours[1], and the beakers were reweighed. The total volume of water in the bucket was calculated and the concentration of sediment was calculated as a proportion of this, from which the amount of fine sediment stored in the bed at each sample point was determined.

3.6 Biological sampling

The biota within the stream were assessed for species richness and distribution in relation to physical and flow properties. The biota which inhabit an ecosystem are important indicators of the overall properties of habitats within it. Changes in the habitat itself will result in changes in the types of organisms and therefore organisms can be used as indicators of change or stability in a habitat (Graham *et al.*, 2004). In this case, biological assessment was used to monitor the relationship between habitat condition and biota, through the response of biota to changes in the habitat condition. A number of biological sampling methods exist and a wide variety have been used in the literature underpinning this study. However, many of the methods do require specialist biological knowledge and were therefore considered unsuitable for this

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[1] The drying time for samples depended on the sediment type i.e. clay, sand, silt, and the organic matter content of the sample. 24 hours was set as a constant minimum to ensure the majority of samples would be dried.

study. The research therefore made use of an adaptation of the MiniSASS methodology, designed specifically for the South African context.

MiniSASS, a simplification of the South African Scoring System (SASS) methodology, was used in this study to monitor the diversity and distribution of macro-invertebrate species in the three study sites. SASS was originally designed as a biomonitoring technique for water quality which makes use of the sensitivities of different macroinvertebrates to water pollution (Graham *et al.*, 2004). In a full SASS assessment, a datasheet with 90 key macroinvertebrate families is provided. Biotopes are sampled using fine mesh nets and sieves to find biota in sediments, the water column, and instream and riparian vegetation. The presence of certain key species with high sensitivity (such as mayflies or stoneflies) will indicate good water quality, while the presence of only species with low sensitivity (such as worms or leeches) may suggest lower quality water. The macroinvertebrates found in each biotope are counted and recorded on a specially designed spreadsheet, which requires the biotope to be recorded for each taxon found, and includes estimates of abundance per biotope. From this, overall water quality can be calculated based on the scoring system of each taxa's sensitivity to changes in water chemistry and the presence of pollution or disturbance.

MiniSASS employs the principles of SASS, but only uses 13 key macroinvertebrate taxonomic groupings at the order level rather than family (datasheet available online at: http://www.minisass.org.za). While organisms are collected in the same way, from the same range of biotopes, the simplified groupings mean organisms do not need to be individually identified. Instead, water quality is measured based on the general sensitivities or preferences of each group, across all the biotopes (Graham *et al.*, 2004). MiniSASS is simply used to assess water quality and so lacks a measure of biotic abundance as part of its method, but uses simpler terminology than SASS and does not require any prior experience or training in identifying taxa.

Habitat is highly variable over an area due to variable substrate. Therefore, multiple samples were collected from across each site. The ten 1m x 1m patches used for substrate assessment were also used for the MiniSASS assessment. In each patch the depth of the water column and flow velocity at the stream bed were measured for later use in assessing habitat preferences of each group These measurements were not used to describe the properties of each site, as the point style sampling was not considered reliable for site scale descriptions. However, these were used later to conduct a RDA of the correlation between biotic community structure and flow variables (see *Section 3.7*). Filamentous algae was also noted in each patch, which was later used as an indicator of changes in algal growth over time.

In order to identify macroinvertebrates associated with the bed sediment, fine substrates were sieved, while large and coarse substrates were sorted through by hand. Using the MiniSASS key classes, each taxonomic group of macroinvertebrate found (not individual organisms) was listed and the flow and substrate properties of each patch were recorded. Once the biota had been identified, a sensitivity score was calculated for each patch, using Table 5.

Table 5: Taxonomic groups and sensitivity score sheet used in MiniSASS assessment, adapted from: http://www.minisass.org/media/filer_public/2013/06/28/minisass_grade_11.pdf

Taxonomic groups	**Sensitivity score**
Leeches	2
Trueflies	2
Flatworms	3
Worms	3
Damselflies	4
Snails	4
Bugs or beetles	6
Minnow mayflies	6
Crabs or shrimps	7
Dragonflies	7
Caddisflies	9
Other mayflies	13
Stoneflies	14
TOTAL SCORE	
NUMBER OF GROUPS	
AVERAGE SCORE	

While the dichotomous key used for identifying organisms in the MiniSASS system includes 16 groups, the scoring table uses only 13 as it combines some groups, such as beetle larvae, beetles and bugs, and cased and uncased caddisflies, based on their sensitivity score. There are three variations on the scoring table, but all rank organism sensitivity in the same order. Therefore, the table which is used is less important than maintaining consistency throughout the study.

Groups are given a sensitivity score between 2 and 14, where 2 is "tolerant of organic pollution stress" and 14 is "intolerant" (Gordon *et al.*, 2015). The presence of organisms with higher sensitivity scores is therefore considered more indicative of good quality water in the stream, than that of low scoring organisms. Similarly, organisms which have highly specific habitat requirements will have a limited range and will be more strongly affected by changes to habitat condition as a result of physical or chemical variables (Bunn & Arthington, 2002; Jowett, 2003; Oliva-Paterna *et al.*, 2003).

MiniSASS or SASS is normally assessed for a site but, for the purpose of this study, MiniSASS assessments were conducted in individual patches across the site, which were scored separately based only on the biota found within them. Therefore, 10 sensitivity scores were calculated for each site. These were later used in comparison with substrate and embeddedness assessments to assess habitat preferences and sensitivity of each group. Observed habitat preferences were contrasted with expected preferences based on information found through research. Table 6 describes the expected preferred substrate type, feeding habits and other general habitat requirements of each taxonomic group in the MiniSASS system.

The groups were sorted into their preferred habitats, based on the conditions in which they were found and further research was conducted into the behaviour and habitat requirements for each group, to compare existing knowledge with the findings of the study. Each group has different habitat requirements based on the habits of the organisms. In each case, the information is generalised, but the profiles provide a broad idea of the needs and sensitivities of each group.

The frequency of occurrence by patch was used to measure the abundance of each taxonomic group at a site. Organisms with low MiniSASS sensitivity scores, and which occurred in a number of patches of differing habitat categories, were noted as being non-selective in terms of habitat condition and therefore less important markers of variability in biological community composition.

Table 6: Summary of preferred substrate, feeding habits and other habitat requirements of each taxonomic group in the MiniSASS system (summarised from Thorp and Rogers (2010), Brady (2016) and the MiniSASS website created by GroundTruth (2016)).

Taxonomic group	Substrate	Feeding	General habitat
Leech	Non-selective	Carnivore/scavenger	Able to survive in anoxic conditions
Truefly	Fine sediment	Carnivore/filter feeder	Able to survive in a variety of substrates
Flatworm	Rocky	Carnivore/scavenger	
Worm	Fine sediment	Microorganisms in substrate	Able to survive in low oxygen conditions
Damselfly	Rocky	Carnivore/predator	Hunt among riparian vegetation
Snail	Rocky	Grazer	
Bug/beetle	Fine sediment	Grazer	Some beetles are predators
Minnow mayfly	Rocky	Grazer	
Crab	Rocky	Omnivore/scavenger	Feed in fine sediment
Dragonfly	Fine sediment/ gravels	Carnivore/predator	
Caddisfly	Rocky	Cased: scavenger/ grazer Uncased: carnivore/ predator	Cased: cling to rocks Uncased: able to swim and hunt
Mayfly	Rocky	Grazer	
Stonefly	Non-selective	Grazers	Sensitive to low oxygen content.

After each set of field data was collected, the frequency of occurrence of the groups in each site was compared, and general habitat conditions were noted. Other organisms such as fish, algae and riparian vegetation were also noted where present but did not form part of the primary biological assessment of habitat condition.

Having completed all the scheduled field trips, the data were collated into a single Excel document to compare the changes in each data type over time.

3.7 Ecological response

The condition of a habitat, and therefore its suitability for invertebrates, is the result of a complex relationship between depth, velocity and substrate (Jowett, 2003). This relationship means that neat connections between habitat conditions and invertebrate populations are difficult, if not impossible to establish. Jowett (2003) states that species of invertebrates can often be found in a variety of habitat types, suggesting an underlying system of microhabitat interactions which can increase the abundance and range of a species.

The first stage of data analysis for the biological response assessment was to calculate the MiniSASS sensitivity score for each patch. This was done using the average score derived from Table 5. Embeddedness scores recorded during the MiniSASS field assessment were used to establish whether patches were sandy or rocky types. Patches with an embeddedness score of 4 or 5 were considered sandy type, and patches with an embeddedness score of 3 or lower were considered rocky type. Habitat patches were then classified as either sandy or rocky type patches (Table 7). Ordinarily, MiniSASS scores are calculated for the whole site, not per habitat patch. The per-site method was used in this study in comparison with the Hydromorphological Index of Diversity (HMID, as described below). However, patch-scale scoring was also necessary for comparison with substrate assessments, which became a dominant factor in this study and so patch scale scores proved to be more valuable, and indicative of inter-site variability than site scale scores.

Table 7: Sensitivity scores and ecological condition derived from MiniSASS assessment, available from: http://www.minisass.org/en/how/

Ecological condition	**Sensitivity score by patch type**	
	Sandy Type	Rocky Type
Unmodified (Natural)	>6,9	>7,9
Few modifications (Good)	5,8 – 6,9	6,8 – 7,9
Moderately modified (Fair)	4,9 – 5,8	6,1 – 6,8
Largely modified (Poor)	4,3 – 4,9	5,1 – 6,1
Seriously modified (Very poor)	<4,3	<5,1

The Chi-square (X^2) method was used (as described by Silk, 1979) to test for dependence or independence between the number of patches of each ecological condition category, and each of the following variables: sample date, site, and bed material per patch. In each case, the X^2 values were tested for significance at $p < 0,05$, and the critical value (CV) for each was determined using the lower tail CV table (available from: http://www.itl.nist.gov/div898/handbook/eda/section3/eda3674.htm).

A redundancy analysis (RDA) conducted with the CANOCO software, a modification of DECORANA created by ter Braak (1988), was performed to test for correlation between biological diversity and the physical and chemical habitat variables: site, sample date, water chemistry, flow properties (depth and velocity per patch, and discharge and maximum flood peak per site), embeddedness per patch and dominant clast per patch based on Gordon *et al.*'s (2004) categories. Each set of variables was tested individually in eight separate runs. The test makes use of Eigenvectors to assess changes in community structure in response to other variables, i.e. the influence of variables on community composition. RDA is a form of direct gradient analyses which uses both biotic and environmental data to tell us how species composition is related to the measured variables (Lěps & Šmilauer, 2003; Van den Brink *et* al., 2003). This means that the arrangement of species along the axes is constrained by environmental gradients (Prentice & ter Braak, 1988; ter Braak & Šmilauer, 2002). In this ordination approach, the largest and second largest parts of the total variance of the data sets (latent variables) are used to create two sets of axes along which the ordination charts can be created (Van den Brink *et al.*, 2003). This analysis produces tri-plots in which environmental data (habitat variables) are indicated by arrows, where the direction of the arrow shows the direction change across data sets and the length of the arrow shows the strength of correlation, and taxonomic groups are indicated by points, where the position of a point corresponds to optimum environmental conditions for that group (Prentice & ter Braak, 1988). Taxonomic groups found in similar habitat conditions are located close together, while points which are widely dispersed represent groups found in differing environmental conditions. The position of a point in proximity to an arrow is explained by Prentice & ter Braak, (1988: 236): 1) Points lying in roughly the same direction as an arrow have a positive correlation to that variable, 2) points lying at a right angle to an arrow have a "near-zero correlation", and 3) points lying in the opposite direction from the arrow have a negative correlation. The percentage of variance in the relationship between each taxonomic group and habitat variables is used to show the strength of the analysis. It reflects how much variability in the species data can be accounted for by the environmental variables – the higher the percentage, the better the analysis (Lěps & Šmilauer, 2003). The data used only reflected presence or absence of a taxonomic group, presented as 1 = present and 0 = absent. The data did not reflect abundance of taxonomic groups per site or per patch. Therefore, it was not necessary to transform the data, as it did not contain any high values (such as species with highly variable abundance scores) which would affect ordination (ter Braak & Šmilauer, 2002).

Relationships were more closely examined here than in the chi-square analyses, and the data was able to show not only the presence or absence of relationships, but also to provide information on the strength of relationships and negative or positive correlations. The significance threshold for each relationship was $p = 0,05$, tested using Monte Carlo Permutation testing (ter Braak & Šmilauer, 2002). The results of each run were then further examined with other charts and statistical analyses.

Abundance data for each taxonomic group was calculated for each site based on their frequency of occurrence across the 10 patches. Abundance was calculated for each data collection period and used to compliment the sensitivity scores calculated as above. The structure of this research is therefore similar to that of Dufrêne and Legendre (1997) in Southern Belgium, but made more complex by the nature of semi-arid aquatic ecosystems.

A habitat diversity index was created for each site using the Hydro-morphological Index of Diversity (HMID) (Gostner, 2012), based on the average of the depth and velocity readings taken in each patch during the biological sampling (see Appendix 2 for full data set)

$$\mathrm{HMID_{site}} = \left(1 + \frac{\sigma_v}{\mu_v}\right)^2 \times \left(1 + \frac{\sigma_h}{\mu_h}\right)^2$$

where v = velocity, h = depth, σ = standard deviation, and μ = mean.

The index was designed to be a tool for river restoration projects, and uses the coefficient of variation (CV) of flow velocity and water column depth readings from within a section of river (Gostner, 2012). The index was plotted against the number of taxonomic groups found in each site to assess the impact of flow diversity on biotic diversity.

Taxonomic groups in each site were then divided into three categories: fine sediment habitat, rocky habitat, and non-selective habitat (Table 6). At a site scale, the total abundance of biota within each habitat preference group was plotted against stored sediment collected during each data collection period. At a patch scale, presence of biota was compared with embeddedness scores in each patch. The embeddedness categories devised by Minshall (1983, cited in Sylte & Fischenich, 2002) were used to categorise the quantities of fine sediment in each patch. From this, box and whisker plots were created displaying the maximum, upper quartile, median, lower quartile and minimum embeddedness score for each group. Modal embeddedness scores were calculated for the taxonomic groups using the embeddedness scores for all the patches in which each group was found. Thus, the charts show the full range of embeddedness tolerance, while modal scores show the most common, or preferred level of embeddedness per group.

The Shannon diversity index (H') was used to assess heterogeneity of substrate within each patch based on the number of substrate types and proportion of the site made up of each type

(Boyero, 2003). The heterogeneity score was calculated per patch using the substrate description categories from Gordon *et al.* (2004), with the maximum possible score being 2.3, i.e. each substrate type being equally represented within the patch. A low H' score indicates low clast diversity, either due to high embeddedness of the substrate, or high proportion of large clasts. In this study, a low H' score predominantly indicates high embeddedness, as will be explained in the Results and Discussion chapters. The formula below was used to calculate the scores, which were then plotted against the number of taxonomic groups found in the patches.

$$H' = \sum -(P_i \ln P_i)$$

where p_i is the proportion of the ith substrate type in the dataset

A scatter plot was created for each month in which data was collected, to establish trends in species diversity in relation to both substrate and changes in flow conditions. Substrate heterogeneity (H') scores were then plotted against MiniSASS sensitivity scores to assess the degree of correlation between sensitivity scores and habitat condition, to determine whether MiniSASS could be used to measure habitat health. HMID scores were also plotted against sensitivity scores per site to assess the influence of physical flow properties on biotic health.

The results derived from the ecological response methodology were key to this study, with the other measurements and assessments predominantly serving to create raw data sets and to provide a context for the analysis of these results. The range of methods used reflects the complex nature of habitat assessment and the large number of number of contributing factors.

CHAPTER 4: RESULTS

The results of data collection and analysis were grouped into three categories: site properties, habitat quality, and ecological response. While each category was analysed and presented separately, the results are interrelated and are more useful when used together as a means of measuring the health of biotic communities supported by the habitat. Site properties include any data which describe the physical characteristics of the sites, such as site length and width, water slope profile and bed materials. These data provide insight into the potential available habitat within each site, and may help predictions of the kinds of habitat patches one could find in them, based on the steepness of the gradient or the width of the channel. However, they do not show the quality of these habitats and so were used only to contextualise the habitat quality data. Habitat quality was further broken down into flow properties (discharge, velocity, depth and water level), water chemistry, and bed sediment. These data were used to assess and describe the physical condition of the habitat patches within the sites. Ecological response integrates the habitat quality data with biotic data collected through the MiniSASS method. Ecological response was measured using the MiniSASS sensitivity score for the dominant substrate type, the Shannon index of substrate heterogeneity, the Hydromorphological Index of Diversity (HMID) for flow speed and depth heterogeneity, and a number redundancy analysis (RDA) assessments using the CANOCO 4.5 software. Biotic abundance was also assessed based on the frequency of occurrence of each taxonomic group. The bulk of the statistical analysis for this study was conducted using ecological response data.

A redundancy analysis (RDA) was run on all data sets, to test for relationships between the presence or absence of taxonomic groups and sites, sample dates, water quality variables, flow properties, fine sediment percentage per patch, and substrate per patch. Relationships were tested for significance with a threshold of p = 0,05. This analysis produces tri-plots where arrows depict environmental data (in this case, time, site properties, water chemistry, flow properties, and bed sediment and substrate), pointing towards higher correlations between variables and taxonomic groups. Taxonomic groups are presented as points along the gradient of each arrow, where closeness to the arrow shows a strongly positive correlation, while distance shows a weaker or more negative correlation. Taxonomic groups found in similar habitat conditions are located close together, while points which are widely dispersed represent groups which were found in less popular or common conditions. The percentage of variance in the relationship between taxonomic groups and habitat variables is presented for both axes in the figure captions. Each set of data was further examined using different methods that complemented the RDA analysis. The full set of tabulated data used for this analysis is included in Appendix 1.

4.1 Site properties

The lengths of the sites (shown in Table 8) were not standardised in order to avoid breaking up habitat sections, such as riffles or pools. Measurements were taken from the point where one habitat type met another. The top of Site 2 was largely made up of closely grouped riffle sections, therefore the length of this site was greater than the other two, in order to accommodate this. Site 2 was also more complex in terms of habitat diversity, as it included riffles, deep pools, standing water and bedrock sections, thus the greater length also provided the opportunity to better represent this diversity.

The mean width of the multiple cross-sections measured in each channel (shown in Table 8) was fairly similar, but the physical features of the channel area varied within each site.

Table 8: Physical site properties: mean water width (m), mean channel width (m), percentage of channel occupied by water, site length (m) and water slope gradient.

	Site 1	**Site 2**	**Site 3**
Mean channel width (m)	10,4	8,1	9,2
Mean water width (m)	6,7	6,6	5,9
Percentage of channel occupied by water (%)	65	82	65
Site length (m)	147	214	137
Site gradient	0,004	0,013	0,012

Site 1 is not constrained by bedrock and has a lower flow velocity, creating a broader, shallower channel. However, the low flow also results in higher deposition, creating large benches and bars made up of material such as gravel and cobbles. Furthermore, the deposition of very large material during the flood in 2011 has created ‘boulder bars’, where the channel is split by boulders. This produces a wide channel of which a relatively low proportion is occupied by water. Because Site 2 flows through a gorge, the channel width is constrained on the right-hand bank (looking downstream) by dolerite, and the lower end of the site flows over bedrock, preventing lateral erosion and channel widening. The stream section is primarily single thread, and so the mean water width makes up the bulk of the channel width, as the section has few channel bars. Conversely, Site 3 has the lowest mean water width, and a similarly low proportion of the channel is occupied by water, due primarily to the high level of braiding in this section, particularly in the lower end of the site.

Site 1 and 3 are therefore very similar in terms of length and percentage of water in the channel, despite the clear differences in their long profiles, which will be discussed next. Site 2 is both longer and has a larger percentage of water in the channel, despite having a long profile similar to that of Site 3.

The site gradient is an important factor in flow velocity for each site. Figure 8 (a – c) shows the longitudinal slope profiles for each site.

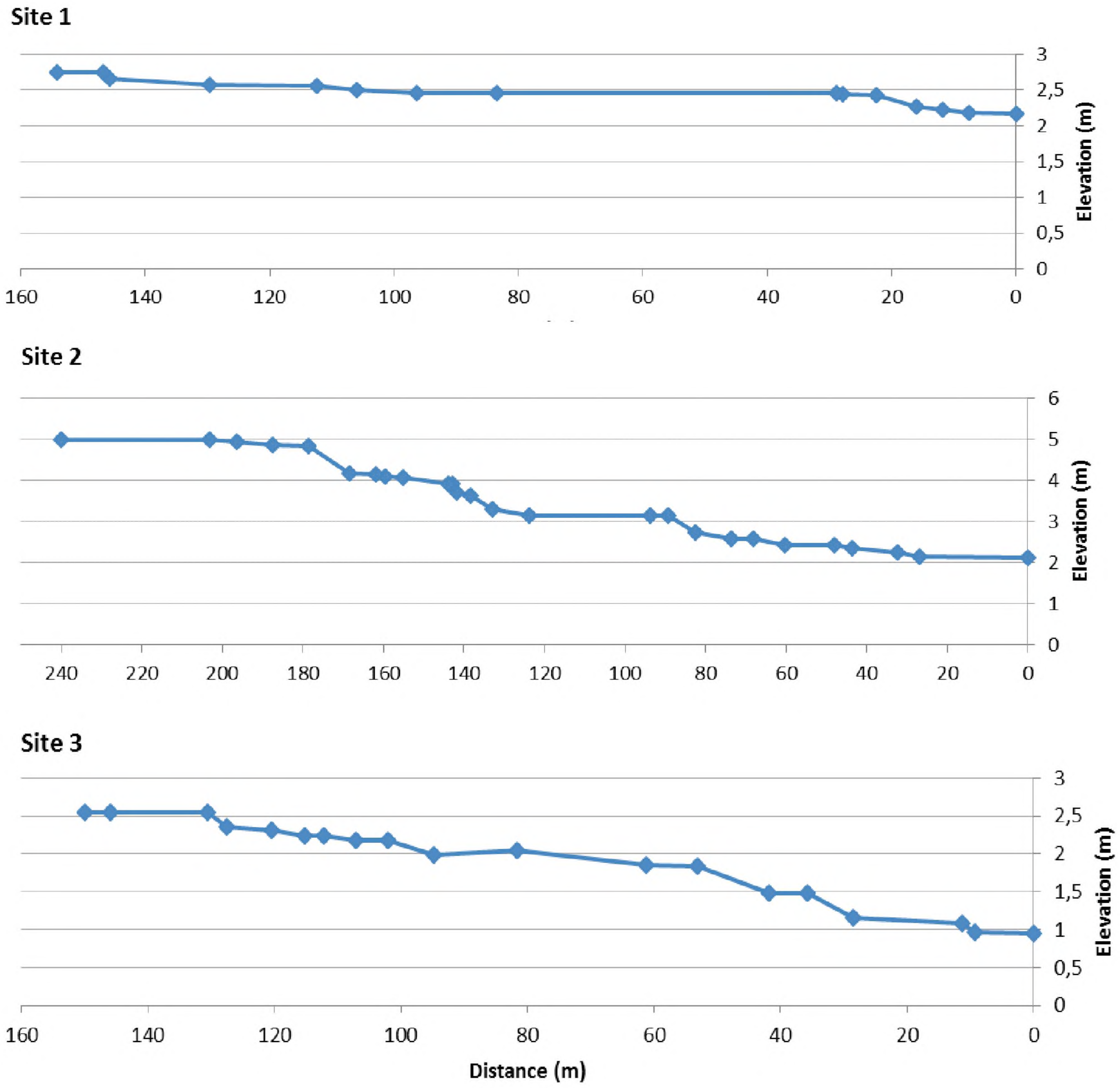


Figure 8 (a – c): Longitudinal slope profiles for Site 1, Site 2 and Site 3

Site 1 has the lowest slope gradient (0,004), creating low-velocity flow as discussed above. Site 2 has the steepest gradient (0,013), with clear knick-points which create fast-flowing riffle sections. Near the middle of the profile, a short stretch of level ground creates a deep pool. The top and bottom end of Site 2 have a low gradient, producing low velocity sections of channel. Site 3 has a similar gradient to Site 2 (0,012), and has knick-points which create riffle sections, as well as stretches of low-velocity flow at the top and bottom of the site. However, the profile is less stepped than that of Site 2 and has no long stretches of low flow other than the top of the site. Diversity of substrate and flow were important considerations in site selection, as the biotic sampling required a variety of habitat conditions to be sampled, in order to give a more reliable indication of habitat quality in the stream.

The presence of algae was also noted in each site during data collection. While the quantity of algae was never quantitatively measured, Table 9 presents the number of patches in each site in which algae was found. This is not an accurate representation of algal growth, but serves to indicate how the presence of algae increased and decreased over time. Algae was generally lower in Site 1 and higher in Site 2. Algae was most common in March 2015, and least common in August 2015. It is worth noting that while August 2015 had low presence of algae in the patches sampled, a number of large clumps of dead algae were observed in the site, but were not anchored to the substrate and no longer lay within the sample patches.

Table 9: Number of patches in which algae was found in each site in July 2014, October 2014, March 2015, May 2015 and August 2015

	Jul 2014	**Oct 2014**	**Mar 2015**	**May 2015**	**Aug 2015**
Site 1	5	2	7	3	2
Site 2	7	3	9	8	5
Site 3	6	6	8	2	1
Total	**18**	**11**	**24**	**13**	**8**

4.2 Habitat quality: Hydrological drivers

Flow properties for each site included measurements of discharge and mean monthly and daily water levels in each site. Rainfall was also an important consideration and monthly and daily rainfall data for 2013 to 2015 was also analysed for the periods between data collection to contextualise the findings.

Rainfall

The Karoo is not a recognised winter rainfall area, but does receive precipitation in winter, in the form of rain and snow (Foster & Rowntree, 2012). Average monthly rainfall data show high rainfall in the summer months and low rainfall in winter months (Figure 3 and Figure 10). Data for 2013 and 2014 followed this trend but the study period experienced a number of unexpected rainfall trends. Daily rainfall data collected on farms higher in the catchment, Quaggasvlei and Dalveen, show the highly variable nature of rainfall events. Rainfall charts (Figure 9 and Figure 10) show that significant storms occurred in December 2013, March 2015, June and July 2015. Despite the relatively high rainfall in March 2015, much of it fell late in the month, so drought conditions prevailed during data collection, which took place in early March.

While typically low rainfall was experienced in July 2014 (only 5 mm at Dalveen), between August 2014 and November 2015 the data for both farms show an unusual trend of consistent, but quite low rainfall throughout the year. Summer rainfall was particularly low, showing none of the peaks seen in the previous years, while winter rainfall was far higher than normal. The highest rainfall for any of the summer months (December to March) was 94 mm at Quaggasvlei in March 2015, in contrast with 123 mm in March 2013 and 187 mm in February 2014. In contrast, July 2015, typically a low rainfall winter month, received 70 mm of rain at Dalveen. This means that the data for the 2015 portion of study reflect unusually dry summer conditions, and unusually wet winter and spring conditions in comparison with the data for 2013/14.

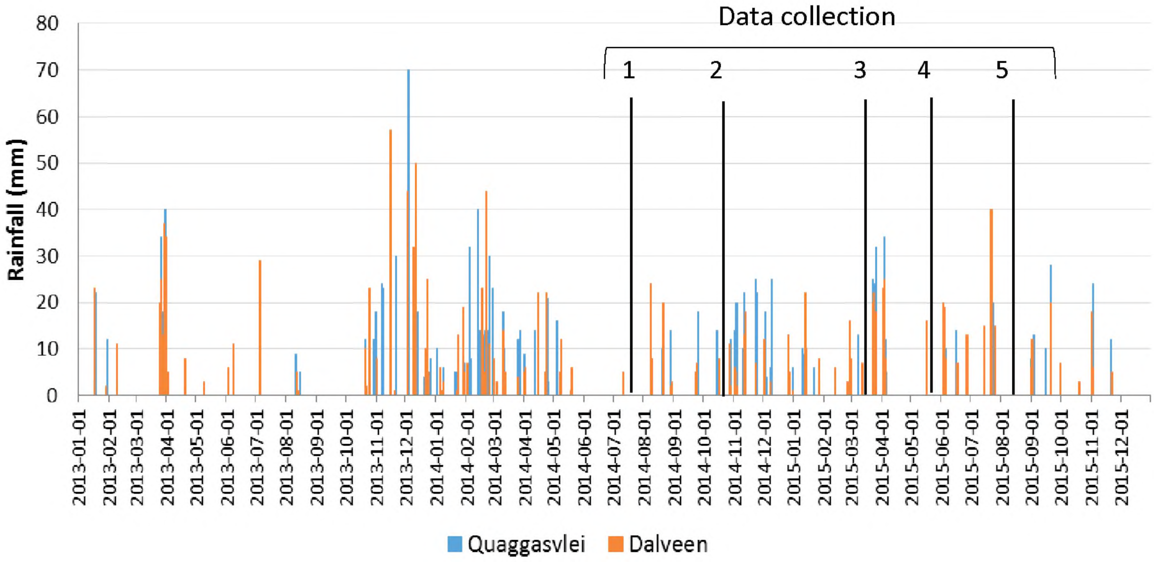


Figure 9: Daily rainfall for Quaggasvlei and Dalveen for the period of January 2013 to November 2015.

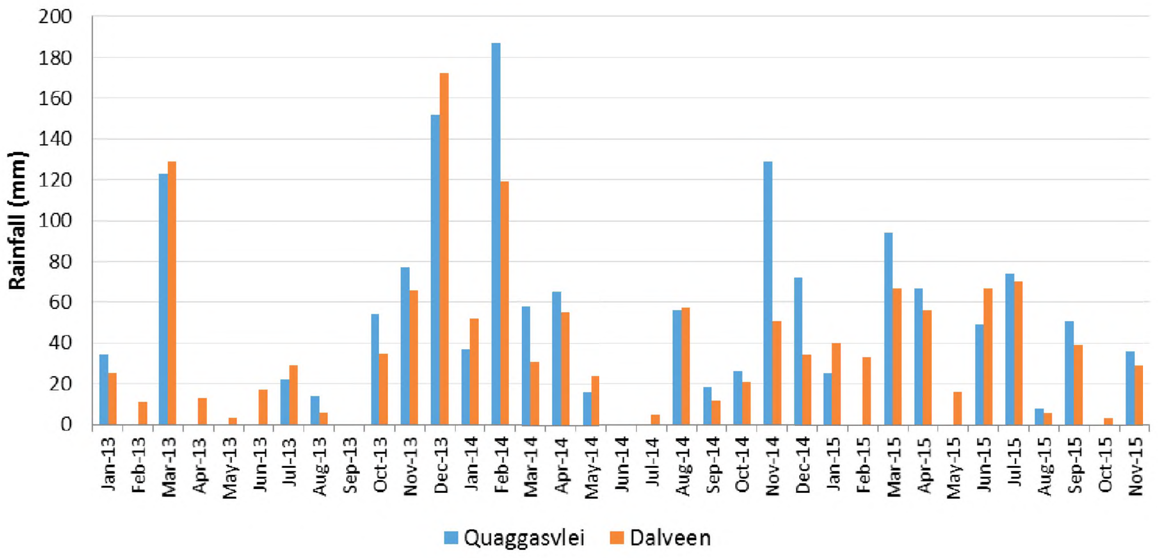


Figure 10: Mean monthly rainfall for Quaggasvlei and Dalveen from January 2013 to November 2015.

June 2014 and July 2014, prior to the first set of data collection, was a dry period during which, only one rainfall event was recorded. In September 2014 and early October 2014, prior to the second set of data collection, both rainfall stations recorded rainfall events >10 mm (Figure 9). Thereafter, between November 2014 and January 2015, both stations experienced a number of rainfall events >20 mm. However, in 2015, both Quaggasvlei and Dalveen received very little rainfall in February and the majority of the summer rain fell in late March 2015 (Figure 10). This occurred shortly after the third set of data collection, hence the drought conditions experienced during that period. Between March 2015 and April 2015 both stations received over 100 mm of rainfall, causing the high flows experienced in May 2015 during the fourth set of data collection. Unusually, Quaggasvlei and Dalveen both recorded high rainfall events over the period of June 2015 to July 2015 (typically dry months), with a number of lower rainfall days in between.

Discharge

Discharge data, shown in Figure 11, followed a similar trend in each site. Except for July 2014, Site 1 consistently had the lowest discharge of the three sites. Site 2 had the highest discharge for all three datasets in 2015, while Site 3 it had the highest discharge in July and October 2014. In addition to similarities in length and wetted channel, Site 1 and 3 also follow a similar pattern of changes in discharge, with their highest readings in July 2014, while Site 2 had its highest readings in August 2015. All three sites had their lowest discharge in March 2015 and readings then increased in all sites in May 2015 and August 2015.

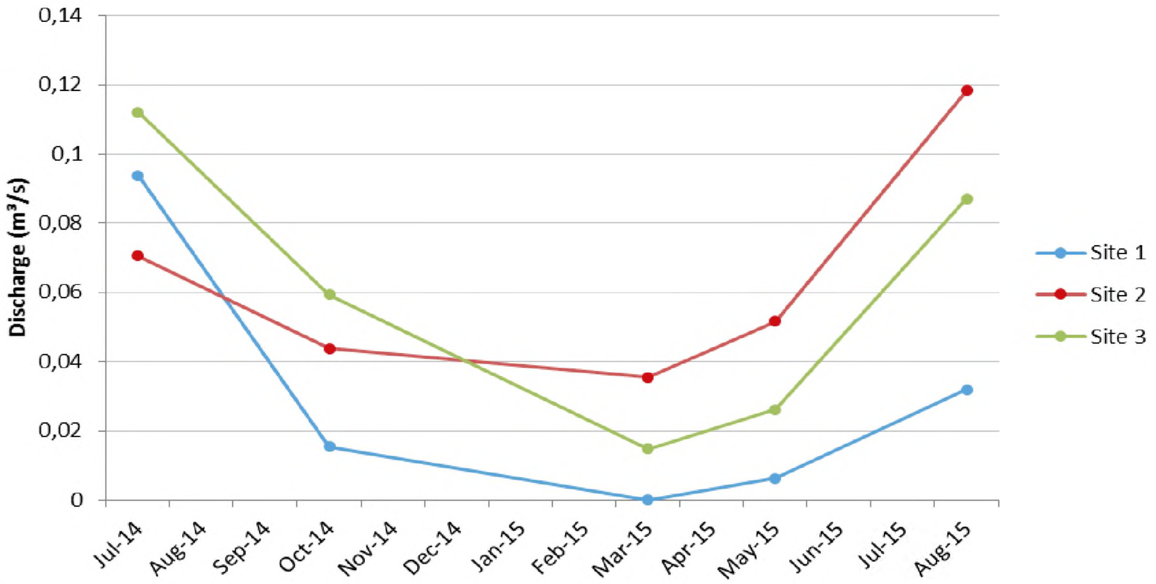
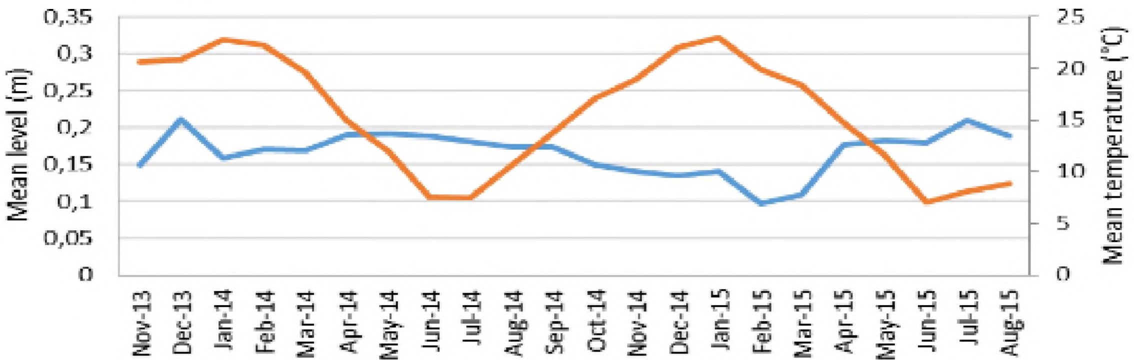


Figure 11: Discharge for all three sites for July 2014, October 2014, March 2015, May 2015 and August 2015

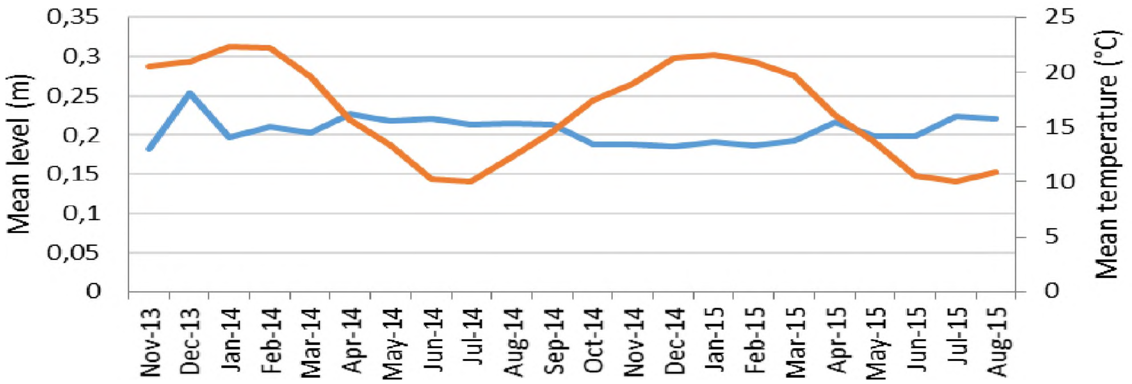
Patterns in discharge are largely a product of external influence on the river system. While flow in the Wilgerbos River is maintained by groundwater, Site 1 is directly below a water extraction point and a weir which reduces the amount of flow entering the site, and as a result is more severely affected by droughts. During the drought in March 2015, approximately 40% of Site 1 was dry, with only standing pools and shallow riffles remaining in the other 60 %. Site 2 and Site 3 are largely natural, and groundwater supply is uninterrupted, so the sites are able to continue flowing even when rainfall has failed. Groundwater flow enters the channel at the beginning of a dolerite intrusion above Site 2 (Steynberg, J.P, Pers. Comm. 2015). The lower flow in Site 3, relative to Site 2 is most likely due to transmission losses, evaporation and uptake by riparian vegetation. The general shallowness of the channel makes it particularly vulnerable to evaporation.

Mean monthly water level and temperature derived from the level logger data yielded largely similar results for the three sites (Figure 12 a – c). In general, data fluctuated predictably in seasonal cycles; temperature increased in the summer months and decreased in the winter months, and water level followed an opposite trend, decreasing in summer, when evaporation is higher, and increasing in winter, partly due to the out-of-season rainfall. However, as the Karoo normally receives the largest portion of its annual rainfall in the summer months (Rowntree & Foster, 2012), and is prone to high intensity thunderstorms, there are also peak flows evident in the level logger data caused by these events.

a) Site 1



b) Site 2



c) Site 3

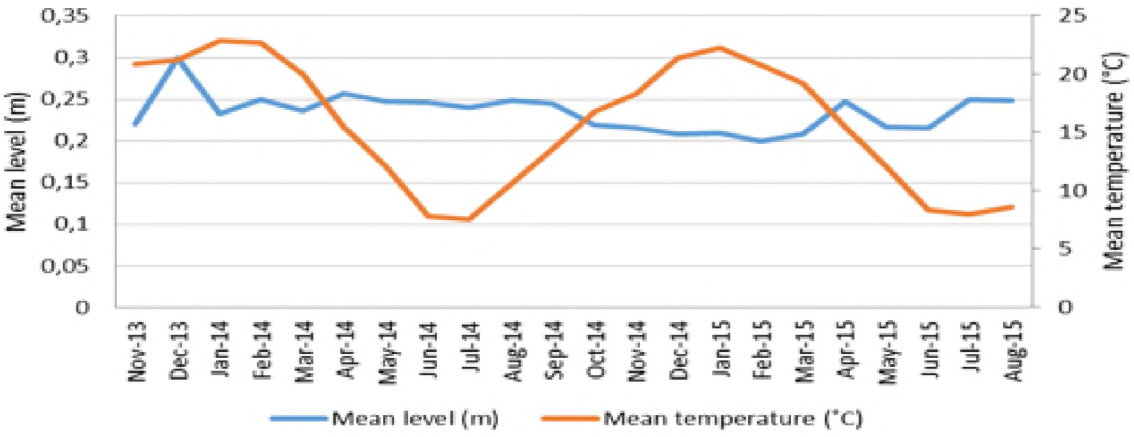


Figure 12 (a – c): Mean water level (m) and temperature (° C) for Site 1, Site 2 and Site 3, for the period of November 2013 to August 2015.

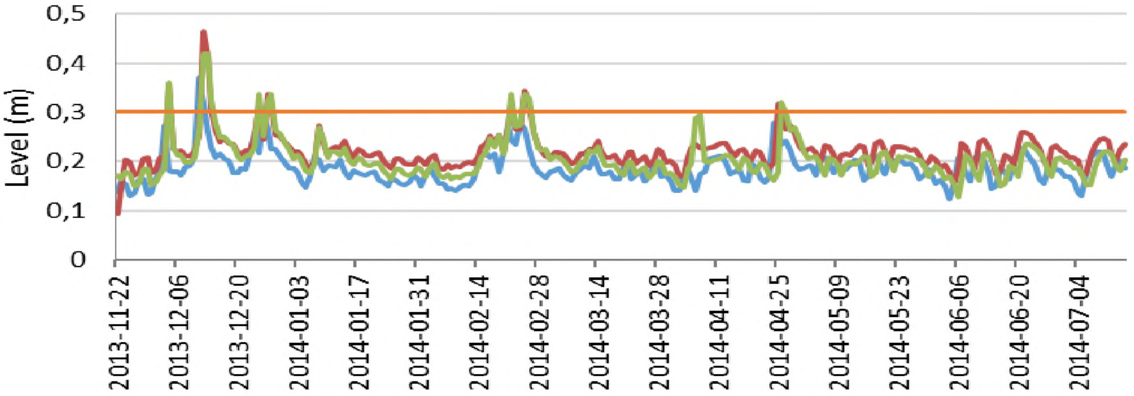
All three sites experienced such a peak in December 2013, with smaller peaks in the following months, a second large peak in April 2015 and a third in July 2015. The large peak in flow in December 2013 is clearly accounted for in the rainfall data, as the highest rainfall for the whole period fell in this month and high rainfall was also recorded in late March and April 2015, and during June and July 2015 (Figure 10). Input from snow melt may also have been a factor in

July 2015, as it was during the May 2015 and August 2015 data collection. The drought in March 2015 is reflected in the water level data, beginning with a sharp decline in September 2014, especially in Site 1. Furthermore, the charts also reflect the previously discussed effect of groundwater in Site 2 and 3, which experienced less severe decreases in flow than Site 1 which has restricted flow.

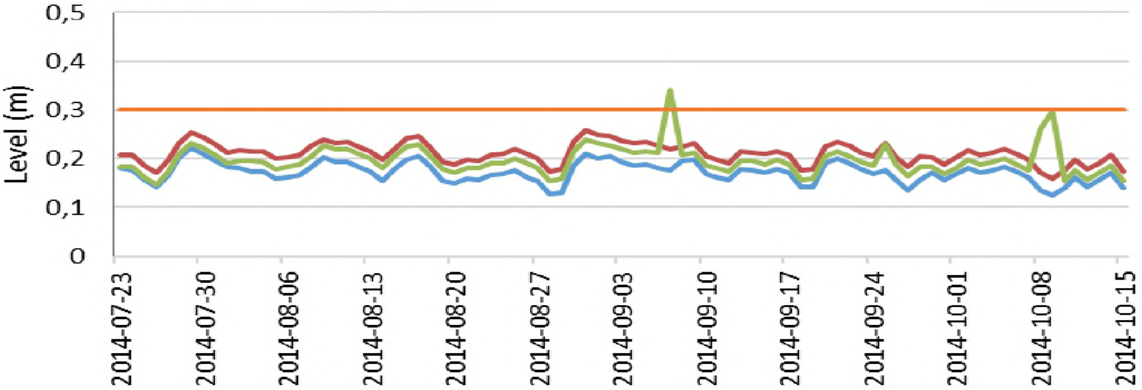
A closer study of data from the level logger shows that high flows in the system were typically caused by a number of flood events of varying size, rather than one large event. Daily flow graphs created for the periods between data collection explain the flows measured during each of the data collection periods (Figure 13a – e). A level of 0,3 m was chosen as the minimum level for flood peaks, as indicated on the graphs below. Average water levels in all three sites ranged between 0,12 m and 0,22 m, and each site experienced flows of up to 0,27 m on a fairly regular basis, making it unlikely that these were flood events. Peaks of 0,3 m and higher coincided with a number of large rainfall events, which can be seen in Figure 9 and Figure 10, suggesting that these were floods.

In most cases the three sites responded almost simultaneously to flood events. However, some events (seen in Figure 13 a and d) appear to have taken up to two days, after being measured in Site 1, to noticeably increase the water level in Site 2 and 3. Flood peaks in Site 1 were typically smaller than those seen in Site 2 and 3, most likely due to the lower initial water level. Between November 2013 and July 2014 (Figure 13a) seven flood peaks occurred: four between late November 2013 and January 2014, and three more widely spread between February 2014 and May 2014. Snowmelt is a likely contributing factor to flow in winter. The large flood peaks in December 2013 and February 2014 are a result of the high rainfall events as recorded at Quaggasvlei and Dalveen. The low discharge measured in Site 2 in July 2014 may have been due to groundwater depletion as a result of the low winter rainfall for that year.

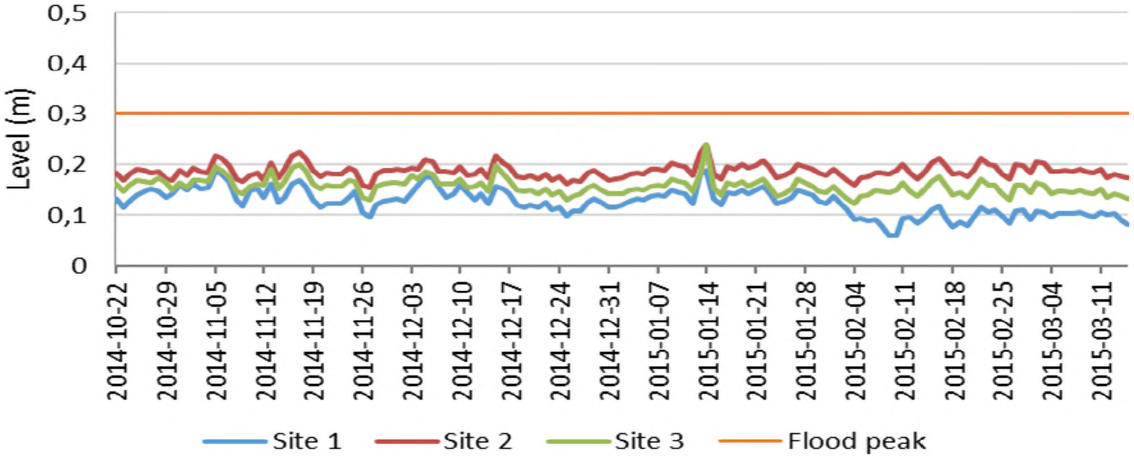
Similarly, the low rainfall measured in the second half of 2014 (Figure 13 b) also resulted in lower flow with far fewer, and smaller flood peaks between July 2014 and October 2014. The two flood peaks measured in Site 3, which did not occur in Site 1 and 2, were possibly a result of a localised storm above the tributary which feeds in between Site 2 and 3, causing an increase in flow only downstream of the confluence.



a) November 2013 - July 2014



b) July 2014 - October 2014



c) October 2014 - March 2015

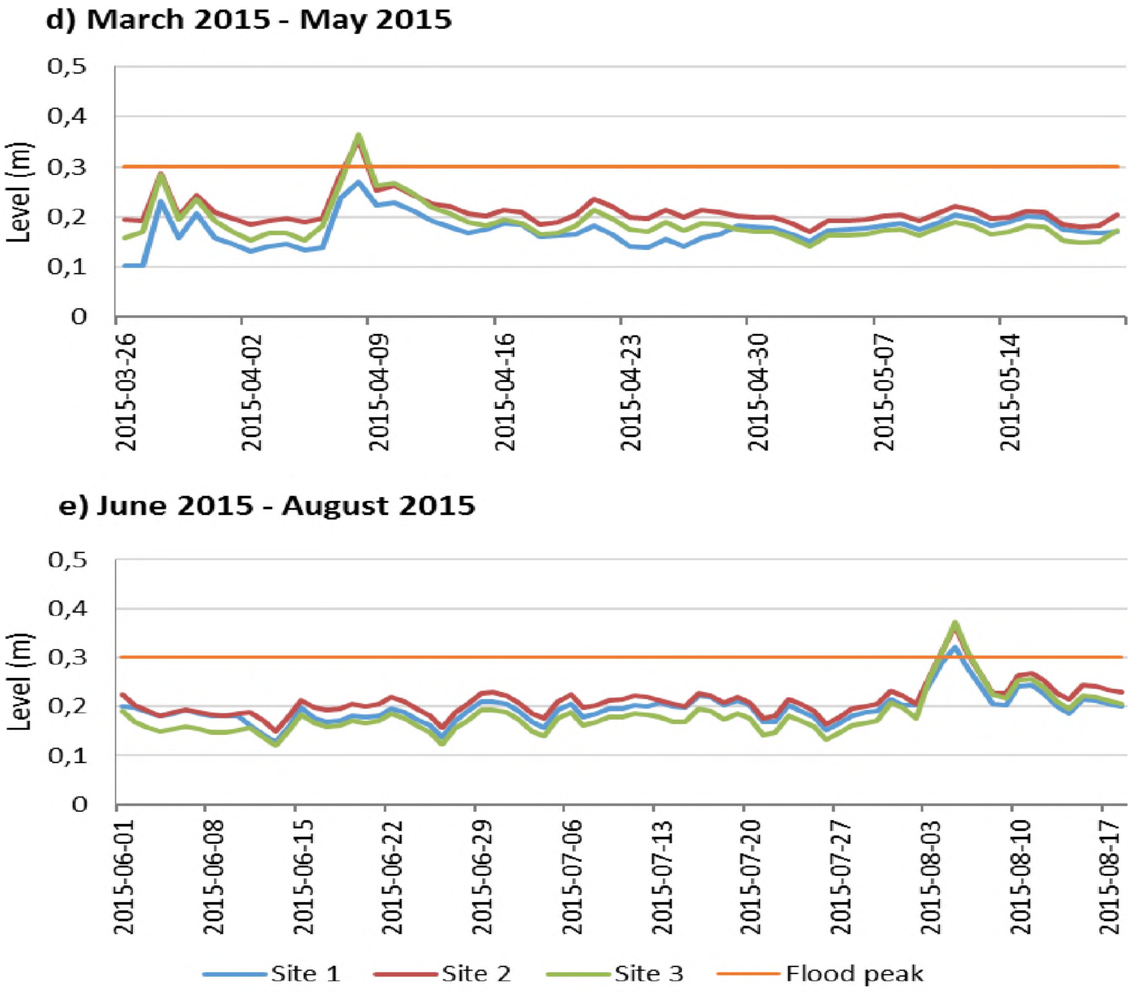


Figure 13 (a – e): Daily flow for each site for the periods between field data collection: a) November 2013 to July 2014, b) July 2014 to October 2014, c) October 2014 to March 2015, d) March 2015 to May 2015, and e) May 2015 to August 2015.

The drought in March 2015 is reflected in the data (Figure 13 c), as none of the sites experienced a flood peak, and water levels were consistently low and declined, particularly in Site 1, from February 2015 to March 2015. Again, the influence of an uninterrupted supply of groundwater can be seen in Site 2 and 3, as their water levels remained around 0,2 m, while the water level in Site 1 dropped below 0,1 m in mid-February 2015.

Thereafter, the increased rainfall in the form of large events in late March 2015 and early May 2015 (Figure 13 d) raised the water level and caused two peak events, but only the second event was large enough to cause a flood. After the second peak, the water levels stabilise; however, the graph shows that Site 1 experienced slightly higher flows than normal at the beginning of May 2015, and through June and July 2015. The first instance coincides with a set of high rainfall events in late April 2015, which may have created high surface flow as groundwater stores had already been recharged by the rainfall in late March 2015 and early April 2015.

Figure 13 e shows that flow in Site 1 decreases again over the course of May 2015 during which time no rain fell, but increases again throughout most of June and July 2015, sustained by out-of-season winter rainfall. The flood peak in early August 2015 follows a high rainfall event at the end of July 2015, following a period of approximately a month without rainfall, thus maintaining the high flow as measured during the final set of data collection in mid-August. This event was followed by a second smaller flood a week later.

4.3 Habitat quality: Flow depth and velocity

Velocity and depth measurements taken in each patch during biological sampling were used to create a hydromorphological index of diversity (HMID), comparing variation in flow with variation in depth across each site. Further analysis of velocity and depth heterogeneity produced quite clear-cut results, which show a degree of uniformity of flow among the sites over the course of the study. HMID scores for each site reflect the diversity of flow speed and depth of the water column, where a higher score indicates greater heterogeneity. As Table 10 shows, the scores did not vary greatly across the sites. Excluding the significant outlier at Site 1 in March 2015, the data produces an average HMID of 10,98, which suggests a fairly low level of heterogeneity overall. The high score for Site 1 in March 2015 can be attributed to the drought conditions as the combination of dry river bed, standing pools and flowing riffle patches created a high diversity of flow conditions in the site. Site 2 and 3 had the benefit of groundwater to maintain relatively stable flow conditions throughout, and so only experienced a slight increase in HMID scores.

Table 10: HMID scores per site for each data collection period

	Jul 2014	**Oct 2014**	**Mar 2015**	**May 2015**	**Aug 2015**
Site 1	9,24	8,81	38,87	18,21	12,26
Site 2	7,91	12,34	14,94	11,40	11,11
Site 3	6,21	8,96	12,36	10,93	9,02

In July 2014, all three sites had similarly low HMID scores, with Site 1 showing slightly higher flow diversity than the other two sites. In October, Site 2 has a notably higher HMID score. The cause of this is unclear, but it is possible that as flow began to reduce in all three sites the deep pool in Site 2 had a more pronounced impact on the heterogeneity of the site, thus raising the score. In March 2015, Site 1 has a significantly higher score than the other two sites. This can be easily explained by the drought conditions. While Site 1 maintained a higher HMID score in May 2015, all three sites experienced a decrease in flow heterogeneity, and a further drop in August 2015, as rainfall restored more stable flow conditions.

4.4 Habitat quality: Water chemistry

The trends in water quality variables (Table 12 a – c) are influenced by out-of-stream factors such as underlying geology, groundwater, and sediment inputs, as well as interactions between the instream variables. All three sites were shown to have good water quality and to be well within safe parameters for the protection of aquatic ecosystems, as specified by DWAF (1996), in the case of all the variables for the majority of the study period.

Table 11 a - c: Water quality variables for Site 1, Site 2 and Site 3, for July 2014, October 2014, March 2015, May 2015 and August 2015

a) Site 1	Nitrate (mg/l)	Soluble Phosphate (mg/l)	EC (µS/cm)	pH	Temp (°C)	DO (% saturation)	Turb. (ppm)
Jul-14	<2	<0,2	448	8,31	4,5	88,2	11,41
Oct-14	<2	0,5	530	8,62	15,2	95,6	8,93
Mar-15	<2	0,7	532	9,00	18,2	121,0	20,20
May-15	<2	0,2	632	7,62	10,4	92,1	3,11
Aug-15	<2	<0,2	419	8,14	9,6	72,5	2,07

b) Site 2	Nitrate (mg/l)	Soluble Phosphate (mg/l)	EC (µS/cm)	pH	Temp (°C)	DO (% saturation)	Turb (ppm)
Jul-14	<2	0,2	470	8,19	10,5	91,5	10,20
Oct-14	<2	0,5	572	8,66	14,1	90,1	9,38
Mar-15	<2	1,5	455	8,24	18,9	81,5	10,50
May-15	<2	0,5	651	7,95	10,9	96,2	3,23
Aug-15	<2	<0,2	466	8,19	10,2	76,2	2,33

c) Site 3	Nitrate (mg/l)	Soluble Phosphate (mg/l)	EC (µS/cm)	pH	Temp (°C)	DO (% saturation)	Turb (ppm)
Jul-14	<2	0,2	440	8,54	14,3	103,1	10,58
Oct-14	<2	0,3	547	8,92	20,2	108.0	10,12
Mar-15	<2	0,7	545	8,86	16,6	96,2	14,23
May-15	<2	0,2	594	8,39	11,7	92,6	2,97
Aug-15	<2	<0,2	451	8,44	11,2	83,9	2,27

Nitrate levels remained consistently low across all three sites for the duration of the study, always below the minimum detectable value of 2 mg/l. Research by DWAF (1996) states that groundwater can have very high concentrations of nitrates and that increases in dissolved oxygen usually result in increased nitrate levels, as ammonia and nitrites can be oxidised more efficiently. In well-oxygenated water, more than 80% of nitrogen should be present as nitrate (DWAF, 1996). However, despite groundwater inputs and even during periods of super oxygen saturation (> 100%), which occurred on three occasions, the nitrate levels in the study sites remained low. It is worth noting that nitrate is the most accessible form of inorganic nitrogen, and is easily taken up by instream vegetation and algae, which may be the cause of the consistently low nitrate levels recorded (DWAF, 1996).

Soluble phosphate levels also remained generally low, ranging between 0 (undetectable) and 0,7 mg/l in Site 1 and 3, and up to 1,5 mg/l in Site 2, but all three sites experienced a peak in concentration during the drought in March 2015. DWAF (1996) describe phosphates as an essential macronutrient which is actively taken up by vegetation and so short-term fluctuations are quickly regulated and therefore the increase seen in data is not likely to have had an impact on biotic health.

Overall EC levels range from 419 µS/cm to 651 µS/cm over the course of the study. Most streams in South Africa range from 30 µS/cm to over 1100 µS/cm, depending on factors such as amount of rainfall, underlying geology and evaporation rates (DWAF, 1996). As the study area falls within the Beaufort Group sedimentary rock formation (Schlüter, 2008), and is exposed to high annual evaporation, DWAF (1996) guidelines state that high concentrations of total dissolved salts are to be expected in the streams, which would cause the EC to be high (between 200 µS/cm to 1100 µS/cm). Guidelines state that EC fluctuations should not exceed 15% of the normal range (DWAF, 1996). However, given the lack of long term water quality data for this river, an average margin of change of 98 µS/cm was assumed from the range stated by DWAF (1996), producing a range of 552 µS/cm – 748 µS/cm. While all three sites experience EC levels below 552 µS/cm, none exceed the upper boundary.

Overall, Site 1 had the lowest water temperatures, and Site 3 had the highest. None of the sites showed unusually high or low temperatures, according to the water quality criteria laid out by DWAF (1996). Site 1 is shaded by a row of poplar trees along the bank, and therefore receives fewer hours of sunlight than the other two sites, whereas Site 2 and 3 are both exposed to direct sunlight from early in the morning and only become shaded in the late afternoon. The water temperature in Site 2 was likely moderated by the direct input of cooler groundwater, accounting for the lower temperatures in this site than in Site 3.

Water quality data show a weak correlation between water temperature and pH, with warmer water having a more alkaline pH, and colder water being slightly more acidic. However, throughout the course of the study the water pH at all three sites remained between 7 and 9. South African rivers can have a pH ranging between 4 and 11, typically within the limits of 6 and 8 for a pristine system, but fluctuations can occur on a daily basis in response to temperature changes, photosynthetic activity (increased alkalinity) or biotic respiration and decomposition (increased acidity) (DWAF, 1996). All three sites also experienced the lowest pH, that is the most neutral pH, in May 2015. This can be attributed to the increased volume of water in all three sites due to higher rainfall. As rain water tends to be slightly acidic, the high input of rain water into the stream system would counteract the alkaline groundwater flow and cause the stream to have a more neutral pH.

Site 1 and 3 did not reflect the expected correlation between temperature and DO content. Typically, cold water is able to store more oxygen than warm water (USGS, 2015), but only Site 2 reflected this trend. Site 1 and 3 experienced irregular DO concentration trends, including periods of super saturation (>100%). Site 1 had a high oxygen concentration in March 2015, during the drought. The expected reason for this is the large quantity of filamentous algae present within the channel at this site (Table 10) and the lack of flow, resulting in an over-production of oxygen within a confined area. Site 3 experienced super-saturation on two occasions when algal growth had been particularly successful, but good flow through the site prevented oxygen levels from reaching the very high level seen in Site 1 in March 2015. Low oxygen concentrations in all three sites in August 2015 may be attributed to a large quantity of dead algae, particularly in Site 1 and 2. The process of decomposition would have removed oxygen from the water, creating lower oxygen conditions.

All three sites showed two peaks in turbidity values: a small peak in July 2014 and a much larger peak in March 2015. Site 1 had the largest range of turbidity values, while Site 2 had the lowest range. In contrast to expected trends, turbidity was inversely related to flow levels. This could have been due to the presence of suspended organic material rather than to suspended sediment. Higher baseflows may have flushed out the organic matter without adding suspended sediment, thus causing a decrease in turbidity. As the data were collected through point measurements (in time) the highest flows, normally responsible for high suspended sediment concentrations, were not sampled. Following the end of the drought in March, a significant decrease in turbidity was measured in all three sites, when flow increased due to heavy rains in May 2015 and the contribution from snowmelt in August 2015. The low turbidity in Site 2 and 3 can also be attributed to groundwater inputs and the weir above Site 1 acts as a sediment trap, leading to the low turbidity in Site 1.

Baseflow turbidity levels in the study sites remain well within safe limits (<100 mg/l), according to DWAF (1996), and turbidity was therefore not expected to be a major contributor to changes in the biotic community; however, data presented in *Section 4.8* in this chapter suggests that turbidity had a significant relationship with the presence or absence of biota.

4.5 Habitat quality: Bed sediment

Bed sediment is important in habitat assessment as it reflects mineral matter and organics which have fallen out of suspension, i.e. are not currently being transported downstream. While turbidity provides a measure of immediate stream condition factors, bed sediment responds to processes over time, specifically recent changes in flow. Two methods were used to measure bed sediment in this study: embeddedness scores, and stored sediment quantities.

Embeddedness

Visual assessments of embeddedness (Figure 14 a – e) revealed that all three sites had varying degrees of embeddedness throughout the course of the study. The rating table adapted from Minshall (1983, cited in Sylte & Fischenich, 2002), was used to create two descriptive categories for dominant substrate. On the embeddedness scale, where 1 is the least embedded and 5 is the most, a score of 4 or 5 (50% - 100% embeddedness) was described as a sandy type patch, while a score of less than 4 was described as a rocky type patch. This was used for the MiniSASS sensitivity score calculations discussed in *Section 4.6.* None of the sites experienced high overall levels of embeddedness over the course of the study, with rocky and sandy patches often quite evenly distributed within the sites. This suggests good habitat availability, with a wide variety of substrate types, covering the full range of scores on Minshall's rating table. Site for site, there was no clear trend as all three sites experienced fluctuating embeddedness scores. Although flow must be a significant driver of embeddedness, the process is complex and unpredictable. Certain flow events had a visible influence on sediment dynamics, but no clear long term trends can be derived from the data.

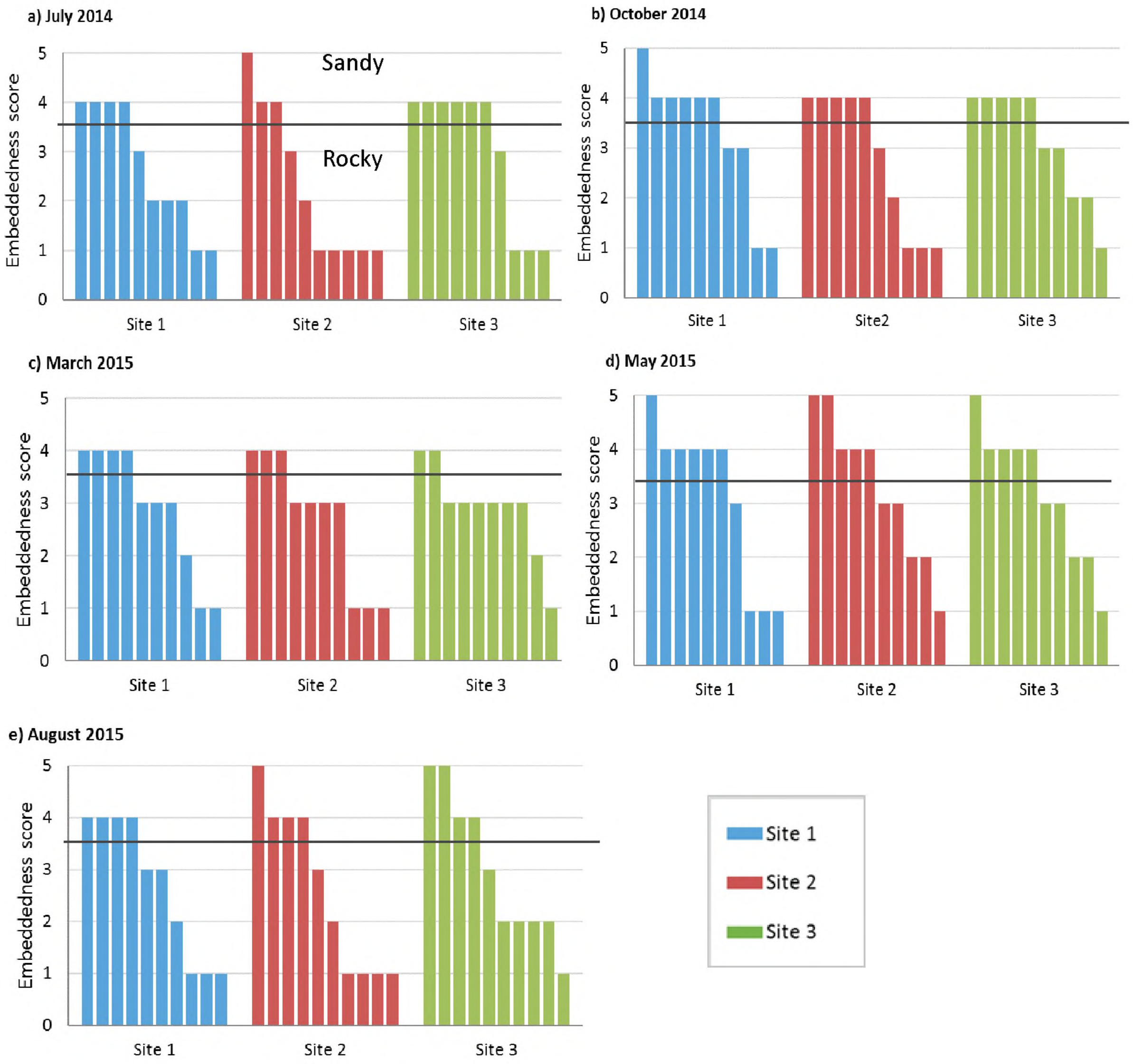


Figure 14 (a – e): Embeddedness score per patch for all three sites for July 2014, October 2014, March 2015, May 2015 and August 2015. Horizontal lines show 50 % embeddedness mark.

Overall, sandy and rocky type patches were fairly evenly distributed across the sites. However, rocky type patches were generally more common, as shown in Table 12.

Table 12: Number of sandy and rocky type patches per site, for July 2014, October 2014, March 2015, May 2015 and August 2015.

	Site 1		**Site 2**		**Site 3**		**Total**	
	Sandy	Rocky	Sandy	Rocky	Sandy	Rocky	Sandy	Rocky
Jul-14	4	6	3	7	6	4	13	17
Oct-14	6	4	5	5	5	5	16	14
Mar-15	4	6	3	7	2	8	9	21
May-15	6	4	5	5	5	5	16	14
Aug-15	4	6	4	6	4	6	12	18
Total	24	26	20	30	22	28	66	84

Lower embeddedness in July 2014 (Figure 14 a) is best accounted for by observing daily flow data prior to data collection (Figure 13 a). A number of small peaks in flow occurred between November 2013 and July 2014, which would not have been sufficient to wash sediment out of the weir above Site 1, but would have had a higher capacity to scour the bed, resulting in low overall embeddedness within the sites. The higher number of sandy patches in Site 3 may be due to the settling out of sediment transported from upstream into the low flow sections of the site.

Median embeddedness scores (Table 13) show that while all sites contained patches with a large range of embeddedness scores, patches in Site 1 and 3 overall had slightly higher embeddedness scores than Site 2. However, median scores were very similar, except for July 2014, when Site 2 contained a large number of patches with an embeddedness score of 1, while the majority of patches in Site 3 had a score of 4 or higher. There were no temporal trends and median scores fluctuate independently. This shows that while rocky and sandy type patches were quite evenly distributed in all three sites, the level of embeddedness of the patches was more variable across the sites.

Table 13: Median embeddedness scores and score range for all three sites for July 2014, October 2014, March 2015, May 2015 and August 2015.

	Site 1		**Site 2**		**Site 3**	
	Median	Range	Median	Range	Median	Range
Jul-14	2,5	3,0	1,5	4,0	4,0	3,0
Oct-14	4,0	4,0	3,5	3,0	3,5	3,0
Mar-15	3,0	3,0	3,0	3,0	3,0	3,0
May-15	4,0	4,0	3,5	4,0	3,5	4,0
Aug-15	3,0	3,0	2,5	4,0	2,5	4,0

All three sites showed an increase in the number of sandy patches from July 2014 to October 2014 (Table 13), and Site 1 and 2 showed an increase in embeddedness (Table 14). This period experienced little flow variability, and no floods, so the build-up of sediment is most likely a result of gradual deposition in low flow sections. Embeddedness decreased slightly in March 2015, particularly in Site 1 and 3. This is a direct result of the decrease in flow, as no sediment could be transported into Site 1 and, as groundwater is not a source of sediment, Site 2 and 3 were experiencing more scouring than deposition, creating more rocky patches than sandy patches.

May 2015 again shows high quantities of bed sediment, following large flood events in late March and early April. These events would have been large enough to overtop the weir above

Site1, introducing fresh sediment into the site, which could also be transported downstream. August 2015 experienced a slight decrease in embeddedness in all three sites, despite experiencing a significant flood early in the month. However, the smaller peaks in flow following the event may have scoured the bed, removing what sediment was introduced by the flood peak.

Fine sediment storage

Fine sediment storage (Figure 15) was calculated for each site based on the concentration of sediment in water samples taken from disturbed areas of river bed. Site for site, stored sediment shows no clear trend. Site 1 had a continuously declining amount of stored sediment from July 2014 to August 2015, while Site 2 and 3 both had a variable trend, with the highest quantities recorded in March 2015, during the drought.

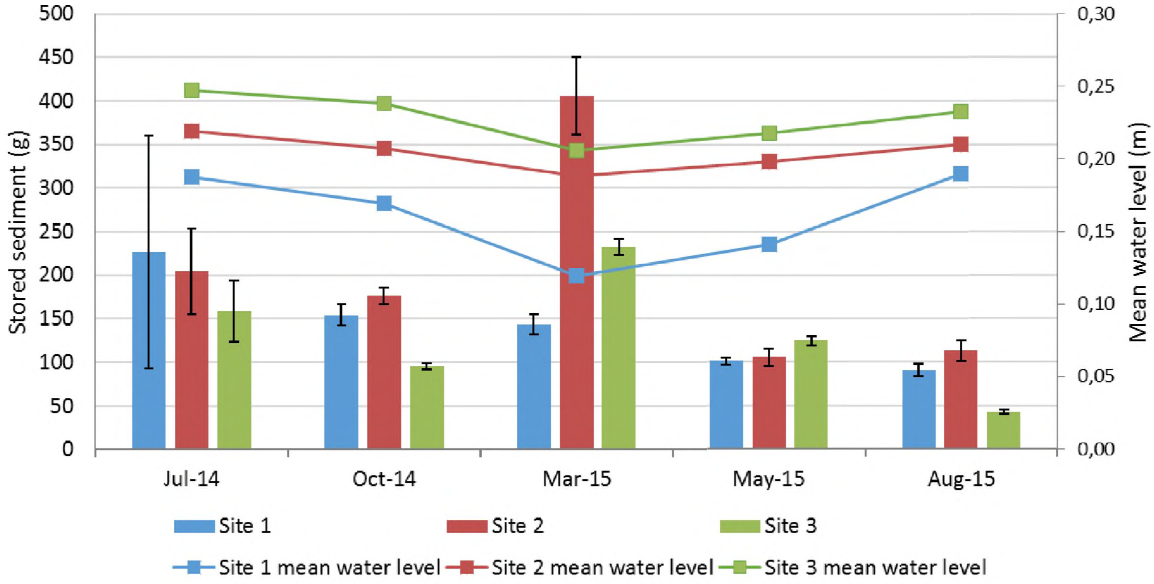


Figure 15: Stored sediment load for all three sites and mean water level for three months preceding sampling in July 2014, October 2014, March 2015, May 2015 and August 2015.

Surprisingly, the highest quantity of stored sediment for Site 1 was recorded in July 2014. This may be accounted for by the slight peak in water level that month, as well as the flood events in summer 2013/14. During the higher flow periods, more sediment was washed from the weir into Site 1, in contrast with March 2015 where the low flow was unable to transport sediment into the site. Furthermore, at Site 1 abstraction of water from the weir greatly reduces the flow downstream. Surface flow generated by rainfall on the land may also transport limited amounts of sediment into the channel from the banks. The low gradient of Site 1 results in low velocity flow, which has low transportation potential, allowing more fine sediment to settle on the channel bed. Site 2 and 3 receive water from tributaries, as well as surface flow, with limited impact from sediment traps or dams as the nearest weir is 3 km upstream of Site 2. While the rest of Site 2 and 3 has relatively little fine sediment in the channel bed, the low gradient sections at the top and bottom reaches of Site 2 and 3, and the deep pool in the middle of Site 2 act as natural sediment traps. Limitations of the stored sediment sampling technique make it far more suited to gravel and fine sediment dominated patches, and is therefore not reliable as a standalone measure of bed sediment in a stream with rocky patches. This will be discussed in greater detail in *Section 5.5* of the Discussion chapter.

While embeddedness assessments (Figure 14 a – e) and stored sediment measures (Figure 15) suggest that sediment quantities varied over time and that sandy and rocky type patches were both well represented by this study, a comparison of the two data sets (Figure 16) showed that there was no clear relationship between measured embeddedness and stored sediment, and in fact there was a weak negative correlation between the two variables in Site 1 and 3.

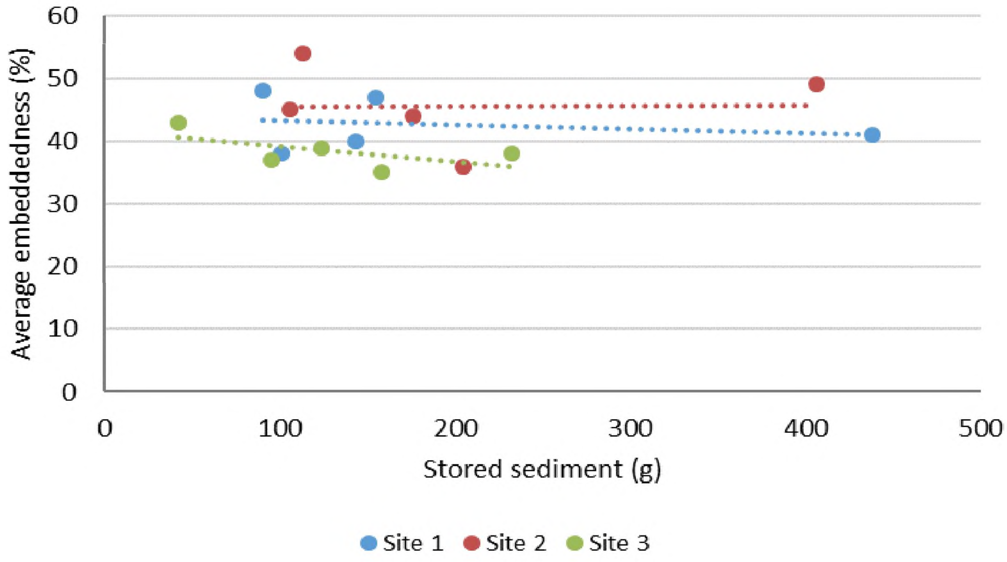


Figure 16: Comparison of average embeddedness (%) and stored sediment (g) per site.

The difference in sampling technique may account for this. Embeddedness is visually assessed based on the surface area of fine sediment within a patch, regardless of its depth. In contrast, measuring stored sediment is dependent on the depth of fine sediment, as samples can only be collected in areas with deep enough sediment to dig the bucket in to. Therefore, a patch with high embeddedness may produce a low amount of stored sediment because the fine substrate is relatively shallow. However, even in a shallow layer, the presence of fine sediment can have an impact on habitat availability, which in turn dictates biotic community structure.

4.6 Ecological response: MiniSASS sensitivity score analysis

Species sensitivity scoring, based on the MiniSASS method, was conducted first at a patch scale then at the site scale based on the accumulated data from each of the patches sampled. The embeddedness scores for each patch were used to create a measurable scale for the MiniSASS "sandy type" (embeddedness $\geq$ 4) and "rocky type" (embeddedness < 4) river categories, as discussed above. Sections of bedrock in this site could not be included in this aspect of the assessment as bedrock does not fit either of the specified patch types, and therefore could not be compared. Bedrock patches covered with other substrates, such as gravel or cobbles, were measured according to the top substrate layer, and were not categorised as bedrock patches.

Patch scale assessment

At a patch scale (Figure 17 a – e), sensitivity scores varied greatly within sites and between sites, regardless of substrate type. In July 2014, Site 1 and 3 showed a fairly even distribution of sandy type and rocky type patches, whereas rocky type patches were far more dominant in Site 2 (Table 13). Site 3 had the highest scores for sandy type patches, while Site 1 had the highest scores for rocky type patches. However, it must be noted that all three sites had similar average scores.

In October 2014, the distribution of sandy and rocky patches was even in Site 2 and 3, and sandy patches were slightly more prevalent in Site 1. Site 2 had the highest average sensitivity score for rocky type patches, and Site 3 had the highest scores for sandy type patches (Figure 17 b).

In March 2015, rocky type patches made up the majority of Site 2 and 3. The highest average scores for sandy type patches were in Site 2. Site 1 was difficult to sample in March 2015 as many of the patches did not contain enough water to support biota, but a number of high sensitivity organisms were found in all three sites during this period.

In May 2015, Site 2 and 3 showed almost equal distribution of sandy and rocky type patches, but Site 1 contained more sandy type patches. Site 1 had the highest average sensitivity score for both sandy type and rocky type patches.

In August 2015, rocky patches were more common in all three sites. Site 1 had the highest average rocky type patch score, and Site 1 and 2 had similar sandy type patch scores.

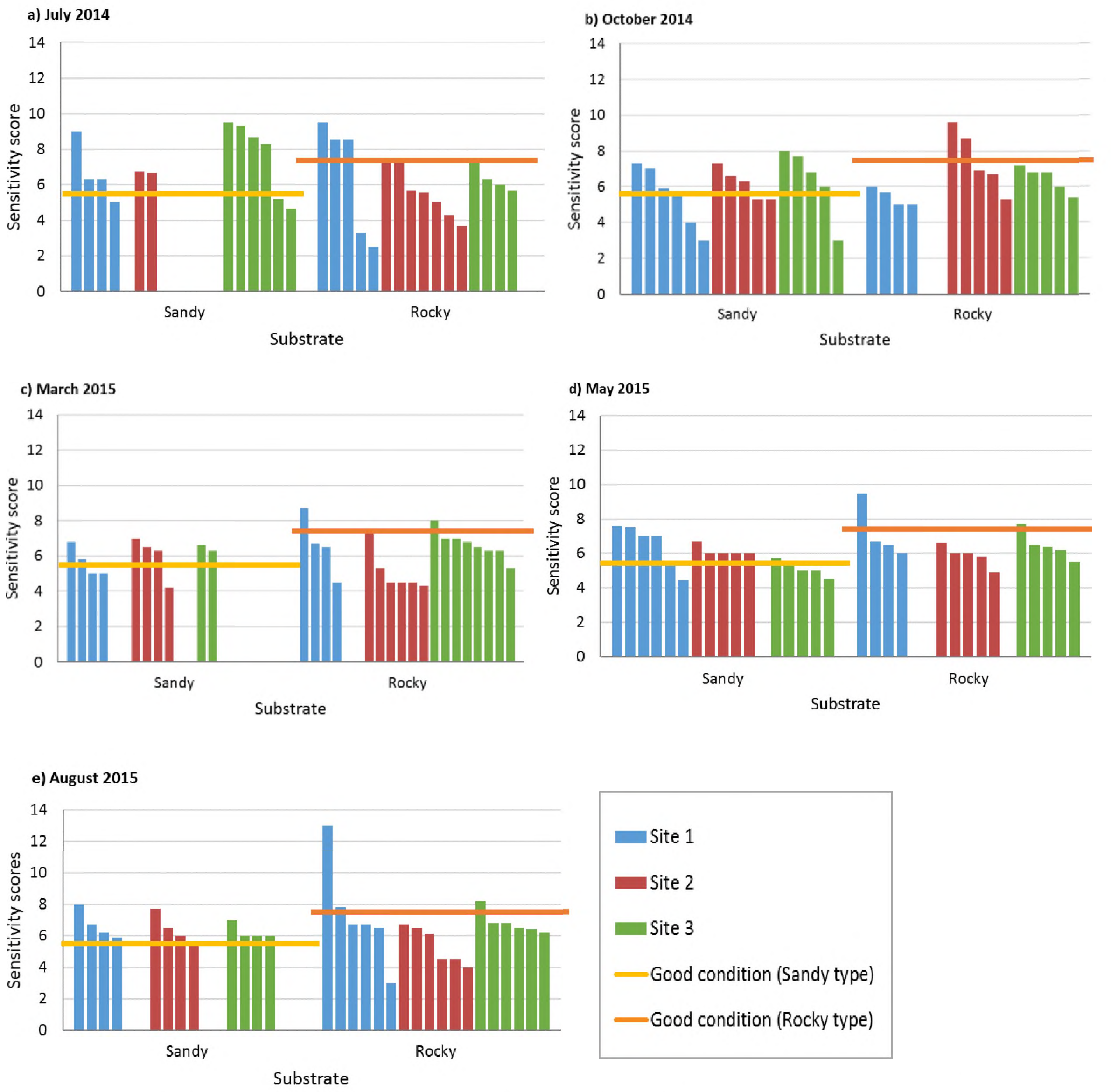


Figure 17 (a – e): Sensitivity scores by patch type for all three sites for July 2014, October 2014, March 2015, May 2015 and August 2015. Horizontal lines show minimum score required for good condition category in sandy and rocky type patches.

May 2015 (Figure 17 d) had the highest number of "good" condition patches, as well as a relatively good diversity of patch types. The observed presence of stoneflies and mayflies in May 2015 and August 2015 is indicative of good water quality, while the number of "good" condition patches is also indicative of overall good water quality and habitat condition as a result of increased flow. The sensitivity score suggests that Site 2 was in poorer condition than the others. In this case, the abundance of low sensitivity organisms, such as damsel flies and worms, produced a lower average score for the site, despite high overall biotic diversity and the presence of sensitive organisms such as other mayflies and caddisflies

Chi-square analyses (Table 14 a – c) were conducted to assess dependence of the sensitivity scores derived from the MiniSASS methodology on three variables: sample date, site, and bed material type (sandy, rocky, or combination). A significance level of $p < 0,05$ was used for all tests. The data showed no significant relationship between sensitivity scores and sample date, a weak relationship with site, and a strongly significant relationship with substrate type.

Table 14 a - c: Chi-square dependency analysis for patch-scale sensitivity score against sample date, site and substrate type

a) Date	**Natural**	**Good**	**Fair**	**Poor**	**Very poor**	**Total**	**X^2 Total**
Jul-14	8	7	3	4	6	28	4,51
Oct-14	7	9	4	5	5	30	2,39
Mar-15	3	10	7	2	6	28	2,59
May-15	5	7	11	6	1	30	7,12
Aug-15	5	11	10	0	4	30	5,17
Total	28	44	35	17	22	146	**21,77**
Critical value							**26,30**

b) Substrate	**Natural**	**Good**	**Fair**	**Poor**	**Very poor**	**Total**	**X^2 Total**
Sandy	18	28	13	3	4	66	12,47
Rocky	10	16	22	14	18	80	10,29
Total	28	44	35	17	22	146	**22,77**
Critical value							**9,49**

c) Site	**Natural**	**Good**	**Fair**	**Poor**	**Very poor**	**Total**	**X^2 Total**
Site 1	14	10	12	3	8	47	5,27
Site 2	5	17	8	7	12	49	6,77
Site 3	9	17	15	7	2	50	5,35
Total	28	44	35	17	22	146	**17,38**
Critical value							**15,51**

Sandy-type patches most commonly scored “good” condition, and rocky-type patches most commonly scored “fair” condition. This is somewhat unexpected, because higher substrate heterogeneity generally supports greater species diversity, and mayflies, the second most sensitive taxonomic group, favour rocky habitats. However, a number of groups with low sensitivity scores, such as flatworms, damselflies and snails also favour rocky habitats and their presence will have lowered the average score for patches in which they were found. Furthermore, the score bracket for "good" condition in rocky patches is higher than that for sandy type patches, meaning that more patches can qualify as "good" condition in sandy substrates than in rocky substrates, given the same biotic community composition. As shown in Table 14, rocky patches were slightly more dominant overall in the study, especially in March 2015. Sandy patches were more common in October 2014 and May 2015. However, sandy and rocky type patches were fairly evenly distributed spatially and temporally. While sensitivity scores did not prove to be dependent on time of year or site, individual taxonomic groups showed clear preferences for certain sites and times of year, as will be discussed in *Section 4.7.*

Site scale assessment

Sensitivity scores were calculated at a site scale (Figure 18 a – e), using the biotic data from each patch type to derive an average sandy type and rocky type score for each site, as well as an overall score. The sampling method used in this study was more spatially focussed than the standard kick-and-sweep method used in MiniSASS sampling, producing patch-specific results rather than the more general reach scale results produced by MiniSASS. Overall, the health of the Wilgerbos River was "fair" according to the MiniSASS scoring system, despite a number of sites scoring highly for one or both substrate types.

In general, sensitivity scores fluctuated for all three sites over the course of the study and there is no consistent pattern. Site 1 had the best overall sensitivity score in May 2015 and August 2015. Site 2 had the highest overall score in October 2014, and Site 3 the highest overall score in July 2014 and March 2015. Sensitivity scores were particularly low for Site 1 and Site 2 in March 2015, and for Site 3 in May 2015. Given that rocky type patches were more common over the course of the study (Table 13), the Wilgerbos River would be classified as a rocky type river bed. In terms of overall scores, Site 3 scored the "good condition in July 2014, Site 2 in October 2014 and Site 1 in May 2015 and August 2015. None of the sites scored the "good" condition in March 2015. This again reflects the "fair" condition score for the river.

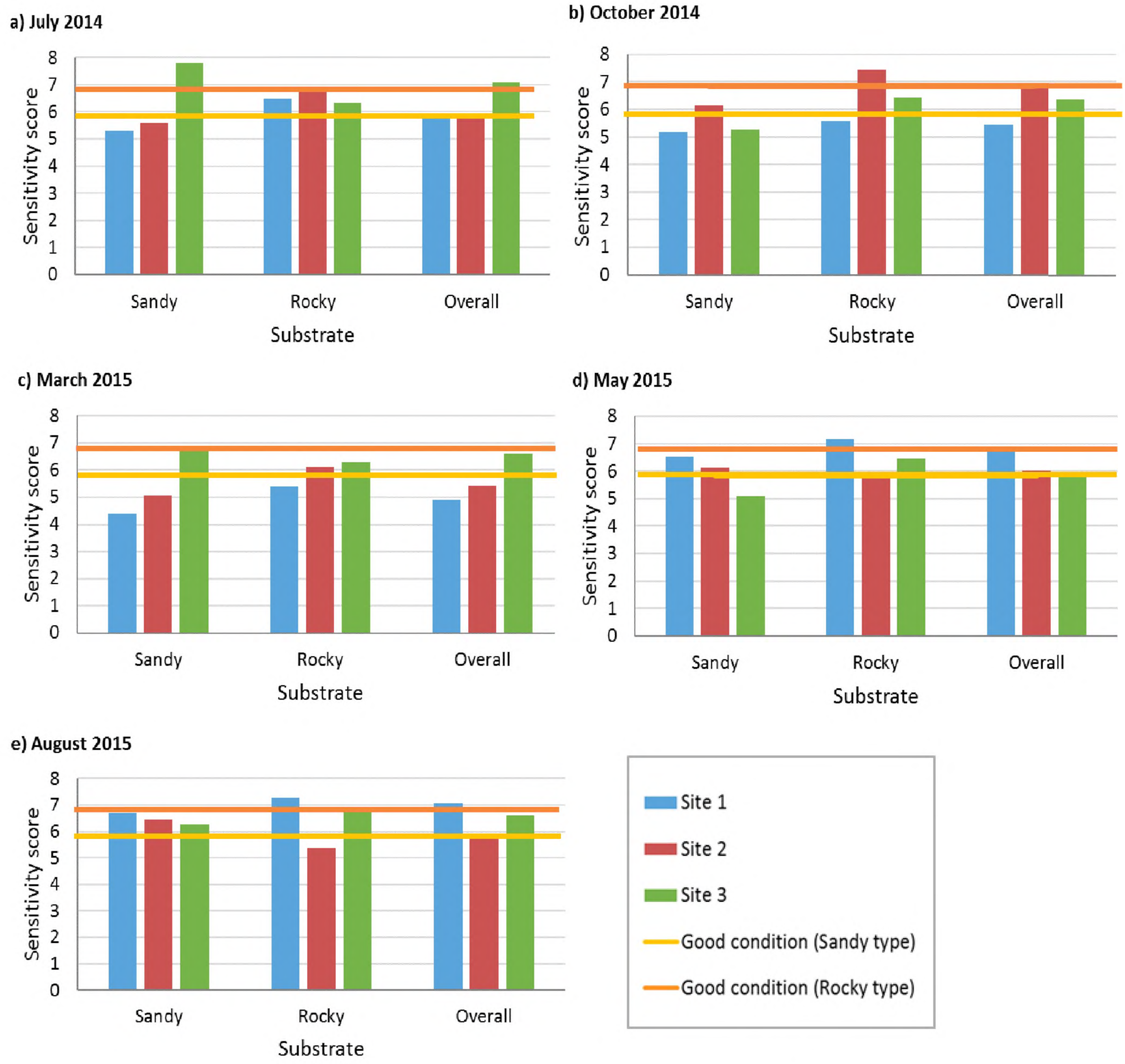


Figure 18(a – e): Sensitivity scores by substrate and overall score for all three sites for July 2014, October 2014, March 2015, May 2015 and August 2015. Horizontal lines show minimum score required for good condition category in sandy and rocky type rivers.

4.7 Biotic community analysis: Spatial and temporal variability in biotic communities

The data produced for the sensitivity scores should be considered in relation to the abundance of each taxonomic group in order to more accurately interpret the data. As MiniSASS scoring is primarily focussed on the sensitivity of individual taxonomic groups to changes in water quality, it lacks a measure of abundance or biotic diversity. Therefore, the sensitivity scores do not provide any information on the biotic community composition, which is an important consideration for habitat assessment.

Frequency of occurrence

Further analysis of biotic diversity and abundance was conducted through analysis of the frequency of occurrence per taxonomic group (Figure 19 a – e).

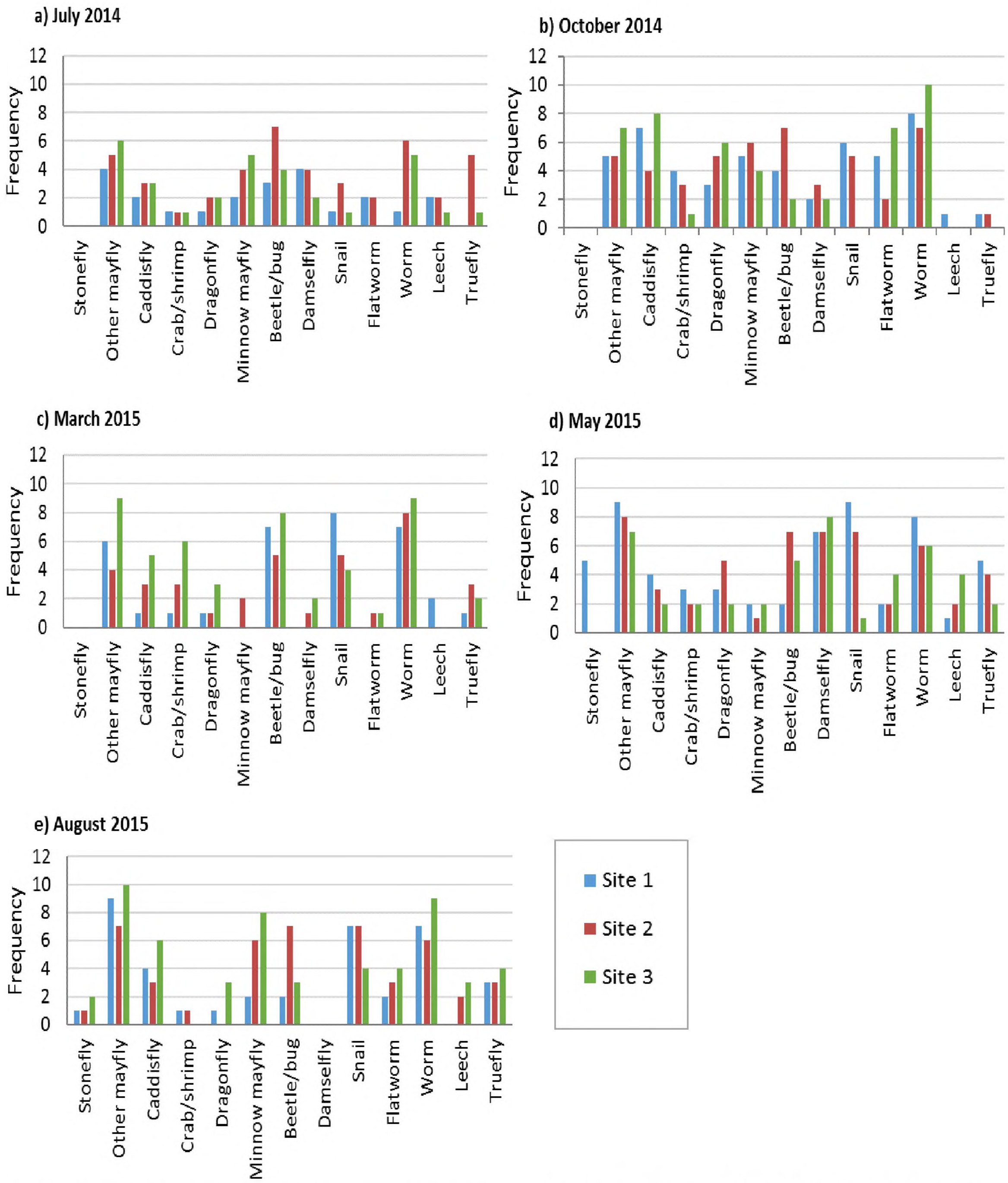


Figure 19 (a – e): Frequency of occurrence for each taxonomic group in all three sites for July 2014, October 2014, March 2015, May 2015 and August 2015.

All three sites contained diverse biotic communities, where each taxonomic group is quite well represented. The other mayfly group, the second most sensitive on the MiniSASS scale, was common in all three sites throughout the course of the study, and other sensitive groups, such as caddisflies, dragonflies, and minnow mayflies were also common.

However, there is a mismatch between abundance scores and sensitivity scores. Low sensitivity groups were also common across the three sites, and often their presence negatively skewed the sensitivity scores. Conversely, a limited number of groups, but with high sensitivity scores, can positively skew the score for the site.

Although sensitivity scores (Figure 18 a – e) suggested that Site 1 was the healthiest of the three in July 2014, abundance data shows that in fact it contained fewer of almost every taxonomic group compared to the other two sites. While in October 2014, Site 1 scored a low sensitivity score, the site also contained high numbers of sensitive organisms, such as mayflies and caddisflies. Conversely, Site 2, which had the highest sensitivity scores for sandy and rocky substrate, and a "good" overall sensitivity score, had low abundance scores for most groups.

Abundance data for March 2015 corresponds fairly well with the sensitivity scores. Both show that Site 1 had unexpectedly good results overall despite being harshly affected by the drought, and Site 2 scored surprisingly poorly, with low sensitivity scores and fairly low abundance of most groups. Site 3 scored highly in the sensitivity scores, which is reflected by high abundance of a number of groups.

In May 2015, abundance data supports the high sensitivity scores for Site 1, but somewhat contradicts the overall low scores for Site 2 and 3. Figure 19 d shows that 12 of the 13 taxonomic groups were present in both sites, but because low sensitivity groups were more abundant in general, their sensitivity scores were negatively skewed.

Finally, abundance data for August 2015 shows very different scores in all three sites. Site 1 had low abundance of a number of groups, but scored a high sensitivity score, while Site 3 had a high overall sensitivity score and had the highest abundance of the majority of taxonomic groups, including high sensitivity organisms. Site 2 had low abundance of a number of groups, which correlates somewhat with the low sensitivity scores for sandy type patches, but not with the overall "good" sensitivity score for the site.

The presence of a diverse collection of organisms, including sensitive groups like mayflies and stoneflies, shows healthy habitat patches with a wide variety of microhabitats to support a complex community. Taxonomic groups which favour fine sediment habitats include worms, dragonflies, bugs and beetles, and trueflies (see Table 6). Groups which favour rocky habitats include flatworms, crabs, minnow mayflies, other mayflies, damselflies, caddisflies and snails. Stoneflies and leeches were categorised as non-selective, as they were found in a variety of substrate types, with no clear preference. Over the course of the study all 13 taxonomic groups were found in all three sites. While some biota were fairly spatially and temporally ubiquitous, such as the other mayfly group, others varied greatly in their abundance over time (such as stoneflies) and between sites (such as the bug/beetles group).

Redundancy analysis

Analysis of the correlation between sampling dates and taxonomic groups using RDA (Figure 20 and Table 15) produced significant relationships (p = 0,002) for all dates except August 2015. The tri-plot (Figure 20) shows the changes in community composition across all three sites in each of the data collection periods. The presence or absence of organisms in each period was recorded and plotted as data points, alongside the time periods which are represented by arrows.

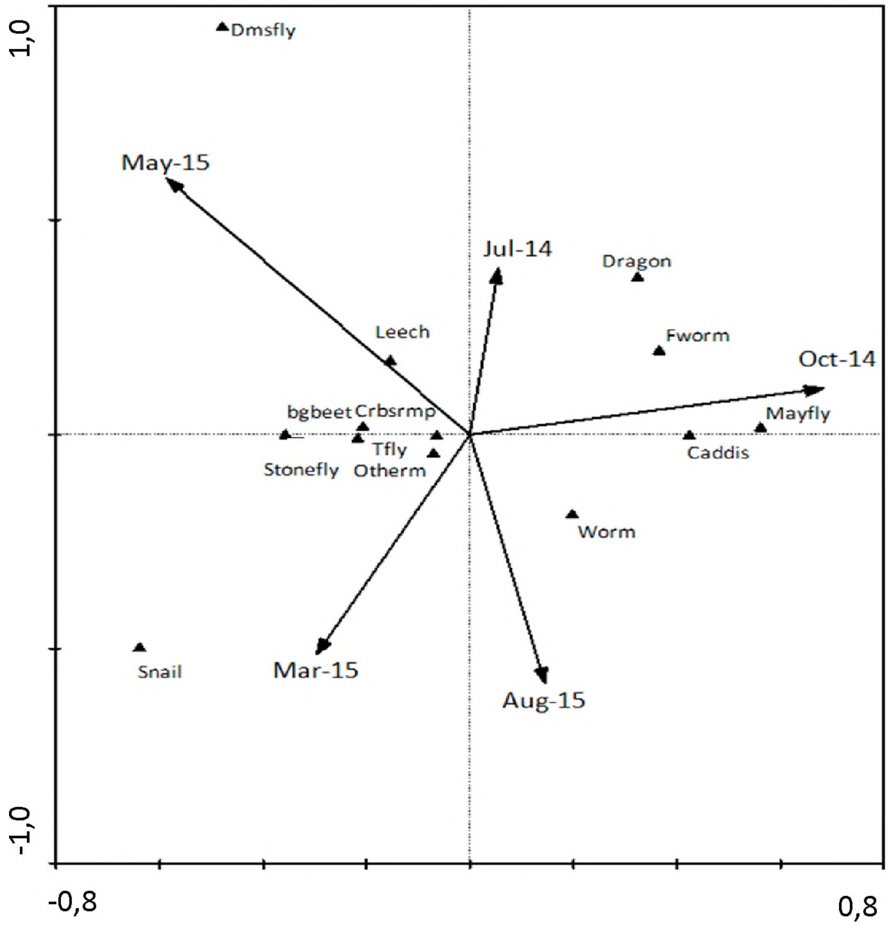


Figure 20: Total prevalence of each taxonomic group during each data collection period, for all three sites. (First axis variance = 34.8%, second axis variance = 27,3%)

The proximity of the data points to the arrows and their position in relation to the centre of the triplot indicates positive or negative correlation. Data points falling within 90° of an arrow are considered positively correlated with the associated period, a 90° angle indicates zero correlation, and points falling between 90° and 270° from the arrow are considered negatively correlated. The distance of a data point from the centre of the triplot indicates the strength of the correlation, strength increasing with distance.

Damselflies (*Dmsfly*) were far more common in May 2015 than any other period, and were entirely absent in August 2015. This creates a strong positive correlation with the arrow for May 2015, and a negative correlation with August 2015. The snail group was low in abundance in July 2014 (a negative correlation) but was highly prevalent in March 2015, May 2015 and August 2015 (a positive correlation). Notably, the bug/beetle group (*bgbeet*), crab/shrimp group (*Crbsrmp*), trueflies (*Tfly*), and other mayflies (*Otherm*) are closely clustered on the chart, which shows that they shared similar trends in prevalence during the study periods. All four groups experienced their lowest overall abundance in July 2014, and high overall abundance in March 2015 and May 2015. Other mayflies were present throughout the study, showing no preference for date, hence the closeness of the point to the centre of the tri-plot (0,0). Stoneflies were only present in May 2015 and August 2015; hence this data point is situated between these two arrows on the chart. This is somewhat misleading, as the chart seems to show that stoneflies were also present in March 2015, and so it is necessary to supplement the RDA with other forms of analysis.

Table 15 presents the numerical data from Figure 20. Of particular importance is the Conditional Effects section, which reflects the statistical value or importance of each of the variables. As explained by ter Braak and Šmilauer (2002), LambdaA reflects the power of each variable to explain the variance in biotic community. It considers each variable individually and indicates how well it can account for the observed variance, i.e. how much does each variable contribute to the variance shown in the tri-plot? Low LambdaA scores, as seen in Table 15, indicate that the variables do not have enough explanatory power on their own to account for the level of variance, and so it is a combination of all the variables, or other variables not shown in the tri-plot, which has caused variance in the biotic community composition.

As mentioned previously, the P-value refers to the significance of the correlation between each variable and changes in biotic community composition, where $p < 0,05$ indicates a statistically significant correlation. The F-value is the result of an F-test to assess the power of the RDA against the null-hypothesis (Frost, 2016). In this case, the null hypothesis states that one variable affects community composition, and that a simple intercept-only model would account for variance as sufficiently as the RDA. The higher the F-value, the greater the power against the null hypothesis. The P-value is therefore also a measure of the strength of the F-value, where p < 0,05 indicates that the F-value is high enough to reject the null-hypothesis (Frost, 2016). In the case of Table 15, all the sample periods produced statistically significant correlations, indicating that the null hypothesis can be rejected for all variables. This compliments the low LambdaA values, showing that biotic community variance is a factor of multiple variables and cannot be sufficiently explained by each variable in isolation.

Table 15: Eigenvalues, species-environment correlations, cumulative percentage variance or species data and species-environment relation and statistical significance (P) of total prevalence of each taxonomic group during each data collection period, for all three sites (refer to Figure 20).

Axes	**1**	**2**	**3**	**4**	**Total variance**
Eigenvalues	0,04	0,03	0,02	0,02	1,00
Species-environment correlations	0,61	0,53	0,38	0,42	
Cumulative % variance of species data		3,5	6,2	8,4	10,0
Cumulative % variance of species-environment relation		34,8	62,1	83,5	100,0
Conditional Effects					
Variable	Var.N	Lambda A	P	F	
May-15	24	0,03	0,002	4,77	
Oct-14	22	0,03	0,002	3,81	
Jul-14	21	0,02	0,002	3,90	
Mar-15	23	0,02	0,002	3,20	

Data comparing the abundance of taxonomic groups in each site (Figure 21 and Table 16) shows significant differences between the sites in terms of community composition. Certain taxonomic groups showed clear preferences for one or two sites, such as the bug/beetle group (*bgbeet*) which shows a strong correlation with Site 2, and the snail group which correlates with Site 1 and 2. Other groups showed slight preferences, such as stoneflies which were slightly more common in Site 1, trueflies (*Tfly*) in Site 2, and other mayflies (*Otherm*) in Site 3.

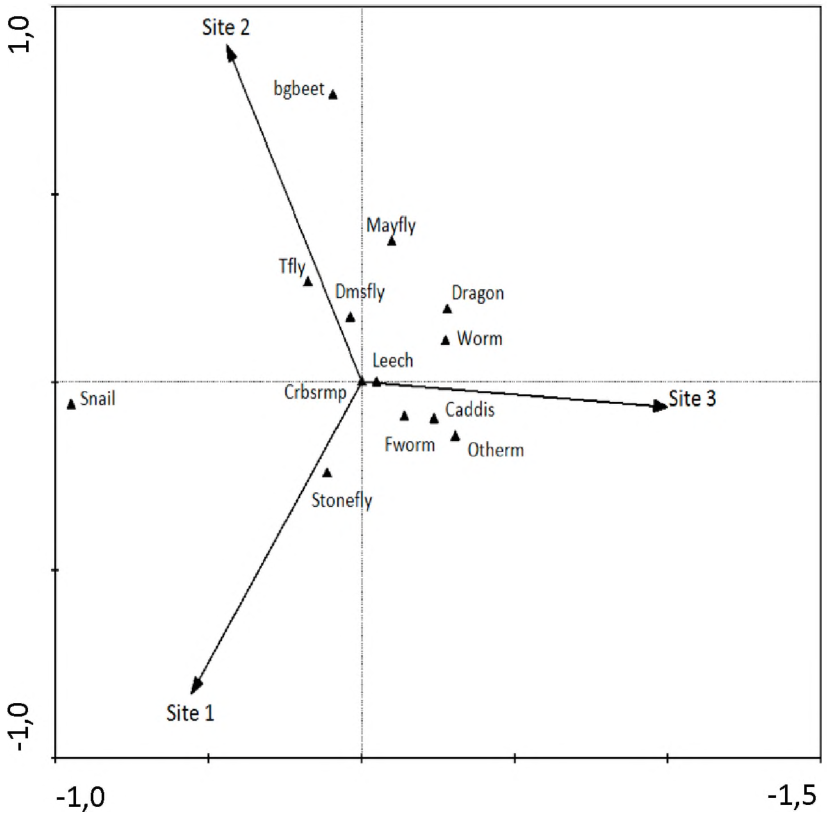


Figure 21: Total prevalence of taxonomic groups with respect to each of the three sites, for all data collection periods. (First axis variance = 57,1%, second axis variance = 42,9%)

Overall, Site 3 had the highest prevalence of a number of taxonomic groups, and a strongly significant relationship with presence and absence (p = 0,002), suggesting that these groups had a preference for habitat conditions in this site. However, for the most part, groups were fairly cosmopolitan. The crab/shrimp group (*Crbsrmp*) and the leech group were almost evenly distributed across all three sites, hence the closeness of these two points to the origin of the bi-plot (0,0).

Table 16 shows that the study sites produced low LambdaA values, indicating that individually they do not have sufficient power to explain the observed biotic variance. The low values also indicate that variables not assessed in this RDA likely account for biotic variance more strongly than physical location. However, both Site 1 and Site 3 produced statistically significant P-values, indicating that the null hypothesis can be rejected for both.

Table 16: Eigenvalues, species-environment correlations, cumulative percentage variance or species data and species-environment relation and statistical significance (P) of total prevalence of taxonomic groups with respect to each of the three sites, for all data collection periods (refer to figure 21).

Axes	**1**	**2**	**3**	**4**	**Total variance**
Eigenvalues	0,020	0,015	0,179	0,115	1,00
Species-environment correlations	0,486	0,397	0,000	0,000	
Cumulative % variance of species data		2,0	3,5	21,4	32,9
Cumulative % variance of species-environment relation		57,1	100,0	0,0	0,0
Conditional Effects					
Variable	Var.N	Lambda A	P	F	
Site 3	28	0,02	0,002	2,98	
Site 1	26	0,01	0,012	2,27	

4.8 Influence of flow properties on biotic community structure

Redundancy analysis

Data for velocity and depth measured in each patch as part of the MiniSASS data collection process was assessed using RDA in CANOCO (Figure 22). In Figure 22, Caddisflies (*Caddis*) show a strong preference for shallow water, along with flatworms (*Fworm*), other mayflies (*Otherm*), worms and minnow mayflies (*Mayfly*). This is indicated in Figure 22 by the position of these points in the opposite direction from the arrow representing the depth variable. Snails and the bug/beetle group (*bgbeet*) show a preference for medium depth water (150 mm – 250 mm) and are therefore more positively correlated with this variable in the triplot. However, at the time of data collection no organisms showed a preference for deep water (>250 mm).

Table 17 shows that only depth had a significant correlation with biotic diversity (p = 0,018). While both variables produced low LambdaA values, velocity had a nonsignificant correlation with biotic community (p = 0,122), indicating that the F-value was too low to confidently reject the null hypothesis. Further, it indicates that velocity cannot be considered a major driver of biotic variance. Leeches, trueflies (*Tfly*), dragonflies (*Dragon*), damselflies (*Dmsfly*), stoneflies and the crab/shrimp group were largely unselective regarding depth or velocity and were found in a variety of flow conditions.

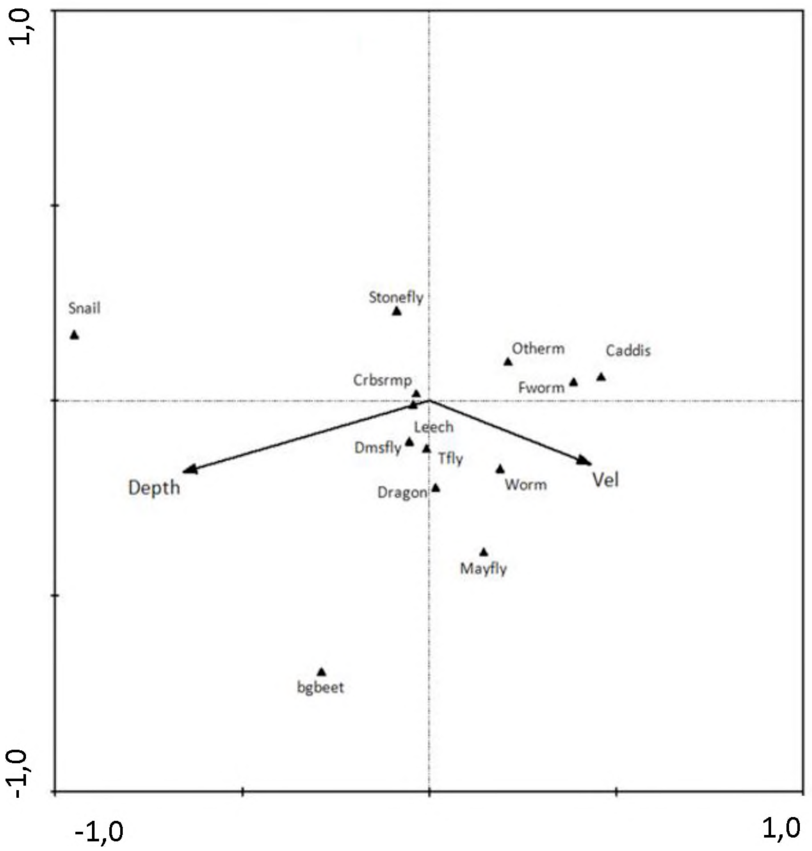


Figure 22: Total prevalence of each taxonomic group in relation to water column depth and flow velocity, for all data collection periods. (First axis variance = 75,4%, second axis variance = 24,6%).

Table 17: Eigenvalues, species-environment correlations, cumulative percentage variance or species data and species-environment relation and statistical significance (P) of total prevalence of each taxonomic group in relation to water column depth and flow velocity, for all data collection periods (refer to Figure 22).

Axes	**1**	**2**	**3**	**4**	**Total variance**
Eigenvalues	0,019	0,006	0,177	0,112	1,000
Species-environment correlations	0,422	0,292	0,000	0,000	
Cumulative % variance of species data		1,9	2,5	20,3	31,5
Cumulative % variance of species-environment relation		75,4	100,0	0,0	0,0
Conditional Effects					
Variable	Var.N	Lambda A	P	F	
Depth	10	0,02	0,018	2,30	
Velocity	9	0,01	0,122	1,51	

Although velocity did not have a significant relationship with taxonomic groups, it is worth noting that when sampling was conducted bugs/beetles, snails and stoneflies showed a very strong preference for low flow speeds, while caddisflies, flatworms and worms showed a preference for medium (0,1 m/s – 0,19 m/s) to fast (0,2 m/s – 0,3 m/s) flowing water.

Hydromorphological index of diversity

HMID scores were compared with the number of taxonomic groups found in each site to assess the impact of flow heterogeneity on biotic community composition in the Wilgerbos River. It is important to note here that while the MiniSASS datasheet depicts 13 primary taxonomic groups, Figure 23 includes a larger number of taxonomic groups, as subgroups such as beetle larvae and cased caddisflies were distinguished from the primary groups due to differences in flow property preferences.

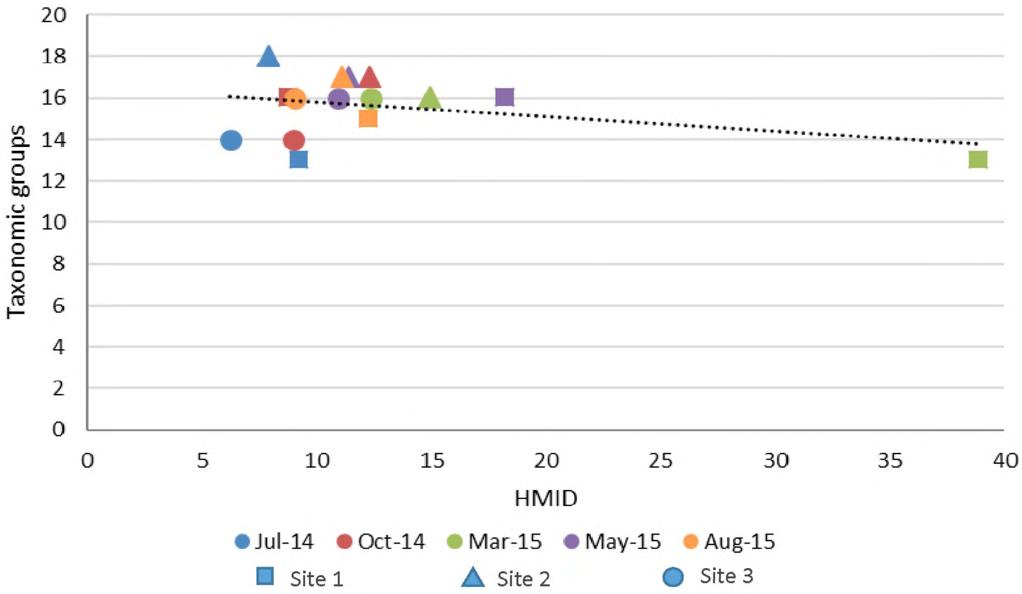


Figure 23: Comparison of Hydromorphological Index of Diversity scores with number of taxonomic groups found in each site for July 2014, October 2014, March 2015, May 2015 and August 2015. Dotted line shows R^2 = 0,01.

The HMID scores for each month produced a cluster pattern along the vertical axis. This reflects the similarity of the scores in each site over the course of the study, with the majority of the data sets falling within an HMID score bracket of 10 to 13, as was shown in Table 10. The number of taxonomic groups is similar in each data set, generally between 15 and 17 unique taxonomic groups in each site. Overall, Figure 23 suggests that while intra-site heterogeneity was variable, the difference between the sites was often lower than within the sites. As Figure 23 shows, there is no statistically significant relationship between the number of taxonomic groups and flow heterogeneity. However, a high HMID score can have a negative impact on biotic diversity. This suggests that high flow heterogeneity is not inherently a positive driver of habitat quality, which agrees with the findings of Gostner (2012) that showed greater heterogeneity places stress on biotic communities and limits their presence in a stream.

As HMID scores are calculated at a site scale, it was considered valuable to compare them with site-scale sensitivity scores, which were calculated using the same score table as was used for patch scale scores. However, comparing sensitivity scores per site with HMID scores created a very similar graph to that comparing HMID score with number of taxonomic groups (Figure 24), and produced an insignificant correlation ($R^2 = 0{,}01$). The data is, again, tightly clustered along the vertical axis with a distinct outlier created by Site 1 in March 2015. The trend line illustrates the similarity in biotic community among the three sites, as the sensitivity scores only range between 5,5 and 6,7.

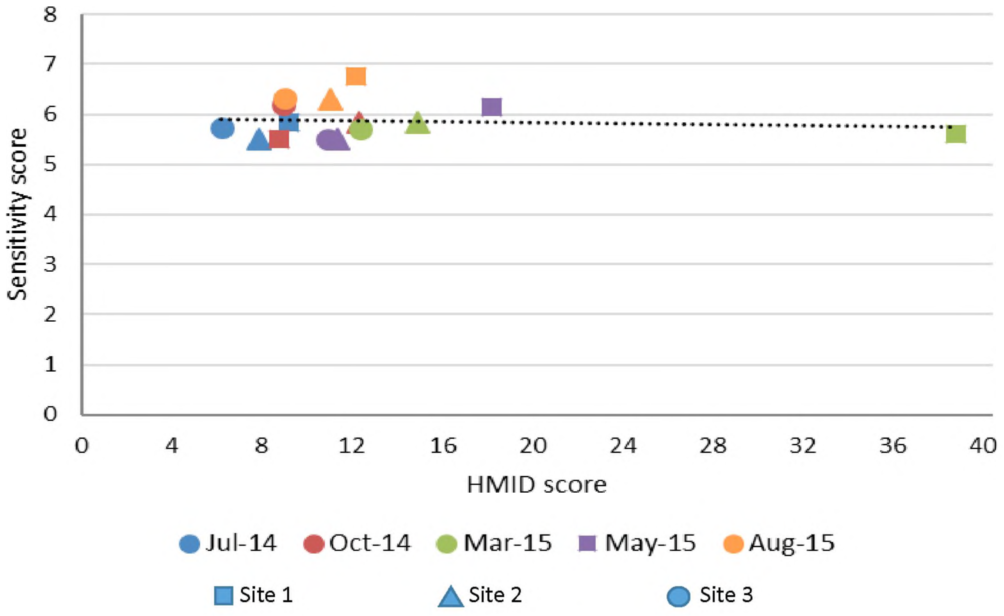


Figure 24: Comparison of Hydromorphological Index of Diversity scores with sensitivity scores for all three sites for July 2014, October 2014, March 2015, May 2015, August 2015. Dotted line shows R^2= 0,01.

4.9 Influence of water chemistry variables on biotic community structure

When correlating taxonomic groups with water quality variables using RDA (Figure 25), nitrate values were omitted from the dataset as they remained constant throughout the study. DO was the only variable not found to have a significant relationship with biotic diversity.

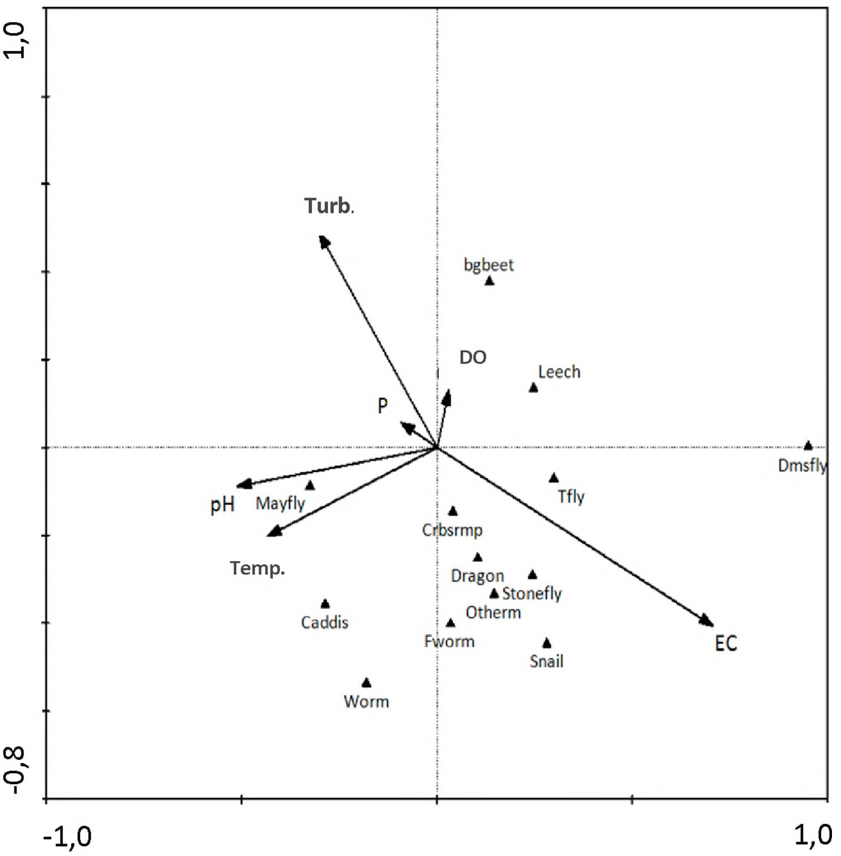


Figure 25: Total prevalence of each taxonomic group with respect to water quality variables, for all data collection periods. (First axis variance = 34,8%, second axis variance = 26,5%).

Electrical conductivity (*EC*) and phosphate (*P*) were the most significant (p = 0,002), followed by water temperature (*Temp*) (p = 0,006), turbidity (*Turb*) (p = 0,01) and then pH (p = 0,02). It is important to note that phosphate and DO both have very short arrows in the figure. Longer arrows indicate a stronger correlation with biotic variability, so short arrows indicate that biotic response is weakly correlated to phosphate and DO, with limited variation, i.e. all groups decreasing in abundance as phosphate and DO level increased.

Drought conditions in March 2015 drove changes in many water quality variables, as well as changes in flow and substrate. This makes drawing conclusions about relationships between biotic community structure and water chemistry difficult for this period as variables are not reflective of baseline conditions.

A number of organisms were closely associated with EC, as indicated by the clustering of points around this arrow. Interestingly, high biotic abundance in May 2015 correlates with high EC values in all three sites. Given that EC values are variable in the Wilgerbos River, it is likely that biota have a naturally high tolerance for dissolved salt concentrations (DWAF, 2006), allowing them to thrive in water with higher EC values. Damselflies (*Dmsfly*) are one of the biotic groups with higher abundance scores in May 2015, but their presence and absence data seems to be strongly linked to seasons (higher in winter, lower in summer), so the position of the point for this group may more clearly reflect the negative correlation with water temperatures, rather than a correlation with EC. Similarly, in the case of leeches, during sampling this group also showed a preference for lower water temperatures, so the position of this point is also more likely to reflect a negative correlation with water temperature than a correlation with either EC or DO.

Minnow mayflies (*Mayfly*) showed a strong correlation with pH, but a visual comparison of abundance data (Figure 19 a – e) and water chemistry data (Table 11 a – c) shows that the group was only present within a small range of pH scores (7,62 – 8,92). This suggests that the group has a narrow tolerance for pH levels and is therefore strongly influenced by this environmental variable. Caddisflies (*Caddis*) and worms show a positive correlation with water temperature (*Temp*). Figure 19 a – e shows caddisflies and worms to both be far more abundant in October 2014, when water temperatures ranged between 14,1 °C and 20,2 °C, than in other months when water temperatures were lower.

While the bug/beetle group (*Bgbeet*) is shown to be closely associated with DO, abundance and water chemistry data do not indicate correlation between this group and any of the water chemistry variables measured. This suggests that another, unmeasured variable is responsible for the presence or absence of this group, which either coincides with or is also correlated to changes in DO.

Table 18 further explains the observed relationships. All the variables produced low LambdaA values, which indicates that water chemistry cannot sufficiently explain biotic variance in isolation, and that other factors must be considered in the analysis. The nonsignificant P-value for DO ($p = 0,682$) reflects the low F-value (0,78) which indicates that DO does not significantly correlate to biotic variance, and is therefore not a major driver of community composition.

Table 18: Eigenvalues, species-environment correlations, cumulative percentage variance or species data and species-environment relation and statistical significance (P) of total prevalence of each taxonomic group with respect to water quality variables, for all data collection periods (refer to Figure 15).

Axes	**1**	**2**	**3**	**4**	**Total variance**
Eigenvalues	0,034	0,026	0,021	0,009	1,000
Species-environment correlations	0,597	0,430	0,478	0,287	
Cumulative % variance of species data		3,4	5,9	8,1	9,0
Cumulative % variance of species-environment relation		34,8	61,3	83,4	92,8
Conditional Effects					
Variable	Var.N	Lambda A	P	F	
EC	5	0,03	0,002	4,01	
P	2	0,02	0,002	3,08	
Temp	1	0,01	0,006	2,73	
Turb	4	0,02	0,010	2,45	
pH	6	0,01	0,020	1,98	
DO	3	0,01	0,682	0,78	

4.10 Influence of fine sediment on biotic community structure

Site scale redundancy analysis

Analysis of the influence of substrate on biotic communities included an RDA of the relationship between taxonomic groups and stored sediment per site, and a patch scale statistical analysis of taxonomic groups and embeddedness.

The site scale RDA (Figure 26) shows the changes in abundance of taxonomic groups within a site, in comparison with the quantity of stored sediment measured. Figure 26 shows that only the bug/beetle group (*bgbeet*) had a clearly positive correlation with fine sediment. This means that the prevalence of the bug/beetle group increased in sites and periods when stored sediment was higher. Dragonflies (*Dragon*) were also positively associated with fine sediment, but the correlation is near-zero, as the data point is almost at a right angle to the arrow. Similarly, worms also show a near-zero correlation. All other groups showed a negative correlation with fine sediment, showing decreasing prevalence with increasing fine sediment. Damselflies

(*Dmsfly*) and caddisflies (*Caddis*) showed the most strongly negative correlation, as these points are the furthest in the opposite direction from the arrow.

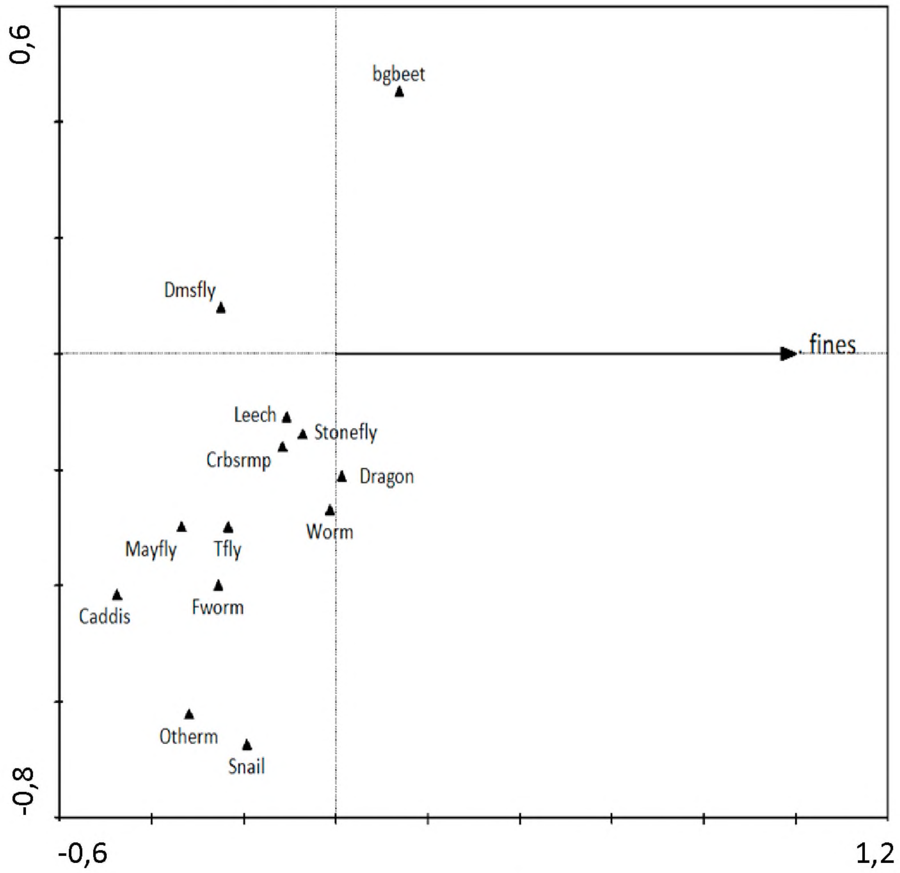


Figure 26: Total prevalence of taxonomic groups in relation to stored fine sediment quantity across all sites, for all data collection periods. (First axis variance = 100%).

Table19 shows that stored sediment had a statistically significant relationship with taxonomic groups (p = 0,002), but, as Figure 26 shows, the correlation was largely negative. While the LambdaA value for fine sediment is low, it is one of the highest values recorded for any variable. This suggests that, though fine sediment cannot fully explain biotic variance in isolation, it holds more explanatory power than many other variables assessed, and is therefore an important variable for biotic community composition.

Table 19: Eigenvalues, species-environment correlations, cumulative percentage variance or species data and species-environment relation and statistical significance (P) of total prevalence of taxonomic groups in relation to stored fine sediment quantity across all sites, for all data collection periods (refer to Figure 26).

Axes	**1**	**2**	**3**	**4**	**Total variance**
Eigenvalues	0,055	0,135	0,116	0,102	1,000
Species-environment correlations	0,564	0,000	0,000	0,000	
Cumulative % variance of species data		5,5	18,9	30,5	40,8
Cumulative % variance of species-environment relation		100,0	0,0	0,0	0,0
Conditional Effects					
Variable	Var.N	Lambda A	P	F	
Fines	13	0,05	0,002	8,54	

Fine sediment storage

A second comparison of the abundance of taxonomic groups per site with stored sediment was conducted (Figure 27 a – e) where groups were separated by habitat preference, into three categories, (refer to Table 6): rocky habitat groups, fine sediment habitat groups and non-selective groups, and then plotted against stored sediment per site. None of the data produced statistically significant correlations (as shown in Table 20) and therefore cannot be considered viable for constructing a profile of sediment preferences for each taxonomic group. However, the data may still be used to compliment further research into the relationship between taxonomic groups and fine sediment dynamics.

Rocky habitat groups had the highest prevalence throughout the study, even when stored sediment levels were high. This is partly a factor of the larger number of groups which favour this habitat type. In March 2015, when stored sediment was at its highest in Site 2 and 3, rocky habitat groups and fine sediment groups were almost equally abundant in all three sites. However, in July 2014 when stored sediment was at its highest in Site 1, rocky habitat taxa were strongly dominant, so no clear trend can be derived from this data.

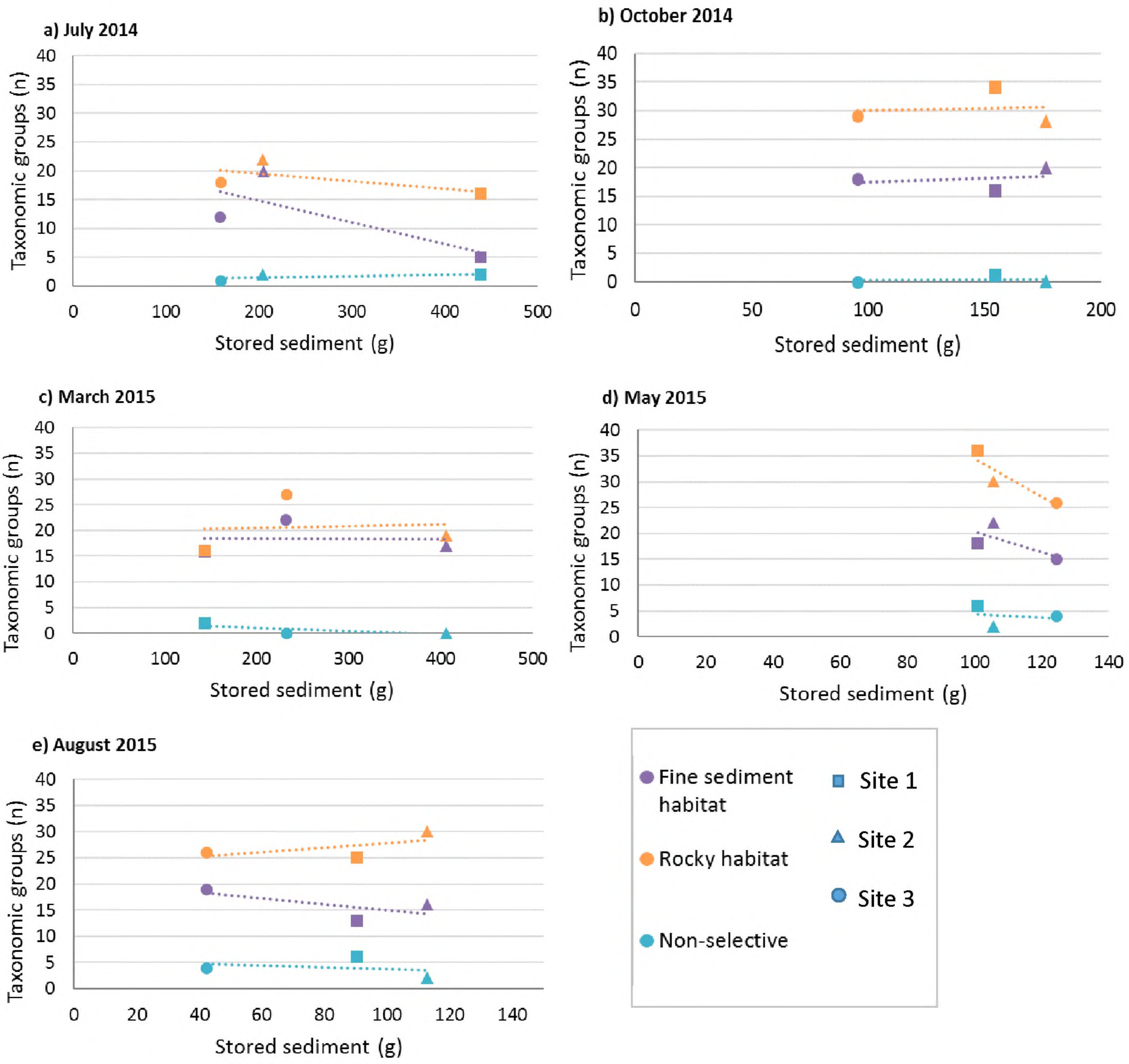


Figure 27 (a – e): Comparison of stored sediment quantities with abundance of fine sediment habitat groups, rocky habitat groups and non-selective groups per site for July 2014, October 2014, March 2015, May 2015 and August 2015.

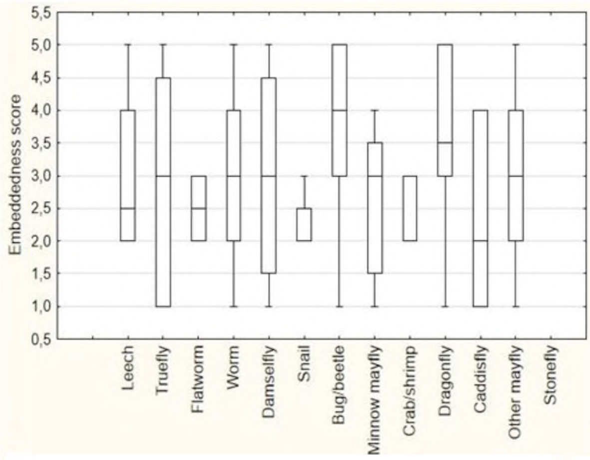
Table 20: R^2 values for each habitat preference group calculated from Figure 27 (a – e).

	July 2014	**October 2014**	**March 2015**	**May 2015**	**August 2015**
Rocky habitat	0,42	0,01	0,01	0,08	0,37
Fine sediment habitat	0,57	0,07	0,00	0,49	0,44
Non-selective	0,39	0,07	0,58	0,04	0,10

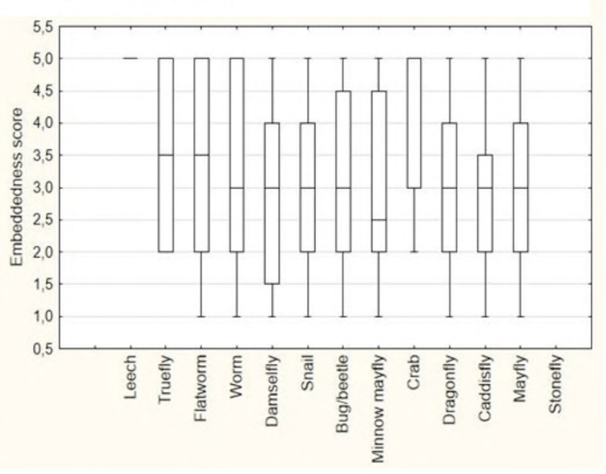
Embeddedness

A patch scale analysis of the relationship between embeddedness and taxonomic groups is presented in Figure 28 a – e. The box and whisker plots show the range of embeddedness scores across all the patches in which each group was found, as well as the median embeddedness score for all patches.

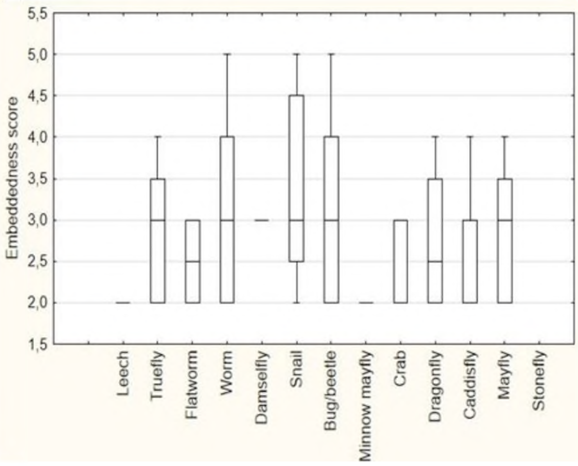
a) July 2014



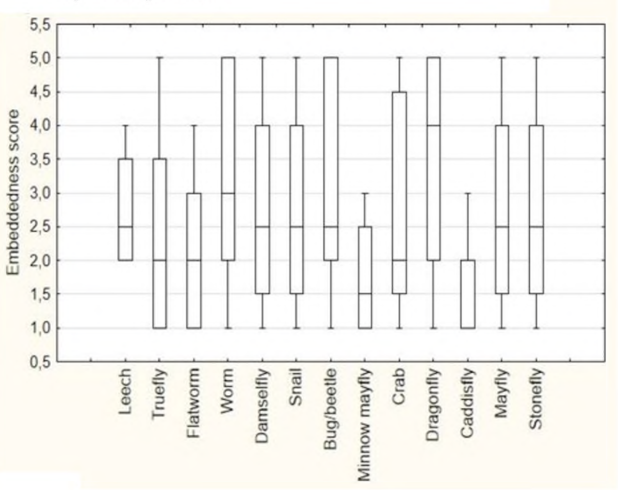
b) October 2014



c) March 2015



d) May 2015



e) August 2015

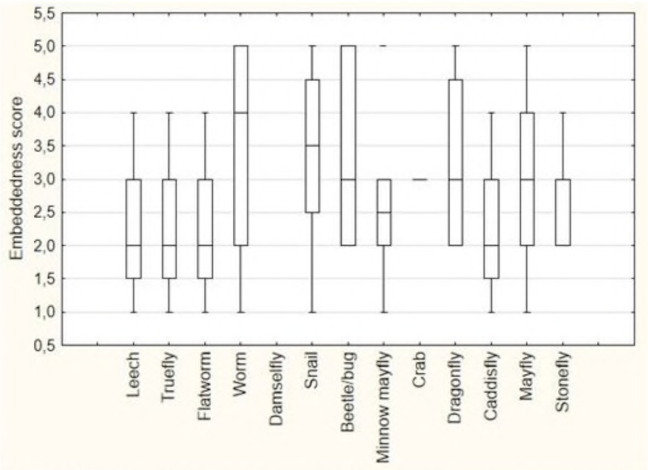


Figure 28 (a - e): Range of embeddedness scores per taxonomic group across all three sites, for July 2014, October 2014, March 2015, May 2015 and August 2015.

The plots showed that many taxonomic groups were found in patches with a wide range of embeddedness scores and often had a high upper limit (maximum value) tolerance of fine sediment, despite habitat preferences of taxonomic groups (refer to Table 6).

Modal embeddedness scores (Table 21) were calculated across all three sites based on the embeddedness score of each patch in which the taxonomic groups were found. These were found to be more reflective of habitat preferences. Maximum and minimum values reflected the range of embeddedness scores for each taxonomic group, whereas modal scores showed the dominant embeddedness value per group for all patches across all three sites.

In general, groups which favour fine sediment (trueflies, worms, bugs/beetles and dragonflies) were less affected by the amount of sediment in a patch than those which favour rocky habitats (flatworms, damselflies, snails, minnow mayflies, crabs/shrimps, caddisflies and other mayflies). Interestingly, modal scores were notably lower in May 2015 (between 1 and 2 for all groups except Dragonflies) than in previous months, which coincides with a significant decrease in stored sediment.

Table 21: Modal embeddedness values across all three sites for each taxonomic group for July 2014, October 2014, March 2015, May 2015 and August 2015

	July 2014	**October 2014**	**March 2015**	**May 2015**	**August 2015**
Leech	2	5	2	2	2
Truefly	1	4	2	1	2
Flatworm	3	5	3	1	2
Worm	2	3	2	2	4
Damselfly	1	3	3	1	Not present
Snail	2	2	3	1	3
Bug/beetle	3	2	2	2	2
Minnow mayfly	3	2	2	1	2
Crab/shrimp	3	3	2	2	3
Dragonfly	3	3	2	5	2
Caddisfly	2	2	3	1	2
Other mayfly	2	2	2	1	2
Stonefly	Not present	Not present	Not present	N/A	2

4.11 Influence of substrate composition on biotic community structure

Patch scale redundancy analysis

Substrate analysis was conducted at a patch scale, comparing biotic presence or absence in relation to substrate composition based on the measurement scale from Gordon et al. (2004). Figure 29 shows a similar result to the analysis of stored sediment in Figure 27.

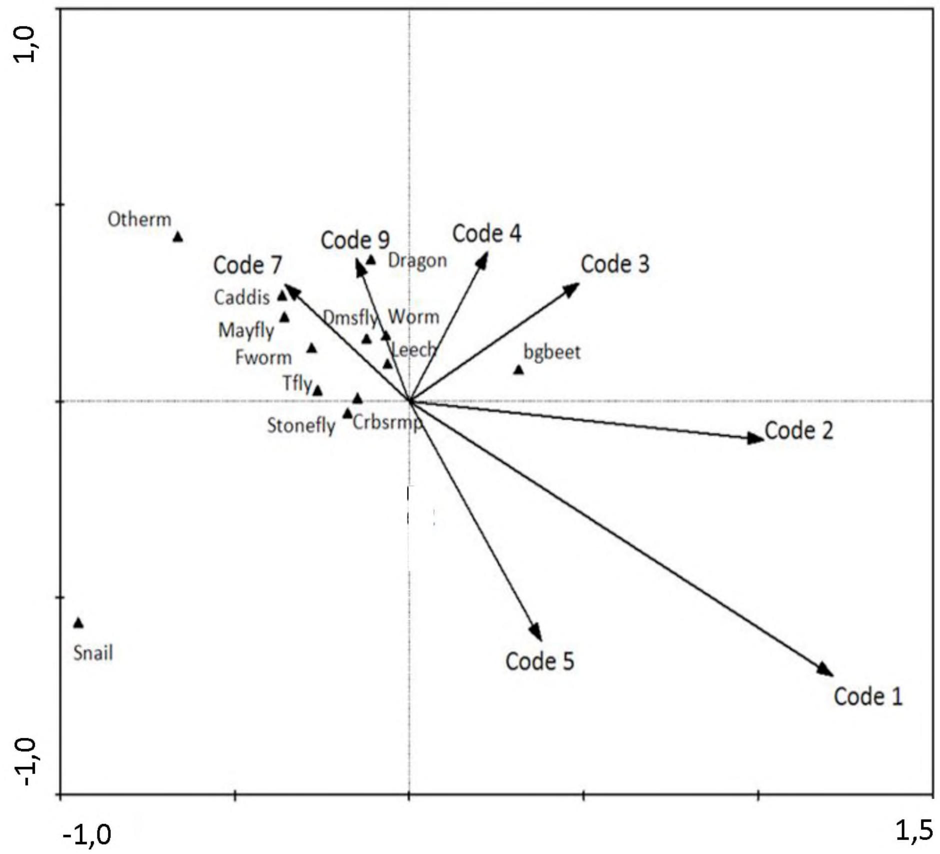


Figure 29: Total prevalence of each taxonomic group in relation to dominant clast type per patch, for all data collection periods (First axis variance = 100%, second axis variance = 100%). Clast codes taken from Gordon et al (2004): Code 1 = fine sediment, Code 2 = small gravel, Code 3 = medium gravel, Code 4 = large gravel, Code 5 = small cobble, Code 6 = medium cobble, Code 7 = large cobble, Code 8 = small boulder, Code 9 = large boulder.

Fine sediment (Code 1) and small gravel (Code 2) were the only substrates which produced a significant relationship with biotic community composition (0,004 and 0,002 respectively). This means that in patches where fine sediment and small gravel were the dominant substrate type, the biotic community was significantly different from patches where other clasts were dominant. However, the correlation was strongly negative, as the majority of taxonomic groups were closely associated with these clasts in the ordination chart. In fact, large cobbles (Code 7) and large boulders (Code 9) were the most important for community structure, with the majority of groups closely grouped around these two clasts.

Table 22: Eigenvalues, species-environment correlations, cumulative percentage variance or species data and species-environment relation and statistical significance (P) of total prevalence of each taxonomic group in relation to dominant clast type per patch, for all data collection periods (refer to Figure 29). Clast codes taken from Gordon et al (2004): Code 1 = fine sediment, Code 2 = small gravel, Code 3 = medium gravel, Code 4 = large gravel, Code 5 = small cobble, Code 6 = medium cobble, Code 7 = large cobble, Code 8 = small boulder, Code 9 = large boulder.

Axes	**1**	**2**	**3**	**4**	**Total variance**
Eigenvalues	0,022	0,161	0,115	0,104	1,000
Species-environment correlations	0,363	0,000	0,000	0,000	
Cumulative % variance of species data		2,2	18,3	29,8	40,2
Cumulative % variance of species-environment relation		100,0	0,0	0,0	0,0
Conditional Effects					
Variable	**Var.N**	**Lambda A**	**P**	**F**	
Code 1	14	0,02	0,004	3,35	
Code 2	15	0,02	0,002	2,56	
Code 5	18	0,01	0,118	1,63	
Code 7	20	0,01	0,128	1,43	
Code 3	16	0,01	0,192	1,29	
Code 9	22	0,00	0,334	1,16	
Code 4	17	0,01	0,394	1,09	
Code 6	19	0,01	0,450	0,96	

This shows that multiple biotic groups have a stronger positive correlation with patches in which larger clasts are dominant (Code 7, large cobbles, and Code 9, large boulders), while only the bug/beetle group (*bgbeet*) shows a correlation with patches predominated by smaller clasts (Code 2, small gravel, and Code 3, medium gravel). As Code 1 refers to fine sediment, which Figure 26 indicated that the majority of organisms have a negative correlation with, the clustering of data points around the Code 7 and Code 9 arrows may also be indicative of this negative correlation with Code 1. Gravel patches were generally found in low flow sections of the study sites and in a number of patches, the spaces between gravels were filled with fine sediment, so these clast sizes (Code 1, Code 2 and Code 3) are spatially linked, which may account for the similarity in their significance and correlation to biotic community composition.

Snails appear to have a strongly negative correlation with Code 3. While data collection revealed that snails were more commonly found attached to larger clasts, it is difficult to account for this particularly negative correlation. The crab/shrimp group (*Crbsrmp*), stoneflies and leeches show the lowest correlation to specific clast sizes, as their data points are situated close to origin (0,0). This suggests that these groups did not show a clear preference for substrate composition.

As stated above, only fine sediment (Code 1) and small gravel (Code 2) produced statistically significant relationships, shown in Table 22. These two variables also produced slightly higher LambdaA values, showing that they have greater explanatory power than the others. However, all the Lambda values remained low. The nonsignificant correlations produced by the other substrate types shows that they do not directly explain biotic variance, and that the null hypothesis cannot be rejected in this case.

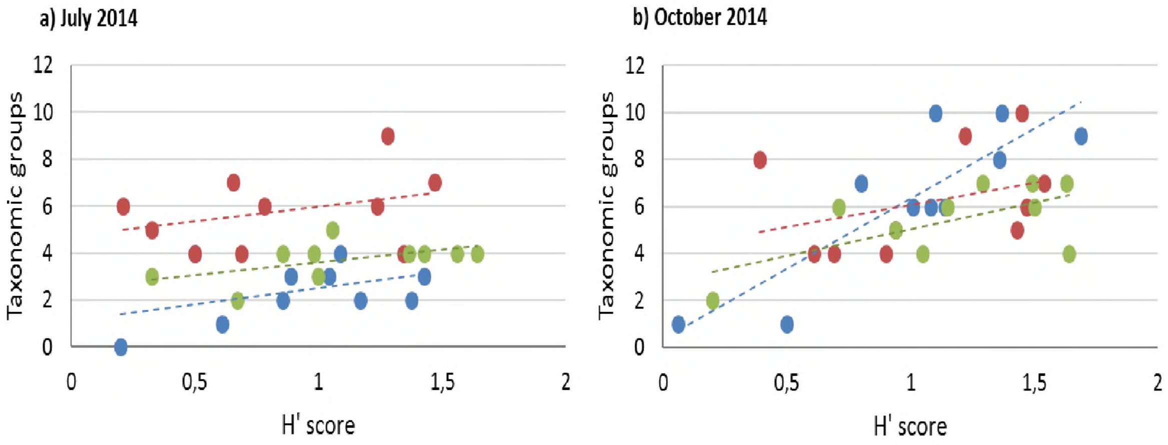
Shannon index of substrate diversity

In addition to measuring biotic response to substrate, the Shannon diversity index (H') was used to measure the substrate diversity in each sampled patch (Figure 30 a – e). R^2 values were calculated to assess the degree of correlation between the heterogeneity score and the number of MiniSASS taxonomic groups found in the patches (Table 23).

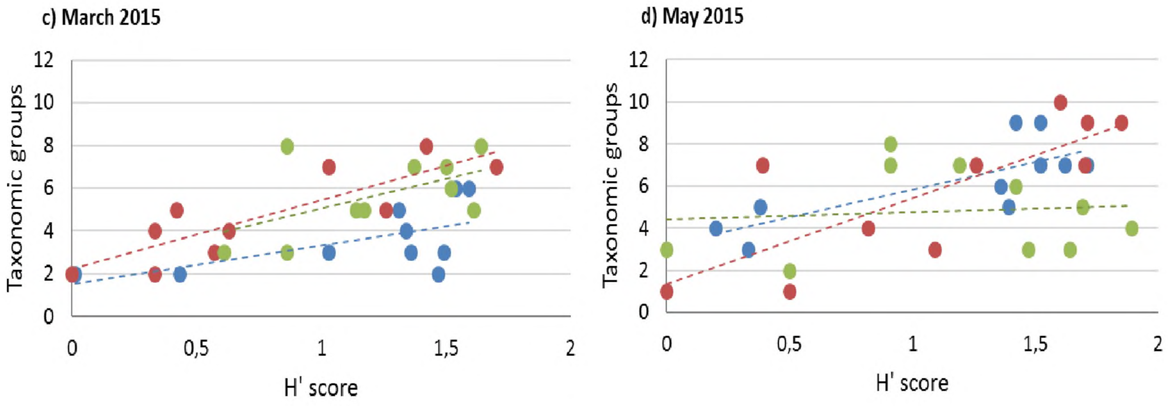
As discussed by Apitz (2012), substrate is an important driver in biotic community composition. Shannon index charts made use of only the 13 primary MiniSASS taxonomic groups. It is important to note that, while the HMID data in Figure 23 showed the sites to be fairly uniform in terms of flow, the H' scores show inter- and intra-site variability. The data shows a positive correlation in each patch throughout the study. Correlations were tested for significance at a threshold of $p = 0,05$.

In general, Site 1 and 2 showed fairly strong correlation between substrate heterogeneity and the presence or absence of taxonomic groups, while Site 3 showed a weaker correlation. July 2014 and May 2015 produced no significant relationships; however, in May 2015 Site 1 and 2 showed strong correlation between substrate heterogeneity and biotic diversity. All three sites produced a wide variety of substrate heterogeneity scores and the number of taxonomic groups found in each patch varied between 0 and 10. In general Site 1 had the lowest biotic diversity, while Site 2 had the highest. Significant correlations occurred in Site 1 in October 2014, and in Site 2 in March 2015 and August 2015.

High quantities of fine sediment resulted in lower biotic diversity, as was seen in Site 1 in July 2014. In addition to fine sediment, there was also a thick covering of green filamentous algae. The smothering effect caused by fine sediment, combined with the reduction of light and flow by algae created inhospitable conditions which precluded many biota from inhabiting this site.


a) July 2014

b) October 2014


c) March 2015

d) May 2015

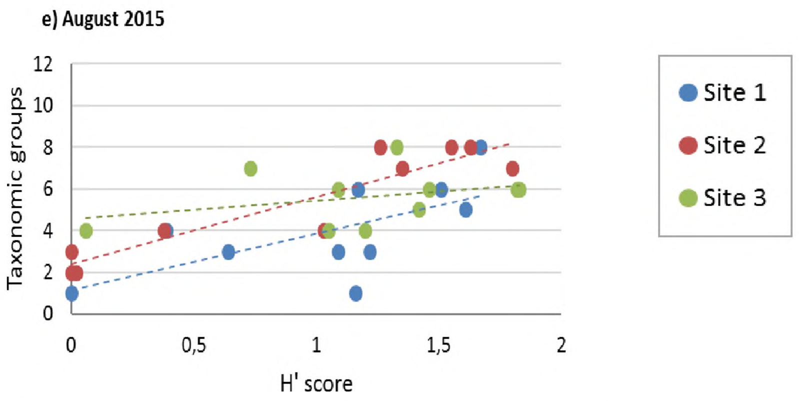

e) August 2015

Figure 30 (a – e): Shannon index (H') scores and number of unique taxonomic groups per patch, including trend lines for all three sites for July 2014, October 2014, March 2015, May 2015 and August 2015.

Table 23: R^2 values for Shannon index scores and number of taxonomic groups per site, for each data collection period, from Figure 23 (a – e) above. Statistically significant data (p <0,05) are in bold font.

R^2	**July 2014**	**October 2014**	**March 2015**	**May 2015**	**August 2015**
Site 1	0,18	**0,73**	0,36	0,60	0,42
Site 2	0,12	0,12	**0,72**	0,62	**0,86**
Site 3	0,29	0,39	0,30	0,01	0,11

Results from October 2014 (Figure 30 b) did not show as distinct a pattern within sites. Site 1 had a strong degree of correlation between H' score and number of groups, and while overall diversity was lower than the other sites, there was an increase in diversity from July 2014. Site 2 and 3 had much lower R^2 values, but Site 2 also continued to score highly in terms of species diversity. The water was clearer in October 2014 than in July 2014, with lower turbidity scores in all three sites, a smaller quantity of stored sediment and very little filamentous green algae in Site 1, creating healthier habitat patches. Furthermore, warmer water temperatures in late spring increased macroinvertebrate activity. This is particularly noticeable in Site 1, where the number of groups was far lower in July 2014 when water temperatures were very low, than in the warmer weather of October 2014 (late Spring).

In March 2015 (Figure 30 c), the effects of the drought are again reflected in the results. It is worth noting that Site 1 was largely made up of disconnected standing pools and small riffles in this period. Many of the pools were isolated refugia, but often had murky water, which made them difficult to sample. Most of the patches sampled in Site 1 were in the riffle sections, as these were easier to sample, and also offered a greater variety of substrate types. While this introduced bias into the sampling method, these patches had only two or three taxonomic groups in them, which does illustrate the negative impact of drought conditions on biotic community. The drought also highlights the importance of longitudinal connectivity, as mentioned by Bunn and Arthington (2002), for maintaining biotic communities. As conditions in Site 1 deteriorated, biota would have moved downstream to find more suitable habitat patches, also contributing to lower biotic diversity in the site. Site 2 and 3 had greater numbers of taxonomic groups. The stored sediment load in both sites was at its highest, which created patches of thick sediment in the low flow areas of the sites. These areas generally had low biotic diversity, but the benefit of groundwater flow allowed for greater habitat availability in the sites and more complex biotic communities to be maintained and supported.

Both May 2015 and August 2015 (Figure 30 d and e) showed similar patterns of data distribution. During both these periods the fine sediment load was low as a result of the increase in flow, and the variation in H' scores shows a wide variety of available habitats. Site 1 and 2 in May 2015 and August 2015 show strong correlation between the number of taxonomic groups and the H' score. The surprisingly low correlation in Site 3 for May 2015 is difficult to account for as the chemical and sediment data suggest good habitat and water quality, yet the biotic abundance was lower than expected in this site. Biotic and habitat health were both high in May 2015. In addition to low sediment loads, the water was well oxygenated (>90 % saturation) in all three sites, the pH was neutral or close to it, the channel was also free from filamentous algae and turbidity was low, allowing sunlight, oxygen and nutrients to filter through the water column. In response, biological diversity was high. It is important to note that May 2015 was the first time stoneflies were found in the sites. Not only are stoneflies highly sensitive to water quality, but many species of stonefly are predators and therefore require a stable food chain (see Table 6).

August 2015 had lower overall diversity in all sites. This may relate to breeding habits of aquatic macroinvertebrates, but a more in-depth study would be needed for more conclusive answers. It is also significant that all three sites had their lowest DO levels in August 2015. The low oxygen content can be attributed to a layer of dead or dying algae that covered large areas of the stream bed in all three sites, but particularly in Site 1 and 2. As algae decays, it uses up oxygen from the water (Twilley *et al.*, 1986) and the dead matter has a smothering effect on the substrate, making the bed less suitable for species which require well oxygenated, flowing water. However, the greater discharge volumes kept some parts of the channel clear of the algal matter, particularly in Site 3, which experienced a small flood in early August 2015, maintaining a higher biotic abundance and creating new patches which species could inhabit.

Finally, patch scale sensitivity scores were compared with the Shannon index scores (Figure 31 a – e), and R^2 values were calculated to assess the level of correlation. The analysis showed little to no correlation between the data sets and, as Table 25 shows, no statistically significant R^2 values were produced, nor were any spatial or temporal trends found.

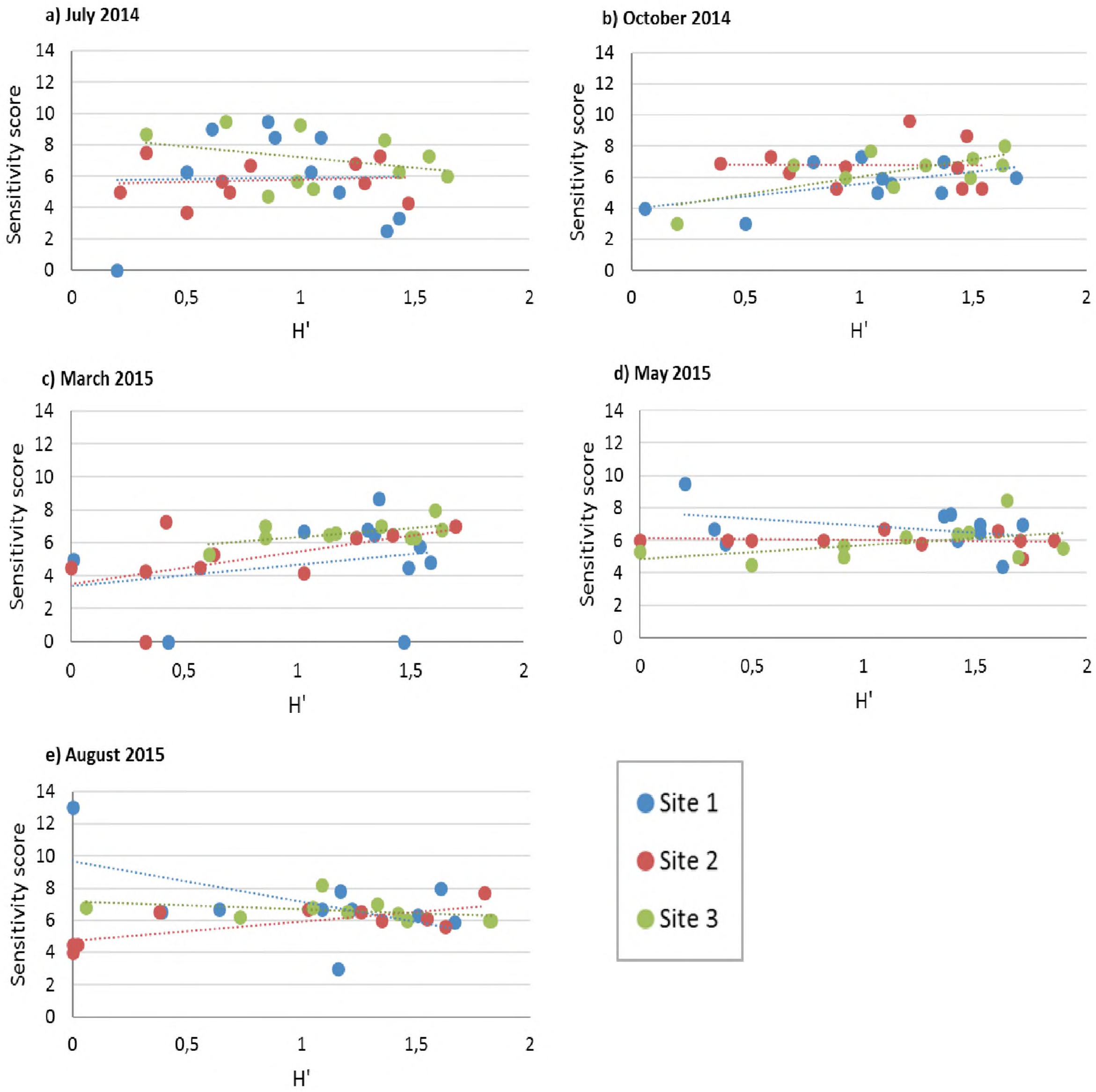


Figure 31 (a – e): Shannon index (H') scores and sensitivity scores per patch, including trendlines for all three sites for July 2014, October 2014, March 2015, May 2015 and August 2015

Table 24: R^2 values for Shannon index scores and sensitivity scores for each site, from each data collection period, calculated from Figure 31 (a – e).

	July 2014	**October 2014**	**March 2015**	**May 2015**	**August 2015**
Site 1	0,00	0,29	0,06	0,14	0,30
Site 2	0,01	0,00	0,27	0,02	0,58
Site 3	0,10	0,52	0,33	0,20	0,13

Sensitivity scores per patch plotted against H' scores show that in most data collection periods the data is clustered along the vertical axis, spanning a relatively small range of sensitivity scores, with a few outliers where patches scored very high or very low on the sensitivity scale.

July 2014 is the exception to this, as the sensitivity scores cover a wider range, but the data still do not reveal any relationship between the variables. As none of the data can be considered statistically significant ($p > 0,05$) there is little which can be concluded from these figures. As discussed earlier, this indicates that sensitivity scores are not reliable indicators of habitat quality.

4.12 Key findings

This chapter has presented a large amount of data, covering all four parts of the two key objectives:

1. Describe the spatial and temporal instream habitat condition and availability
2. Describe the biotic community composition
3. Investigate the influence of flow and sediment, as system drivers, on habitat quality
4. Investigate the influence of flow and sediment, as system drivers, on biotic response to habitat changes,

and the secondary objective to assess the usefulness of the MiniSASS methodology and scoring system as a measure of habitat quality as well as water quality.

However, as much of the data was used to create the RDA ordination charts, the HMID scores and the Shannon index scores, it is possible to summarise the results into a number of key findings which are particularly important for this study. The main findings which will be carried forward into the Discussion chapter are as follows:

Ecological response: MiniSASS sensitivity scores

The Wilgerbos River was found to be in good health (as a sandy type river) or fair health (as a rocky type river). Overall, the MiniSASS scores fluctuated over the course of the study, but sandy type patches were generally scored more highly than rocky type patches. Sensitivity scores were shown to be correlated with sampling date, and strongly correlated to substrate. MiniSASS scores were not shown to be useful as a measure of habitat health as they often did not correlate with physical variables.

Biotic community analysis

Biotic diversity was high throughout the study, with the majority of taxonomic groups present in all data collection periods. However, the abundance of taxonomic groups fluctuates spatially and temporally, with lower abundance scores in July 2014, March 2015 and August 2015, and high abundance scores in October 2014 and May 2015. Redundancy analysis shows biotic presence and absence to be strongly correlated with sampling date and site. This shows that biotic community composition different significantly among the sites during the study, in contrast with sensitivity scores which did not show a correlation with site.

Influence of flow properties

Depth was shown to be significantly correlated with biotic presence and absence, but velocity was not. However, as a number of taxonomic groups were closely associated with the velocity arrow in Figure 22 velocity must be an important variable in biotic community structure, suggesting that the relationship is not direct, and the impact of velocity produces other habitat conditions which have a direct impact on biota. Flow heterogeneity was shown to have a little to no correlation with biotic presence or absence, and the data suggests that high heterogeneity can have a negative impact on the biotic community, particularly when paired with low flow conditions.

Influence of water chemistry variables

All water chemistry variables were found to be significantly correlated to biotic community composition, except DO. However, the water chemistry data collected does not show any clear temporal or spatial trends, and the drought in March 2015 created abnormal conditions, making it difficult to draw further conclusions about the relationship between biota and water chemistry.

Influence of fine sediment and substrate

At a site scale, stored sediment showed a significant negative correlation with all taxonomic groups except the bugs and beetles group. All other groups showed a preference for lower sediment habitats. Similarly, at a patch scale, substrate analysis showed fine sediment and fine gravel to have a strongly significant correlation with biotic presence and absence, but the majority of taxonomic groups showed a preference for patches with larger clasts, particularly large cobles and boulders. In this case, the significance of the correlation shows that biotic communities were significantly different in patches with high embeddedness than in patches with lower embeddedness.

Patch scale measures of fine sediment (i.e. embeddedness) were more important for assessing habitat preferences of taxonomic groups than site scale measures (stored sediment), and could be more easily related to biotic abundance and diversity, making them more useful. Substrate heterogeneity was shown to be important for biotic diversity, but biota with a preference for fine sediment habitats were able to inhabit a wider variety of patch types, regardless of substrate composition.

General findings

Although algae and groundwater were not quantitatively measured in this study, they proved to be important considerations in habitat assessment as their presence can affect habitat condition, biotic community structure and stream response to disturbance. Algae had both a negative and positive impact on the physical habitat, sometimes creating highly oxygenated conditions, but also smothering the substrate and preventing light and heat reaching lower levels of the water column. Groundwater had an overwhelmingly positive affect on physical habitat, allowing faster recovery after the drought and creating more stable instream conditions.

With these key findings in mind, the *Discussion* chapter will primarily focus on investigating the influence of flow and sediment on habitat quality and on biotic response to habitat changes, as well as further discussion on the usefulness of the MiniSASS methodology and scoring system as a measure of habitat quality.

CHAPTER 5: DISCUSSION

This study was an investigation into the changes in biotic community structure in response to changes in habitat as result of flow, sediment and water chemistry dynamics over a period of one year. The *Results* chapter focused primarily on the first primary objective of this study, to describe the spatial and temporal variability of habitat conditions and biotic community composition for a groundwater–fed semi-arid river. The *Discussion* chapter will deal mainly with the second objective, to investigate the relationship between system drivers (flow and sediment), habitat quality and biotic response, and the secondary objective to assess the usefulness of the MiniSASS methodology and scoring system as a measure of habitat quality as well as water quality. This section of the thesis will re-present the major finding from the *Results* chapter and draw together the data to investigate the correlations observed.

As with the results, data analysis for this study was conducted in stages before being drawn together. While the overall geomorphic health of the Wilgerbos River was good (see *Results, Section 4.6*), analysis of the data suggests that the natural disturbances in the system have a very strong impact on habitat and biotic community structure. Biotic response to changes in flow is clear from the results, but in some cases the trends observed cannot be easily explained by the data collected, suggesting that either multiple causes were in effect, or that a variable which this study did not account for has a greater influence than expected. One such variable is groundwater. Groundwater inputs and levels were not measured in this study, but the likely influence of groundwater on flow and therefore on biotic communities in all three sites was a very important consideration in the interpretation of the results. The diversity of sites sampled provided a good representation of different habitats and changes in habitat condition and availability with changes in flow and sediment dynamics (see Figure 23 and Figure 30 a – e). In conclusion, this chapter will discuss the limitations of this study, alongside hypotheses generated and recommendations for overcoming these limitations in future studies.

5.1 MiniSASS sensitivity score analysis

Sensitivity scores are ratings of ecological health derived from the sensitivity of biota to changes in water quality, based on their habitat requirements and habits (Graham *et al,* 2004). Organisms such as true flies, flat worms, leeches and worms, which are able to inhabit a variety of habitats and have flexible feeding habits, have low sensitivity scores. In general, organisms which are described as having multiple food sources by GroundTruth (2016) also have low sensitivity scores (see Table 5 and Table 6) as they are more flexible feeders and better able to adapt to changes in habitat conditions (Tomanova *et al.*, 2006). Similarly, organisms which feed in fine substrate (i.e. worms and crabs) or leaf litter, are less likely to experience severe food shortages (Tomanova *et al.*, 2006). Predatory biota listed by GroundTruth (2016), such as stoneflies, caddisflies and dragonfly nymphs, require a more complex ecosystem with a stable food chain in order to feed, and many develop secondary feeding habits, such as scavenging to overcome this (Tomanova *et al.*, 2006). Organisms with limited feeding habits, such as stoneflies and mayflies are more likely to be sensitive to changes in habitat conditions as they are less flexible and therefore more dependent on consistent food sources (Tomanova *et al.*, 2006)

As discussed in *Section 4.6* of the Results chapter, an important difference between using MiniSASS to assess water quality and using it to assess habitat quality is that sensitivity scores were not found in this study to adequately represent the abundance of each taxonomic group, which is an important consideration for assessing habitat health, as it is a significant factor of biodiversity and habitat functionality (Mace *et al.*, 2005). As discussed above, when assessing habitat quality, a diverse population is an indicator of a healthy habitat (Mandaville, 2002). While the MiniSASS assessment does indirectly measure diversity, by recording all the taxonomic groups found in a site, there is no measure of the abundance of biota. Further, as Figure 24 and Figure 31 show, sensitivity scores do not correlate to habitat quality variables, such as flow heterogeneity (HMID) and substrate heterogeneity (Shannon index, H' scores), which suggests that sensitivity scores cannot be considered descriptive of habitat condition beyond water quality.

The river health categories used in MiniSASS assessments are also not relevant measures of habitat quality as they assume a level of human modification to the the stream channel. While some patches scored the "poor" condition descriptor in the MiniSASS assessment, this is less useful in the context of this study, as it implies a high degree of modification in the river. While all three sites experience some human impact, none would be considered largely modified. However, it is important to note that in these poor condition patches, the dominant biotic groups were those with low sensitivity scores. These are due to a variety of factors, such a water chemistry, sedimentation, and water depth, but no clear causal relationships emerge which could be applied as a rule to the scores.

This does not mean that the sensitivity score is not valuable for habitat quality assessment, but rather that a sensitivity score should be more carefully considered in the context of habitat health and biotic diversity. The sensitivity scores are primarily intended to measure water quality and habitat quality is a factor of multiple drivers and variables other than water quality. However, no major water quality issues were found during the study, and biotic abundance and diversity were both good, which supports the "fair" to "good" sensitivity score for the river. The sensitivity rankings allocated for the MiniSASS system are useful for interpreting how healthy the biotic community is, as an abundance of sensitive organisms suggests a healthy, stable habitat, while an abundance of less sensitive, hardier organisms suggests a harsher habitat. Furthermore, the MiniSASS taxonomic groups are easily recognisable and can be quickly identified and counted even by inexperienced assessors.

5.2 Spatial and temporal variability in biotic communities

The results of the RDA (Figure 20) showed the correlation between biotic abundance and sample date to be statistically significant for all months, except August 2015. In general, taxonomic groups were fairly common throughout the course of the study, but in some cases, the data reflect the seasonality of some taxonomic groups. For example, as shown in Figure 19 a – e stoneflies, were present only in May 2015 and August 2015, damselflies were very prevalent in May 2015, but were entirely absent in August 2015, and snails had very low abundance in July 2014. Chi-square analyses (Table 14 a) showed that there was no significant relationship between sensitivity scores and date. Again, this creates a polarisation of abundance and sensitivity data. While the RDA reflects the seasonality of biotic groups, showing when individual groups are present or absent in the stream, the Chi-square analysis reveals that there is no uniformity in how sensitivity to water quality influences seasonal cycles. In short, the analyses show that the sensitivity of organisms has no relationship with their breeding cycles, and seasonality is entirely unique to each group.

In addition to being common through the study period, most taxonomic groups were also cosmopolitan and quite evenly distributed across all three sites. Redundancy analysis (Figure 21) showed Site 1 and 3 to have a significant relationship with biotic community structure, and Chi-square analyses (Table 14 c) showed a weak relationship between site and sensitivity score. However, the apparent site preferences of some groups are largely a factor of differences in substrate among the three sites. Both the bug/beetle group and the truefly group, which favour fine sediment habitats, showed a preference for Site 2 as it had the highest number of sandy patches overall, while the other mayfly group, which favours rocky habitats, was slightly more common in Site 3 which had a higher number of rocky patches. The deep pool in the middle section of Site 2, has very thick sediment on the bed, which many organisms would find inhospitable, and as a result, the site had low biotic diversity. Conversely, Site 3 had a diverse habitat arrangement with good habitat accessibility, which may explain why many groups had a preference for the site.

In general, high biotic diversity was taken as an indicator of a healthy habitat, with a well-developed ecosystem. The presence of a number of groups with varying habitat preferences and feeding habits shows good availability of a variety of habitats within a site, and also a functional food-web, able to support browsers, filter feeders and predators alike. As shown in Figure 19 a – e, all three sites supported a variety of organisms over the course of the study. However, in general Site 1 had the lowest abundance and diversity of taxonomic groups, suggesting a less complex ecosystem than in Site 2 and 3.

Because the sensitivity scoring system relies on averages, sites can be poorly represented in terms of habitat health, by the presence of low sensitivity groups. Conversely, some sites received high sensitivity scores due to the presence of a few high sensitivity scores, but had low abundance of taxonomic groups. This highlights the challenges of using sensitivity scores to assess habitat health. There are a number of examples where the sensitivity score for a site did not correspond to abundance or diversity of taxonomic groups. For example, in July 2014 and May 2015, Site 2 received a low sensitivity score, despite a high abundance of a number of taxonomic groups, including those with high sensitivity scores. The same is true for Site 3 in May 2015. In all three cases, the presence of low sensitivity organisms negatively skewed the average scores for the sites, producing some of the low scores shown in Figure 17 a – e and Figure 18 a – e.

Higher abundance of low sensitivity organisms, such as was recorded in Site 3 in May 2015 (Figure 17 d) is considered to be indicative of harsher habitat conditions, as low sensitivity organisms are better able to cope with changes in habitat condition (Graham *et al*, 2010), and as some are scavengers (see Table 6) it can be assumed that they are less susceptible to reduced food availability. Conversely, an abundance of high sensitivity organisms is taken to indicate good habitat conditions as they often have more specific habitat requirements and are more sensitive to changes (Graham *et al.*, 2010). Many are also carnivores or grazers and therefore would require stable food sources. In this respect, the use of sensitivity scores is valuable as it can provide an indication of habitat condition. However, the use of abundance data is a necessary supplement to provide a fair assessment of habitat health.

In summary, biotic abundance fluctuated both spatially and temporally, but most taxonomic groups showed a preference for Site 2 and 3, with the former being diversely populated throughout the study. Further, while some groups were abundant during all data collection trips, damselflies, stoneflies and snails had notable increases and decreases in abundance at certain times, sometimes being entirely absent from a site. However, while RDAs showed strongly significant spatial and temporal correlations with biotic community composition, Chi-square analyses did not produce strong relationships with sensitivity scores. This indicates that temporal and spatial abundance is not a factor of sensitivity, and must be measured at an individual group level.

5.3 Influence of flow properties on biotic community structure

The RDA (Figure 22) showed that velocity did not have a significant relationship with biotic community composition as only a small number of groups were closely associated with this variable. It is necessary to acknowledge the greater potential for errors in velocity measurements than in depth measurements, as in shallow water the Flowmate probe could not always be fully immersed in water. However, there is an important indirect relationship, as low flow creates highly sedimented habitat patches, while fast flow creates rocky habitat patches. Therefore, in this regard velocity is an important factor in biotic diversity through habitat creation, but as flow is variable, changes in sediment dynamics can lag behind changes in flow, so no direct relationship could be identified from the data collected. Velocity and depth tend to be inversely proportional, with lower velocity in deep water than in shallow water. As velocity affects mixing in the water column, deep water has lower nutrient levels, oxygen content and temperature than shallow water, all of which influence which organisms will reside in a habitat patch. Therefore, while velocity may not have produced a statistically significant relationship with biotic presence and absence, the relationship between velocity and depth is important to keep in mind. It is possible for an organism to inhabit patches with a variety of flow speeds, but as depth increases, many organisms become more selective, often as an indirect result of reduced velocity. For example, stoneflies are particularly sensitive to oxygen content, and therefore will not be found in deep water, as shallow water is easier to mix, even at low velocities, and therefore has a higher oxygen content.

Rocky type patches seemed to be less affected by flow conditions, as no clear relationship could be derived from the sensitivity scores and flow data for these patches. There are a number of possible reasons for this: larger clasts increase friction along the river bed, creating more turbulence and promoting better mixing of heat, oxygen and nutrients even in low flow; high turbulence can make patches inhospitable for organisms which cling to rocks and cobbles; larger clasts create refugia for organisms which rely on filter feeding from the water column and organisms with gills; larger clasts are able to support communities of algae and lichen which also support browsing grazing biota. These conditions create confounding properties which can alter biotic presence and absence in these patches in unexpected ways.

Despite the significant relationship between depth and biotic diversity and the non-significant relationship with velocity shown in the analysis, the fact that some organisms in data collection showed preferences for certain velocity conditions, suggests that this variable is important in some cases, but some taxonomic groups are clearly more sensitive to velocity than others. However, further analysis shows that, while depth was significant at a patch scale, at a site scale flow heterogeneity was not a significant driver of biotic diversity and that biotic diversity was fairly even across sites, despite variations in flow conditions.

The hydromorphological index of diversity scores (HMID) (Table 10 and Figure 23) showed that biotic communities remained relatively constant across the three sites, despite high variability in flow properties. The scores were clustered along the vertical axis of taxonomic groups, showing a similar number of groups present in each site. Along the horizontal axis of HMID scores, the scores were more widely dispersed, showing varying degrees of diversity in each site. Data for July 2014, shows lower HMID scores, which indicates greater homogeneity of flow properties than in the following periods. In May 2015, Site 1 showed greater diversity of flow than the other two sites. This is an unexpected finding, as Site 1 is considered the most homogeneous of the three sites due to it being mostly a single thread channel with the lowest slope gradient. Site 1 was also a clear outlier in March 2015, as the dryness of the lower half of the site produced highly heterogeneous conditions. Nonetheless, when viewed in terms of the whole site, rather than per patch, it becomes clear that the effect of the drought was not as drastic as it initially appeared, and Site 1 supported as many taxonomic groups in March 2015 as it did in July 2014, but in a lower abundance.

The slightly negative relationship between HMID and biotic diversity shows that low flow increases heterogeneity whereas high flow drowns out variability. This indicates that high heterogeneity may not in itself be a good thing. It may be more useful for flow diversity to be combined with another flow measure, such as mean flow depth, when assessing habitat condition.

5.4 Influence of water chemistry variables on biotic community structure

Overall, water chemistry in the Wilgerbos River showed the river to be healthy according to guidelines provided by DWAF (1996), Behar (1997) and USGS (2015). No clear, long term trends could be found between variables and time, or site; however, as Figure 25 shows, most water quality variables were shown to have a significant relationship with presence and absence of taxonomic groups, suggesting that water chemistry is correlated to community structure.

Phosphate was shown in Table 18 to have a statistically significant relationship with biotic community composition, and concentrations fluctuated throughout the study. However, Figure 25 shows that phosphate (P) had a weak influence on biotic presence or absence, indicated by the short arrow. Behar (1997) states that biota in general are sensitive to changes in phosphate levels of greater than 0,1 mg/l and that very high phosphate concentrations will certainly have a harmful effect on biotic communities. All three sites experienced a large increase in phosphate levels in March 2015 and, as shown in Table 11 a – c, Site 2 had the highest concentration of phosphates of the three sites. Figure 19 c shows that Site 2 almost uniformly has lower abundance of each taxonomic group than Site 3, except for low sensitivity organisms including snails, flatworms and trueflies. Other than phosphate concentration, Site 2 and 3 have no other remarkable differences in measured water chemistry, making it likely that phosphate is a factor in biotic community structure. However, unmeasured parameters may also be affecting the sites, and the apparent influence of phosphate may be coincidental, which would account for the weak influence shown in Figure 25. The low biotic abundance in Site 1 in March 2015 is likely due to multiple factors, of which phosphate may be one, which will be discussed in greater detail later.

Groundwater contributions offer a likely explanation for increased phosphate concentrations in Site 2 and 3, as groundwater may introduce phosphate leached from the underlying geology (Vanek, 1991). Mazurov et. al. (2007) and Cook and Shergold (2005) state that sandstone, mudstone and dolerite, the dominant geologies of the region, are all sources of phosphate, and the granular nature of sandstone and mudstone allows for storage of phosphate nodules around nuclei such as quartz and calcite. Groundwater is known to leach nutrients from soil and rock, and concentrations of nutrients, including phosphate, are often higher in groundwater-fed systems, particularly in the shallower waters of the riparian zone (Vanek, 1991). The shallow water depth of the Wilgerbos River and the underlying geology of sandstone, mudstone and dolerite, it is likely that during a period of drought, when groundwater is the major contributor to channel flow with no dilution from surface flow or rainfall, phosphate levels will increase.

While groundwater does not fully account for the increased phosphate in Site 1, phosphate concentrations can be strongly linked to channel bed sediment. Fine sediments, such as silt and clay have a high sorption and storage capacity for phosphate (DWAF, 1996; Carlyle & Hill, 2001). The bed sediments in the Wilgerbos River are very fine, and while the flow of the river generally washes much of the sediment away, during the drought large quantities had accumulated in standing pools in Site 1. The warmer water temperature in these pools likely caused soluble phosphate to dissolve more quickly out of the sediment, causing the peak to occur within a relatively short timeframe (Busman *et al.*, 2009). The higher phosphate levels experienced in March 2015 may have contributed to the reduced biotic diversity in each site, and may account for the high variability in biotic abundance between the three sites. A more detailed study of phosphate concentrations in over time would be necessary before a conclusion could be drawn.

Electrical conductivity (EC), like phosphate, was also highly significant in shaping biotic community composition (Figure 25), but abundance data (Figure 19 a – e) contradicted expected findings on the relationship between EC and biotic presence and absence data during the study. As EC is a reflection of total dissolved salts, increased EC can affect metabolic and osmotic processes in organisms, and therefore it was expected that increased EC would result in decreased biotic abundance. However, this was not the case. All three sites experienced a peak in EC in May 2015, which is contrary to expected findings, as the high flow due to rainfall, combined with the more neutral pH recorded in all three sites, should have caused a decrease in EC (Leveling, 2002). However, increased groundwater contribution to the water column due to rainfall may have raised the EC by introducing more dissolved salts. However, the biotic abundance data show that biotic diversity and abundance were higher in May 2015 than in months with lower EC, such as October 2014 and August 2015. This unexpected relationship is suggestive of situation specific tolerance and adaptation to the naturally higher EC in the system. DWAF (1996) states that in some systems organisms adapt to fluctuating levels of total dissolved salts (directly proportional to EC). However, EC remained consistently high throughout the study, with a slight decline in August 2015 due to the dilution effect of snow melt contributions to flow (Steynberg, H., pers. comm., 2015).

In general, the pH of the Wilgerbos River was slightly basic, with minor fluctuations throughout the course of the study. Although pH appears to be significantly correlated to the biotic presence and absence in the RDA (Figure 25), the correlation is unclear when comparing data on pH and the presence of taxonomic groups in Figure 19 a – e. The increase in alkalinity in Site 1 in March 2015 is again a likely factor in the drop in biotic diversity, as DWAF (1996) states that increases in pH can affect metabolic processes and osmotic balance of aquatic organisms, reduce the availability of certain nutrients and create toxic ammonium compounds (NH_3). The more neutral pH in Site 1 and 2 in May 2015 may have allowed these sites to have higher diversity of almost all taxonomic groups in this period than Site 3 which remained basic. The neutral pH may also have been what enabled the appearance of stoneflies in Site 1. Apart from these two instances, however, fluctuations in pH do not seem to coincide with any particular trends in biotic community structure or taxonomic group abundance.

Water temperature showed a less clear correlation with biotic communities, but there is evidence to suggest that it contributed to changes in abundance. In July 2014, when the temperature in Site 1 was particularly low (4,5°C), the number of taxonomic groups found was lower than any of the later data collection periods. It is likely that the low water temperatures were unsuitable for many biota. According to DWAF (1996), cold water reduces metabolic activity in aquatic organisms, and so the community was limited to organisms able to survive in the low temperatures, or else those which prefer shallower or more turbid water where the temperature may have been warmer. Many biota also have seasonal breeding cycles, and low water temperatures in winter are unsuitable for some nymphs and larvae (Dickens & Graham, 2002). In October 2014, when temperatures increased, overall biotic diversity increased in all three sites. Higher temperatures in March 2015 were a factor of seasons ,given that March is a summer month, and water levels were lower, allowing the water to heat up more quickly (DWAF, 1996). However, in May 2015 and August 2015, biotic abundance remained high despite the decrease in temperature, which suggests again that organisms in this system have a tolerance for lower water temperatures, and many seemingly thrive around the 10°C mark. "Temperature is a key factor affecting the number and kinds of species in a stream" (Rivers-Moore *et al.,* 2008: 5). DWAF (1996) states that organisms have upper and lower tolerances for temperature changes, but that these are not uniform, because the sensitivity of an organism to water temperature depends on the flexibility of their life stages (Rivers-Moore *et al.,* 2008). Organisms with more flexible life histories rely less on temperature than organisms with more rigid life histories which rely on thermal cues to dictate transitions into different life stages (Rivers-Moore *et al.,* 2008). These more flexible organisms may then be present year-round in rivers with extreme temperature fluctuations, as is common in semi-arid systems (Rivers-Moore *et al.,* 2008).

Turbidity and temperature are often connected (DWAF, 1996), as higher turbidity causes higher temperatures, and so the relationship between turbidity and biotic diversity mirrors that of temperature and biotic diversity. While increased temperatures in summer may also account for warmer water measured in March 2015 (Table 11 a – c), the summer period had been unusually dry (Figure 9 and 10), so increased turbidity is not likely to have been a result of increased rainfall. Changes in turbidity levels of 10 - 20 ppm from the norm are considered significant and may have harmful effects on biota as it significantly reduces light filtration and oxygen content, and increases temperature (van Osch, 2009). This again accounts for the low diversity in March 2015, particularly in Site 1 where turbidity was particularly high. None of the sites experienced prolonged fluctuations of this magnitude and so biotic communities were able to recover quickly when turbidity dropped greatly in all three sites in May 2015 and August 2015.

Similarly, dissolved oxygen did not have a significant relationship with biotic diversity, and this can be explained by understanding how oxygen content can affect organisms. Continued exposure to oxygen saturation of less than 80% can be harmful to biota, but DWAF (1996) state that sub-lethal effects are only witnessed after continued exposure to 60% saturation, and lethal effects only at 40% saturation, which places the study sites well within a healthy range in all data sets but one. In August 2015, Site 1 and 2 experienced DO concentrations lower than 80%, during which time both experienced a decline in species abundance compared with May 2015, when both sites had DO concentrations higher than 80%. Occasional and short-term fluctuations in DO content are less important to study, as they have little to no impact on biotic communities (DWAF, 1996).

Data from October 2014 and May 2015, also reflect the importance of water chemistry in biotic community composition, as warmer temperatures, lower turbidity, well oxygenated water and lower pH were all identified as contributing to greater biotic diversity and abundance in all three sites, alongside high substrate diversity. Figure 25 showed all water chemistry variables, except DO, to have a significant correlation with biotic community. However, as data analysis showed, these factors did not produce consistent results over time, and it was difficult to identify clear trends in changes in community composition, as multiple water chemistry variables can act on a community at one time, and interact with each other (such as temperature affecting DO content). Individual taxonomic groups have particular sensitivities to water chemistry, rather than the more general sensitivity biota have to factors such as substrate and flow. Therefore, water chemistry is a significant driver of biotic community composition, but more specifically, it is a significant driver in the presence and absence of individual biotic groups.

In summary, water quality did not produce any clear relationships, despite the statistically significant correlations shown in Figure 25 and, in some instances, the data showed to be the opposite of what was expected. A number of water quality factors were likely contributors to overall low biotic abundance in Site 1 in March 2015, but these can all be attributed to the drought conditions at the time. Similarly, other correlations can be explained by changes in other, more physical habitat properties, such as groundwater supply and bed sediment, or by unmeasured water quality variables. However, due to the lack of recorded evidence showing significant temporal or spatial causal relationships between water quality and biotic diversity, changes in other variables such as flow and bed sediment must be considered as more significant drivers of biotic change in the long term. However, fluctuations in water chemistry were significant as short-term drivers of community structure in the Wilgerbos River.

5.5 Influence of fine sediment on biotic community structure

For the most part, the results of the CANOCO RDA of stored sediment in relation to the presence or absence of taxonomic groups showed (Figure 26) the expected results in terms of sediment preferences for each taxonomic group, e.g. the bug/beetle group favour high sediment habitats, while snails, which favour rocky habitats (GroundTruth, 2016) are situated far from the arrow. However, some unexpected results were evident too, such as worms and trueflies not being closely associated with the fine sediment arrow, despite favouring fine sediment habitats (GroundTruth, 2016). Stored sediment was a significant factor in community composition, but produced a negative correlation with almost all taxonomic groups, showing a strong preference overall for lower sediment habitats. This was also reflected in Figure 29, which showed a significant negative correlation between most biotic groups and fine sediment and small gravel. There are a number of reasons for this relationship, which were discussed in *Section 2.5* of this thesis, but will be briefly restated here: high sedimentation can cause infilling of niches between larger substrate materials, resulting in loss of habitat for biota which cling to or hide under rocks, physical damage through abrasion and clogging of gills and mouth parts, and restricted movement of water through the substrate, creating colder, lower oxygen conditions (Griffith & Walton, 1978; Harter, 2001; Berry *et al.*, 2003).

Although two methods were used to measure fine sediment in this study, stored sediment proved to be less useful in relation to observed biotic diversity as it was better used for monitoring overall sediment increase or decrease, rather than the proportion of sediment habitat available to organisms. The method requires seeking out relatively deep, loose sediment (gravel or smaller factions) on the stream bed and therefore does not accurately reflect habitat diversity nor realistic habitat conditions in which organisms may actually be found, and so no significant relationships were produced when comparing stored sediment with biotic abundance. The difference in results between the site scale and patch scale measures of fine sediment may also show redistribution of fine material within a site. Higher flows would scour both sandy and rocky patches, while low flows deposit preferentially in gravel patches. If a site experienced high flow there may be a decrease in patch embeddedness as fine sediment is removed, but an increase in measured sediment storage due to deposition in gravel patches as the flow receded. However, more detailed measurements would be needed in order to confirm this

As a result, embeddedness proved a more accurate measure for this study as it considers fine sediment as a proportion of the whole patch, and so provides a better idea of habitat availability. Embeddedness is also measured at a patch scale and is easier to correlate directly with organisms found within the patches. Figure 28 a – e shows that many organisms were found within a wide range of embeddedness scores, which is surprising, as it does not appear to correspond with the habitat preferences stated in literature (see Thorp and Rogers, 2010; Brady, 2016; GroundTruth, 2016). Because embeddedness is a measure of fine sediment, it provides no description of the other clasts found in a patch. A patch with high embeddedness may also contain a small area of large bed material, such as cobbles or boulders, allowing the patch to support biota which favour both rocky and sandy habitats. Similarly, a patch with low embeddedness, may still contain enough sediment to support worms or beetles, which favour sandy habitats This is explored further in *Section 5.6*. In general, organisms which favour high sediment conditions seem to be less selective about the level of embeddedness in a patch, as they are able to survive in relatively small amounts of sediment, among larger clasts. This allows them greater flexibility and explains why the bug/beetle group and worms were so common in all three sites. It must also be considered that some organisms, such as crabs, damselflies, bugs/beetles and dragonflies move across the streambed to find food or to hunt, and therefore may be found in patches with higher or lower embeddedness scores than would be expected for the group. The modal scores in Table 21 are more reflective of the expected habitat preferences for each group. A modal score of 3 (25 – 50% embeddedness) was quite common for both fine sediment habitat groups and rocky habitat groups. This is most likely simply because it is the middle-most category, and is therefore an overlap zone for organisms which favour low sediment and high sediment habitats.

5.6 Influence of substrate composition on biotic community structure

Bed material is an important factor in biotic community structure, as it affects what kind of biota will inhabit a patch (Newson & Newson, 2000). Larger clasts create refugia in which larger biota can shelter, allow better mixing of heat, oxygen and nutrients in the water column, and provide an anchor for clinging biota, but also increase the turbulence of the water (Newson & Newson, 2000; Harter, 2001). Smaller clasts provide material in which biota can bury themselves, and a source of food for some biotic groups, but fine sediment can smother the river bed, leading to low oxygen conditions (Harter, 2001).

The RDA (Figure 29) reflects the substrate preferences of taxonomic groups, as fine sediment and small gravel were shown to be significantly drivers of community composition, yet the majority of taxonomic groups were positively correlated with larger clasts (specifically large cobbles and large boulders). Patches which had high embeddedness would be able to support very few organisms due to the less hospitable habitat conditions created by high quantities of fine sediment. In fact, only worms and bugs/beetles were found in patches with 100% embeddedness. Patches with larger clast categories, such as large cobbles and large boulders often had higher substrate diversity and were able to support a greater number of biota. As discussed previously (see *Section 5.5)*, fine sediment habitat groups are able to inhabit relatively small areas of sediment, and so could often be found in small refugia where sediment had been deposited behind boulders and between cobbles, thus making these biota ubiquitous, while other biota were more limited in their distribution by their preference for rocky habitats.

Substrate diversity proved to be a very important factor in biotic community structure, as shown by the Shannon index (H' scores) (Figure 30 a – e). Each set of data revealed a consistent positive correlation between substrate diversity and the number of taxonomic groups found in each patch. This is in agreement with the "habitat heterogeneity hypothesis" described by Tews *et al.* (2004), which assumes that the more complex a habitat is, the greater the diversity of species it can support. The level of substrate diversity, and its correlation to species diversity, varies over time and space, which is reflected in the data collected in the Wilgerbos River. This emphasises the complexity of habitat analysis, and the interplay of multiple factors which determine the presence or absence of biota.

Higher quantities of fine sediment reduce substrate diversity and habitat availability, by filling in spaces between larger clasts (Griffith and Walton, 1978). This was shown in the data for Site 1 in July 2014, which had the lowest biotic diversity per patch, and the highest quantity of fine sediment, as well as thick filamentous algal growth. Site 2 also experienced a decline in overall biotic diversity in March 2015, when fine sediment increased in the site. Organisms such as bugs, beetles and worms, are able to survive in environments such as this, as their diets (algae and organic matter) mean food is readily available in fine sediment patches. Organisms such as caddisflies require flowing water to feed from and large bed material to attach to, and crabs, which require large niches to hide in, will find highly sedimented patches uninhabitable. Furthermore, when in suspension, fine sediment can cause abrasions to biota with gills, such as caddisflies, and the reduction of light penetration slows metabolic rates of organisms, as aquatic biota are unable to regulate their body temperature (Griffith & Walton, 1978; DWAF, 1996).

No correlation can be found between sensitivity scores and either of the physical heterogeneity indices used (Shannon Index nor Hydromorphological Index of Diversity). This is a result of the issue discussed earlier; sensitivity scores are skewed by high sensitivity or low sensitivity biotic groups in a patch and do not consider biotic diversity or abundance. While sensitivity scores provide a useful indicator of habitat quality, they cannot be used as a direct measure of habitat quality. In other words, the presence of a high sensitivity organism, such as other mayflies and stoneflies, indicates a high quality of habitat, whereas low sensitivity organisms, such as worms or leeches, which are hardier and able to survive in a number of different habitat patch types, can be indicative of harsher habitat conditions. But the presence of both high and low sensitivity organisms in a patch should be taken to indicate good habitat quality rather than poor habitat quality. A site-scale sensitivity score was not useful for this study to measure habitat health as it did not correlate to other factors such as substrate and flow heterogeneity.

5.7 General discussion

In summary, the following key points can be extracted from the data analysis:

1. At a site scale, flow is an important driver of habitat condition as it affects sediment dynamics which are significant in dictating biotic community composition.
2. Flow heterogeneity was not shown to be an important factor in biotic diversity – increased heterogeneity did not lead to increased diversity.

3. At a patch scale, depth has a significant relationship with biotic presence and absence, and many taxonomic groups favour shallower water.
4. Velocity was not found to have a significant relationship with biotic community, but through its influence on substrate and water chemistry, some biotic groups showed clear preferences for specific flow conditions.
5. Water chemistry is also significant in biotic presence and absence, but the interplay of variables is too complex to identify individual trends and relationships from the data collected.
6. Bed material has a very strong relationship with biotic community composition.
    - Fine sediment is a significant driver of the presence and absence of taxonomic groups. Highly sedimented patches support significantly different communities from patches containing larger clasts, with the majority of groups favouring low sediment conditions.
    - Larger clasts promote greater substrate heterogeneity in a patch and are therefore important for biotic diversity, but do not have a direct relationship with community composition.
    - Substrate heterogeneity, at a patch scale and a site scale, creates more diverse biotic communities.
7. The relationship between flow and fine sediment is indeterminate and low flow is equally likely to cause a build-up of fine sediment as a flood event is.

The relationships observed among the variables measured in this study are represented graphically in Figure 32. While a full habitat assessment would produce a far more complex flow chart, this study emphasised devising a simple methodology which could be easily replicated. Therefore, while habitat condition is a complex subject, a factor of multiple relationships at a number of spatial and temporal scales, Figure 32 simply depicts the findings produced during this study, either through data collection, or through research.

Overall, the Wilgerbos River succeeds in meeting the three criteria for good ecosystem health mentioned in Apitz (2012): the system shows vigour as it has both primary productivity, such as algae and rooted instream vegetation, and secondary productivity, in the many different biota supported by the instream habitats; however, productivity was never quantitatively measured so the actual volume of biomass produced is in this study site unknown.

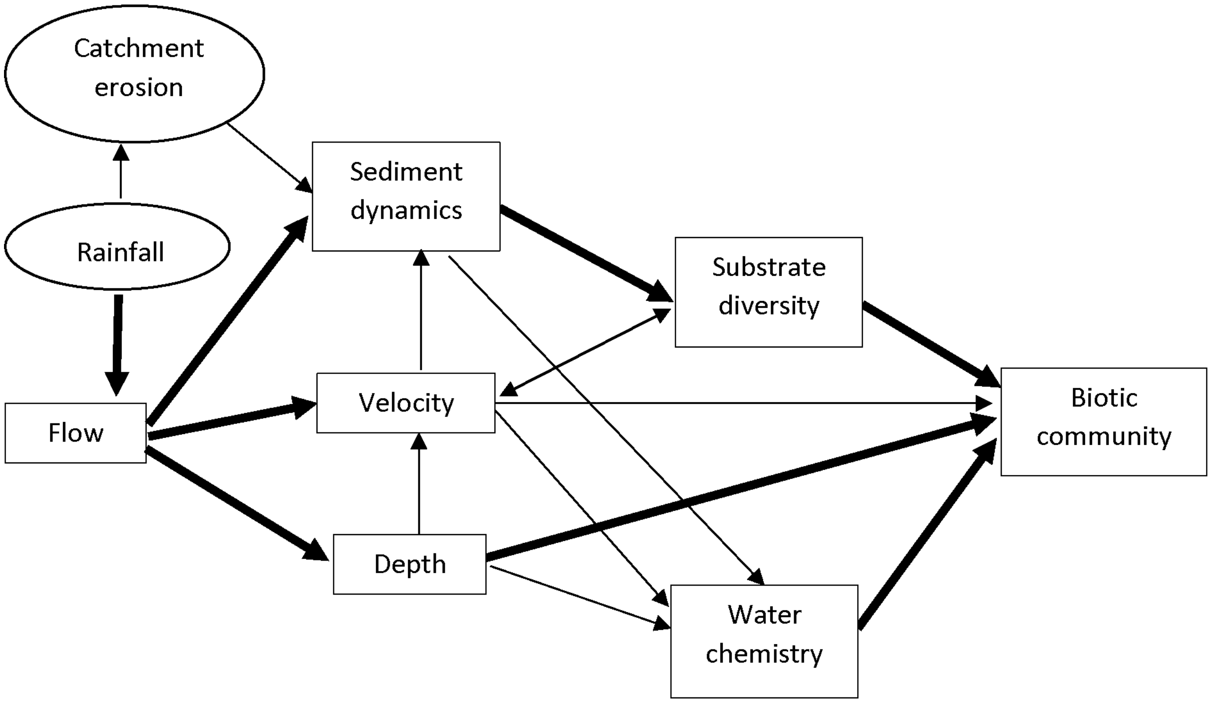


Figure 32: Relationships observed among variables influencing biotic community composition. Thick arrows indicate strong, or statistically significant relationships. Round boxes indicate external factors, square boxes indicate internal factors.

The system also shows a high level of organisation, as a wide diversity of biota was supported, including browsers, filter feeders and predatory macroinvertebrates, as well as fish, tadpoles and otters, which were not measured in the study but whose presence was noted as a point of interest. Finally, the system shows resilience to harsh conditions, which will be discussed in greater detail later in this chapter. According to these components, the river has good habitat health and is able to support complex and diverse biotic communities, even during drought conditions.

The findings discussed above are in line with the two key objectives of this study: they provide a description of spatial and temporal variability in habitat availability and quality and changes in the biotic community, and they investigate and assess the relationship between system drivers (flow and sediment), habitat quality and biotic response. All three sites experienced changes in habitat condition and biotic community composition during the course of this study, but the changes were not uniform, and each site was differently affected by changes in flow. Site 1 reflects a more typical Karoo stream used for abstraction, and therefore is very susceptible to drought. Conversely, Site 2 and 3 are less typical as groundwater-fed streams are uncommon in the Karoo, but they demonstrate the importance of groundwater as a moderating influence on the effects of drought. While biotic diversity did not show a great degree of change over the course of the study, the data does reflect changes in community composition and the abundance of taxonomic groups at different sampling periods. Some taxonomic groups (such as stoneflies and damselflies) showed strong trends in presence and absence at different stages of data collection, while most fluctuated throughout the study. Southwood (1977, cited in Mellado Diaz *et al.*, 2008) suggests that adversity reduces interspecies competition for resources, by limiting the number of species able to survive in a habitat. This is evident to a point in the Wilgerbos River, as taxonomic groups with low sensitivity scores, i.e. hardier and more tolerant of harsh conditions, such as the bug/beetles group, worms and flat worms, were always present, and appeared to be very flexible in terms of flow or substrate composition preferences. The arrival of stoneflies, the most sensitive taxonomic group on the MiniSASS scale, in May 2015 suggests good water quality in that period, and their decrease in abundance as DO concentrations decreased in August 2015 reflects their sensitivity to oxygen content. Seasonality and breeding cycles must also be considered as a factor here, however. The uncommonness of other taxonomic groups such as caddisflies and dragonflies, which have specific habitat preferences, shows the importance of spatial variability and habitat heterogeneity for maintaining biotic diversity. For the most part communities remained constant, with the majority of the taxonomic groups present throughout the study, and the commonness of the other mayfly group throughout the study is indicative of overall high water quality and habitat health. Furthermore, it shows that while adversity is a limiting factor in biotic abundance and richness, organisms which are accustomed to harsh conditions are able to build up a general tolerance and communities may become less susceptible to adversity over time. The data suggest that there is a correlation between physical and chemical habitat properties and biotic communities, and often a direct relationship can be established, especially in the case of variables such as groundwater, sediment dynamics and substrate which had the greatest influence on the results of this study.

In this study flow was an important factor in determining habitat composition and quality. Changes in flow had an influence on all the other variables measured, reinforcing Poff and Zimmerman's (2010) argument that flow is the 'master variable', and Bunn and Arthington's (2002) statement that flow is key in determining physical habitat structure. Although biotic presence and absence was not significantly related to velocity, Figure 22 showed that a number of organisms were closely associated with changes in velocity. By inference, this suggests that the role of velocity is not direct, but rather the structural and chemical changes implemented by velocity, such as changes in substrate and sediment dynamics, and mixing of nutrients, heat and oxygen are what dictate the presence or absence of taxonomic groups, rather than velocity itself. Depth was shown to have a direct relationship with biotic community, as the RDA produced a significant correlation, and a number of taxonomic groups showed an aversion to increasing depth. However, the relationship between velocity and depth is important to consider as shallow water tends to have higher velocity, better mixing and rockier substrate, while deep water has lower velocity, less mixing and sandier substrate. This creates a complicated relationship where the influence of velocity is diluted by other habitat variables which more directly impact on biotic community. However, while the RDA did not produce a statistically significant relationship between presence and absence data and velocity, as shown in Figure 22, the chart does illustrate a close association between the two, suggesting that the influence of velocity on habitat condition is important to biota. Bunn and Arthington (2002) secondly state that biota evolve life histories in response to changes in flow. The time scale of this study did not allow for this statement to be tested; however, as the biota in the Wilgerbos River showed lower sensitivity to fluctuations in water chemistry variables, and as biotic diversity was able to withstand drought conditions, this statement would seem to hold true. The Hydromorphological Index of Diversity scores did not correlate with biotic diversity, as was expected, but a longer study period may reveal a stronger relationship between biota and flow variability. Furthermore, a larger study area may better capture spatial variability in velocity and a larger data set could highlight trends which were not evident in this study.

The direct role of geomorphic substrate and sediment in habitat structure was clear in the study. As shown by the Shannon Index scores, habitat patches with greater substrate heterogeneity were able to support a greater variety of organisms, and patches with fine sediment as the dominant substrate type tended to have the poorest biotic diversity. Fine sediment in this system had a strongly significant relationship with biota. While creating habitats for some taxonomic groups such as worms and beetles, fine sediment often precluded taxonomic groups from inhabiting patches, filling up spaces between gravels and cobbles which would normally provide habitats for other organisms. Fine sands and silts settle out in low gradient areas and thickly cover the stream bed creating a dense layer which covers all other substrates beneath it, fully occupying niches and potential refugia. This suggests that fine sediment is a significant habitat driver, with both positive and negative influences on biotic community. This has interesting implications in terms of disturbance in the system. The presence of the weir above Site 1 has a direct impact on the system. It was constructed for the purpose of water abstraction, but its capacity for sediment storage may also have a positive influence on the downstream environment, as described by Hauer and Lorang (2004), by reducing the sediment load and promoting greater substrate heterogeneity, in turn supporting a greater diversity of biota. The converse of this argument, as stated by Apitz (2012), is that disturbance can cause changes in habitat and water quality which can hamper functionality in the long term. In this case, preventing longitudinal transportation of fine sediment increases the erosive capacity of the stream, which may result in greater sediment loads being produced downstream of the weir and changes in lateral and vertical channel shape. However, there is currently little evidence of extreme erosion in the reaches directly below the weir since its reconstruction in May 2012, which suggests that the weir has had very little negative impact on the stream itself. This may change in the long term, and may need to be monitored.

The relationship between flow and sediment is indeterminate, as the two measures of fine material produced contrasting results under different flow conditions. The site scale measurement of stored bed sediment showed an increase in sediment quantities during low flow (March 2015), while the patch scale measurement of embeddedness showed a decrease in sediment quantities during low flow. The potential for this to be due to differences in sampling technique has been noted, but it is also possible that the difference shows the redistribution of sediment with changes in flow condition (see *Section 5.5*). Furthermore, patch scale analysis shows that embeddedness is equally likely to increase or decrease following a flood event. Embeddedness decreased notably during the drought, then increased following the flood event in May 2015 to produce almost even distribution of sandy and rocky type patches. However, following the flood event in August 2015 embeddedness decreased again. This indicates that some flood events cause more scouring, while others cause more deposition, and the point at which a flood event transports more sediment than it removes would also be valuable for future studies, given the frequency of flood events in the Karoo region. This type of information would require long term measurements which were beyond the scope of this study.

This research has potential application in terms of water management in semi-arid regions, by creating a baseline profile of a Karoo stream in fair to good condition. Furthermore, studies such as this one can begin to fill the knowledge gap around modelling semi-arid stream biotic communities with the same level of detail as in perennial waters. The drought conditions in March 2015 highlight the importance of groundwater in maintaining habitat condition, and more thorough measurements of groundwater levels would be strongly recommended for future studies such as this. However, the drought also made it difficult to create an accurate baseline for normal conditions in the Wilgerbos River. Therefore, this study is more useful as a baseline of biotic response to and recovery after drought in a semi-arid groundwater-fed stream system.

Naturally, as groundwater is an important characteristic in two of the three sites sampled in this study, the findings are relevant to other groundwater-fed streams, but not necessarily to the entire Karoo region. The methods used in this study can easily be applied to any system, and can be useful both for monitoring natural changes in response to seasonal disturbance, and for assessing long term changes in response to anthropogenic disturbances such as farm dams and abstraction.

Like the Karoo, semi-arid regions across the world are water stressed, and in many countries they support communities, as well as farm- and grazing lands, such as northwest America, India and the Middle East (Parr *et al.*, 1990), and so channel modification is largely unavoidable in order to provide a reliable supply of water. However, it is important to constantly monitor the condition of the stream, and to manage the water effectively both upstream and downstream of any constructions or abstraction points so as to ensure the water supply is sustainable and long lasting. While aspects of the data analysis used in this study require a scientific background, many of the in-field data collection methods such as MiniSASS, clast measurements and sediment sampling do not require intensive training or experience and can be easily applied to simply monitor physical changes and biotic response over time and space.

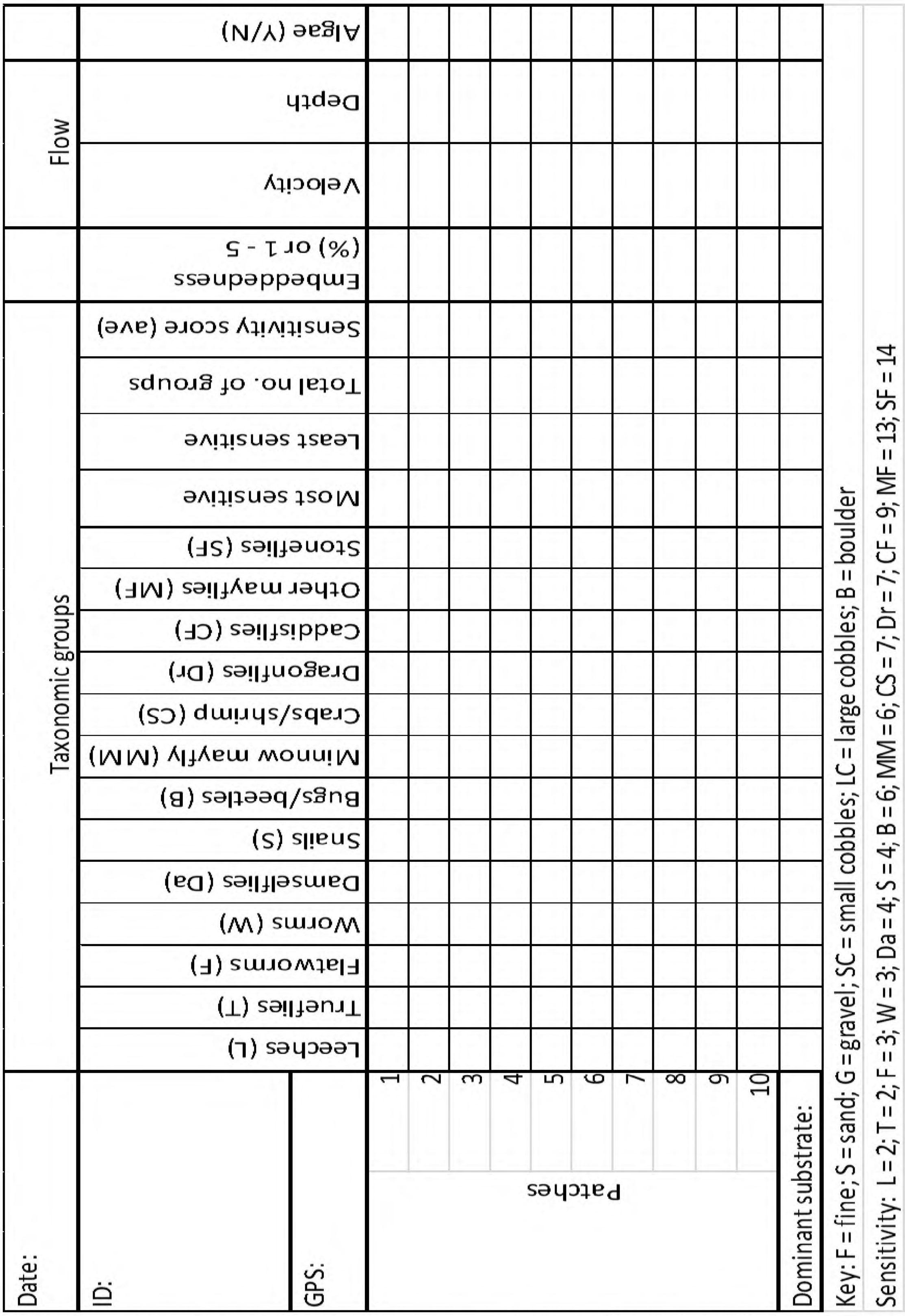
The methods used in this study were successful in achieving the two key objectives laid out, as well as the secondary objective, of assessing the usefulness of MiniSASS as a measure of habitat condition. The MiniSASS method has shown to be effective for data collection, and individual taxonomic groups were useful indicators of overall habitat condition, but the sensitivity score system is not ideal for habitat assessments as it prioritises biotic sensitivity at the cost of biotic diversity. The health category scores could be adapted (similar to the system used for SASS) to account for the number of taxonomic groups rather than just the sensitivity of the species found, or else a new scoring system could be devised to rate habitat quality as a factor of biotic diversity as well as sensitivity.

The findings of this study have identified some significant drivers of biotic diversity and a proposed layout is presented for a simplified habitat health assessment sheet (Figure 33), which incorporates the most important factors for habitat structure. While this sheet does not yet include a scoring or rating system, it does provide a grounding from which a scoring system could be devised. The main reason for including MiniSASS in this study was to work towards a simplified method of habitat assessment, similar to the way MiniSASS is used as a simplified method of measuring water quality. The methodology has been employed in citizen science, allowing school children to collect data as part of projects and field trips, generating a large amount of data on rivers across South Africa (Graham *et al.*, 2010).

The value of citizen science is therefore in the quantity of data it can generate, but as the data often comes from people with little to now scientific background, it is important to have clear and simple methodologies, with no room for misunderstandings. Areas such as the Karoo are home to a number of farmers and small towns which rely heavily on river systems for water. There is a lot of potential for the implementation of citizen science in areas such as this, where farmers and community members could be encouraged to conduct the assessment on a 3 to 6-month basis, to monitor their water resources and their potential impact on river condition.

The assessment makes use of the patch system used in this study, but notes the substrate per taxonomic group, rather than the overall substrate composition of the patch. The substrate key codes given at the bottom of the data sheet would be written into the block for each taxonomic group in each patch. This is in an effort to create a more detailed and specific preferred substrate profile for each group. The dominant substrate for each group is also recorded, based on which substrate type was most common for each group across the patches. This is in the same vein as the modal embeddedness scores used in this study, in order to identify inconsistencies or anomalies. The overall embeddedness per patch is also recorded to give an indication of the amount of sediment on the river bed.

The sheet makes use of the sensitivity scores, but also accounts for the total number of groups per patch, in an attempt to overcome the challenges of using sensitivity scoring to assess habitat. Until a more reflective scoring system can be devised, the sensitivity score does provide an indication of the community composition in the site, i.e. low scores indicate a larger proportion of low scoring groups, and high scores indicate larger proportions of high scoring groups. However, the ecological condition score categories which usually accompany the sensitivity scores should be rejected, as they are not useful at a patch scale and may provide misleading information about the health of the habitat. Flow variables are included, as depth was a significant driver of biotic community composition, and velocity is a driver of sediment dynamics which affect habitat structure and availability. Finally, the presence of algae should be recorded as it has a number of impacts on water quality and habitat availability, which should be accounted for when assessing the presence or absence of biota in a habitat patch. As algae was not quantitatively measured in this study, this aspect of the score sheet requires further study and work to derive a method of quickly but effectively measuring the amount of algae in a stream.



Date:	Taxonomic groups																		Flow		
ID: GPS:	Leeches (L)	Truefilies (T)	Flatworms (F)	Worms (W)	Damselflies (Da)	Snails (S)	Bugs/beetles (B)	Minnow mayfly (MM)	Crabs/shrimp (CS)	Dragonflies (Dr)	Caddisflies (CF)	Other mayflies (MF)	Stoneflies (SF)	Most sensitive	Least sensitive	Total no. of groups	Sensitivity score (ave)	Embeddedness (%) or 1 - 5	Velocity	Depth	Algae (Y/N)
Patches 1																					
2																					
3																					
4																					
5																					
6																					
7																					
8																					
9																					
10																					
Dominant substrate:																					

Key: F = fine; S = sand; G = gravel; SC = small cobbles; LC = large cobbles; B = boulder

Sensitivity: L = 2; T = 2; F = 3; W = 3; Da = 4; S = 4; B = 6; MM = 6; CS = 7; Dr = 7; CF = 9; MF = 13; SF = 14

Figure 33: Proposed layout for rapid habitat assessment sheet based on the most key habitat variables identified in this study.

5.8 Recommendations

The original intention of this study had been to visit the sites on a seasonal basis to monitor habitat changes in response to seasonal weather fluctuations characteristic of a summer rainfall region. However, the weather patterns did not follow the normal seasonality of summer rainfall and site visits became irregular in order to account for this, and so the study was amended to instead monitor habitat changes in response to general changes in flow and flow disturbance. If this study were to be extended or repeated it would be improved by doubling the time frame for field data collection. As Newson and Newson (2000) state, it is essential that ecosystem health be studied at both a spatial and temporal scale to account for a full range of flow variability. It would have been very valuable to gather data over a 12-month period before or after the drought to establish a baseline of other flow conditions with which to compare the drought conditions, and to better represent the variability that is characteristic of Karoo rivers. Similarly, it would have been interesting to measure the effects of a large-scale flood event, in contrast to drought, which is not uncommon to the region. A longer time frame would have allowed more opportunity for such measurements or events to occur. In addition, it would be valuable to expand the area of study, so as to include a second stream which is not groundwater-fed which could be compared with the groundwater-fed Wilgerbos River. Nongroundwater-fed streams are more typical of the Karoo and the ability to compare the two would most likely even further emphasise the moderating effect of groundwater. Boreholes on surrounding farms could be used effectively to measure the depth of the water table over time, allowing the groundwater supply to be monitored as well as the instream conditions. For streams which are heavily used as a source of water, the rate of abstraction could also be measured, which would add a resource management aspect to this study.

The data produced in this study indicates that the river recovered almost immediately after the drought, but whether this is only because of groundwater is unknown. Nonetheless, the system's ability to maintain functionality under stressful conditions shows a high level of resilience, which Apitz (2012) considers to be an indicator of ecosystem health. It is possible that non-perennial stream biota have not only evolved to suit the harsher conditions of semi-arid environments (Mellado-Diaz *et al.*, 2008), but also to be hardier and quicker to re-establish communities after disturbances, which are more common in water stressed regions than in humid, perennial systems. However, this hypothesis requires further study, as the rate of recovery in more seasonal or ephemeral streams may be far slower. An in-depth measurement of groundwater processes and changes in groundwater level would be a valuable line of study to follow were this research to be expanded, as it has proven to be a significant factor in biotic community maintenance. Researchers with SASS training and accreditation may also find a full SASS assessment more accurate as a habitat indicator, than MiniSASS; however, this would require experience with biomonitoring as the full SASS methodology is a more complex process than the simplified methodology used in this study. A final recommendation would be the inclusion of a submerged DO meter to measure continuously, alongside the level loggers, the rapid oxygen fluctuations as described by Pinder and Friet (1994), which could then be compared with changes in temperature and water level at multiple time scales. This would help to create a clearer idea of how oxygen concentrations influence biotic communities, and even individual groups.

As much of the literature previously referred to suggests, an interdisciplinary team is valuable when conducting a habitat quality assessment. This study integrated a number of aspects of stream ecology (hydrology, geology, ecology, and geomorphology), which inevitably leads to a more superficial investigation of each aspect, especially when under a time limit. An interdisciplinary team could provide closer and more detailed study of the different aspects. For example, an ecologist would be able to provide knowledge on the life cycles, which may account for the presence or absence of certain taxa at certain times, and a geologist would have knowledge of groundwater and the influence of underlying geologies on water chemistry. However, specific training should not be necessary for habitat assessments, and this study is evidence that methods such as MiniSASS and the disturbance method for measuring stored sediment can be effectively used to assess habitat quality without a background in ecology or geomorphology, and provide an opportunity for citizen science to augment scientific studies.

CHAPTER 6: CONCLUSION

This study investigated the effects of flow, water quality variables and sediment processes on habitat availability and biotic community structure in a groundwater-fed system in the Karoo, South Africa. The primary objectives of the study were to describe the spatial and temporal changes in instream habitat condition and availability, and biotic community composition, and to investigate the influence of flow and sediment, as system drivers, on habitat quality and in turn biotic response to habitat changes. As a secondary objective, the MiniSASS methodology was tested as a potential measure of habitat quality, to create a quick and easy way to conduct habitat assessments. Data collection was divided into three categories: site properties, habitat variables (including flow, water chemistry, sediment and substrate) and biotic sampling (presence and absence of taxonomic groups, and MiniSASS sampling). A number of statistical methods were used to assess the relationships between each habitat variable and biotic presence and absence, and the sensitivity scores derived from the MiniSASS assessments. The MiniSASS methodology was shown to be useful for sampling, but sensitivity scores were not reflective of habitat quality. Having identified substrate, fine sediment, flow and depth as variables which have a strong influence on habitat condition and biotic community composition, a proposed outline for a rapid habitat assessment data sheet was created, based on the MiniSASS sample method.

The Karoo region is generally recognised as a summer rainfall region, but does receive rainfall in winter, often in small, isolated showers. Rainfall at any time of the year is in the form of intense events with a short duration, but the semi-arid environment is sensitive to these events and river systems are often quick to respond to rainfall input. Lack of vegetation cover in catchments promotes surface run-off and, as a result, soil erosion which transports sediment from the land into the channel. This creates non-perennial or seasonal stream systems which are prone to ‘flashy’ flow and large sediment loads. The region is water scarce and vulnerable to drought, but also experiences flash flooding during large rainfall events. Natural disturbance is therefore a major feature of the Karoo, and in general of semi-arid river systems. However, the Karoo is also an important livestock farming region where many sheep, goat and wild game farmers and breeders settle. Therefore, in addition to natural disturbances, hydrological systems are also subjected to anthropogenic disturbance in the form of dams and weirs, water abstraction, and borehole extraction of groundwater. Consequently, river systems in the Karoo are placed under strain, which can, if not properly managed, have negative impacts on instream habitats and the biotic communities which inhabit them.

This study looked into the habitat properties of three stream sections with different gradients, flow dynamics, substrate types, flow inputs and physical situations. The first section (Site 1) has the lowest gradient and, generally, the lowest discharge, and is situated directly downstream of a small weir used for water extraction, which greatly reduces base flow into the site. Site 2 and 3 both have steeper gradients and higher discharges, and are both fed by groundwater (Site 3 to a lesser degree due to transmission losses). Both have a number of riffle-pool systems creating diversity in flow and substrate, and both flow through a gorge. Site 2 lies 3 km downstream of a second weir, and flows along the fence line of farmland, and is severely constrained by a doleritic intrusion on the western edge. Site 3 lies along the upper boundary of a farm and is therefore exposed to very little direct anthropogenic disturbance, but is oriented such that it is exposed to the sun for more hours each day than Site 2. Furthermore, the gorge in this section channels the wind more effectively, and so Site 3 is also more exposed to wind, which contributes to evaporation.

During this investigation, each site was visited five times over the course of 13 months, on an approximately seasonal basis. During each visit, the discharge, water chemistry, stored sediment load and biotic diversity of the three sites were measured. In addition, longitudinal profiles were created and channel width was measured. The intention was to assess the impact of changes in the quality of the instream habitats on the biotic communities, by looking for correlations between the physical data and the biotic data. The results presented in this thesis show connections between the physical variables themselves, and between the physical variables and instream biota. However, the overall condition of the river is good and the system appears to be under low stress. As a result, only minor changes were found in the biotic community composition, even in drought conditions, and recovery time after the drought was quick. A large part of this is due to the moderating influence of groundwater in Site 2 and 3, as these reaches were able to maintain flow throughout the year, regardless of the drought, thus maintaining stable habitats and healthy biotic communities. Site 1 suffered greater impact during the drought, but was able to recover within a month and the biotic communities were restored to full health, which is indicative of good overall river health, and hardy organisms which have adapted to cope with the harsh environment.

While substrate and flow are said to be the primary drivers of habitat (Bunn & Arthington, 2002; Hughes & Louw, 2010; Poff & Zimmerman, 2010) the results of this study suggest that groundwater is also a key variable for maintaining habitat quality in semi-arid environments, increasing their resilience to drought, and decreasing recovery time after drought. Groundwater moderates changes in flow, sediment dynamics and water chemistry during dry seasons, allowing biotic communities to survive throughout the year, as described by Hynes (1983) and Le Maitre *et al.* (2007). However, groundwater-fed systems are not typical of the Karoo region and in that sense the findings of this study cannot be broadly applied to all Karoo streams. This study could be expanded to incorporate a second, surface flow-fed river which could be used as a control for comparison with the groundwater-fed Wilgerbos River. Explicit measurements of groundwater inputs would also be strongly recommended for the future. A longer time frame could allow for a study to be made of normal seasonal conditions and variations in flow and habitat, which could then be compared with the drought conditions observed in this study to show the difference between the regular seasonality of a semi-arid system, and the impacts of a prolonged drought in summer. Despite the limited scope of this study, it provides a useful baseline study of biotic communities in a Karoo stream, showing that great diversity can be supported in a reach and that biota are resilient to natural disturbance which is common to the area and have higher tolerance for harsher habitat conditions (DWAF, 1996). Greater substrate diversity in a habitat patch is directly correlated with biotic diversity. This is in line with research by Newson and Newson (2000), Harter (2001) and Apitz (2012) into the importance of substrate diversity for habitat availability and biotic community. Flow diversity within a reach did not correlate with biotic diversity, suggesting that substrate diversity has a more direct impact on the biotic community than flow. This does not mean that flow does not have an impact on the community, but rather that the effects of flow are indirectly felt by biota, primarily through the effect flow has on substrate, or physical habitat structure, as described by Bunn and Arthington (2002). Furthermore, this study showed that anthropogenic disturbance can have a positive impact, as the weir above Site 1 traps large quantities of sediment which has promoted substrate diversity within the site. However, it may also increase the erosive capacity of the river in the future. The influence of groundwater is also important for bed sediment, as it does not contribute fine sediment, whereas surface flow in semi-arid environments introduces large quantities of sediment from the land. Further studies into groundwater would be valuable to water management studies in this area in the future.

Part of the methodology of this study included assessing the usefulness of the MiniSASS sensitivity scoring system as a measure of habitat health, rather than water quality as it is commonly used for. For the most part the results were valuable and in many patches the sensitivity scores of individual biota were useful indicators of habitat health. However, there were cases where a single, high sensitivity organism was found in a patch with a low Shannon diversity index score, which produced conflicting results at a site scale, suggesting that average sensitivity scores do not necessarily correlate with habitat health. Therefore, while the MiniSASS taxonomic grouping system is useful for a broad assessment of biotic diversity within a patch, the sensitivity score should be used primarily as an indicator of water quality and sparingly as an indicator of habitat health. In general, habitat health can be quite easily assessed by simply correlating substrate diversity with the number of taxonomic groups found, but a rigorous sampling method must be employed to sample a wide range of habitat patches to ensure the highest possible reliability. While flow is clearly essential for creating and maintaining instream habitats, and has a direct influence on substrate composition and sediment transportation or deposition, biotic diversity can be maintained even in low flow conditions, as experienced in Site 2 and 3 during the drought. Similarly, low flow diversity was able to maintain equally high numbers of taxonomic groups as high flow diversity and, as Site 1 was able to maintain good biotic diversity despite having significantly reduced flow, it is clear that the Wilgerbos River is perfectly able to support healthy biotic communities even in adverse conditions. However, higher abundance of organisms was observed in the two unaltered sites, which suggests that human influence does have an impact on biotic communities.

Finally, the inclusion of an interdisciplinary team would be highly recommended for similar studies in the future. A number of previous studies, like this one, were undertaken by teams with the same or similar professional backgrounds, such as Hauer and Lorang's (2004) work in western USA, Oliva-Paterna *et al.*'s (2003) work in Spain, and Meyer *et al.*'s (2003) work in Germany, all of which specialised in ecology or biological sciences. This can lead to knowledge gaps which are either not identified at the time, or are not fully resolved because of a lack of experience with the area. Interdisciplinarity would mitigate this by covering a wide range of knowledge areas and professional fields, and would encourage the study to be more comprehensive by accounting for a greater variety of variables.

In conclusion, this study was successful in achieving the aim and objectives set out at the start, as it assessed which habitat variables were most important for biotic community composition and thus provided a baseline for future studies in semi-arid stream systems. The study also determined that, while the MiniSASS sampling methodology was useful for sampling biotic communities, the sensitivity scores were not useful as a measure of habitat condition. However, this research would benefit from an expanded study area and a longer period of time. While the results did not show the expected degree of seasonal biotic variability, the data produced were useful in providing a better understanding of Karoo river systems, particularly of the importance of groundwater in habitat health. The potential to use a study such as this in a management capacity has already been discussed, and the inclusion of a second river system would further its usefulness by creating a baseline for both groundwater-fed and surface flow-fed systems, both of which need to be effectively managed. While the Wilgerbos River is in good condition and has not been greatly affected by abstraction practices, many other streams in the Karoo are less well maintained and have been severely altered by poorly placed farm dams, land degradation through overstocking, and the development of erosional features such as gullies (Foster & Rowntree, 2012; Foster *et al.*, 2012). Management of systems such as these is not only important for the maintenance of river health, but also for those who rely on them as a source of good quality water, or those who live downstream of large farms which extract large amounts of water. The lack of knowledge regarding semi-arid system instream habitats can easily be addressed, and the field methods used in this study could be used without extensive training or a scientific background. Therefore, there is a potential to make use of citizen science in the Karoo, which would generate a large amount of data which could be used to create more accurate and reliable models for managing semi-arid systems in the future.

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APPENDIX 1:
Raw data used in CANOCO RDA (July 2014)

Site ID	**Ecological data set (1 = present, 0 = absent)**												
	Leech	Truefly	Flatworm	Worm	Damselfly	Snail	Bug/ beetle	Minnow mayfly	Crab/ shrimp	Dragonfly	Caddisfly	Other mayfly	Stonefly
Site 1 p1	0	0	0	0	0	0	0	0	0	0	1	0	0
Site 1 p2	1	0	0	0	1	0	1	0	0	0	0	1	0
Site 1 p3	1	0	1	0	0	0	0	0	0	0	0	0	0
Site 1 p4	0	0	0	0	1	0	0	0	0	0	0	1	0
Site 1 p5	0	0	1	1	1	0	0	0	0	0	0	0	0
Site 1 p6	0	0	0	0	1	0	1	0	0	0	0	0	0
Site 1 p7	0	0	0	0	0	0	1	1	0	1	0	0	0
Site 1 p8	0	0	0	0	0	0	0	1	0	0	0	1	0
Site 1 p9	0	0	0	0	0	1	0	0	1	0	1	1	0
Site 1 p10	0	0	0	0	0	0	0	0	0	0	0	0	0
Site 2 p1	0	0	0	0	1	0	1	0	0	1	0	1	0
Site 2 p2	0	0	0	1	1	0	1	0	0	1	0	0	0
Site 2 p3	0	1	0	1	0	0	1	1	0	0	1	1	0
Site 2 p4	1	0	1	1	0	1	0	0	1	1	0	1	0
Site 2 p5	0	0	0	0	1	0	1	1	0	0	0	1	0
Site 2 p6	0	1	0	1	0	0	1	0	0	0	0	0	0
Site 2 p7	0	0	0	0	1	1	1	0	0	0	0	1	0
Site 2 p8	0	1	0	1	1	0	1	1	0	0	1	0	0
Site 2 p9	0	1	0	1	1	0	1	1	0	0	0	1	0
Site 2 p10	1	1	1	0	0	1	0	1	0	0	1	0	0

	Leech	Truefly	Flatworm	Worm	Damselfly	Snail	Bug/ beetle	Minnow mayfly	Crab/ shrimp	Dragonfly	Caddisfly	Other mayfly	Stonefly
Site 3 p1	0	0	0	1	1	0	1	1	1	0	0	0	0
Site 3 p2	0	0	0	0	0	0	1	0	0	0	0	1	0
Site 3 p3	0	0	0	1	0	0	0	0	0	0	1	1	0
Site 3 p4	0	0	0	1	0	0	0	1	0	0	0	1	0
Site 3 p5	0	0	0	0	0	0	1	0	0	0	1	1	0
Site 3 p6	0	1	0	0	0	0	0	1	0	0	1	0	0
Site 3 p7	1	0	0	1	0	0	0	0	0	0	1	1	0
Site 3 p8	0	0	0	0	0	0	1	0	0	1	0	1	0
Site 3 p9	0	0	0	0	0	1	0	1	0	0	0	0	0
Site 3 p10	0	0	0	1	1	0	0	0	0	1	0	0	0

Site ID	**Site descriptors**								
	Temp (°C)	Nitrate (mg/L)	Soluble Phosphate (mg/L)	DO (% saturation)	Turbidity (ppm)	EC (µS/cm)	pH	Discharge (m^3/s)	Maximum flood peak (m)
Site 1 p1	4,5	<2	0	88,2	11,41	448	8,31	0,094	0,094
Site 1 p2	4,5	<2	0	88,2	11,41	448	8,31	0,094	0,094
Site 1 p3	4,5	<2	0	88,2	11,41	448	8,31	0,094	0,094
Site 1 p4	4,5	<2	0	88,2	11,41	448	8,31	0,094	0,094
Site 1 p5	4,5	<2	0	88,2	11,41	448	8,31	0,094	0,094
Site 1 p6	4,5	<2	0	88,2	11,41	448	8,31	0,094	0,094
Site 1 p7	4,5	<2	0	88,2	11,41	448	8,31	0,094	0,094
Site 1 p8	4,5	<2	0	88,2	11,41	448	8,31	0,094	0,094
Site 1 p9	4,5	<2	0	88,2	11,41	448	8,31	0,094	0,094
Site 1 p10	4,5	<2	0	88,2	11,41	448	8,31	0,094	0,094
Site 2 p1	10,5	<2	0,2	91,5	10,2	470	8,19	0,071	0,118
Site 2 p2	10,5	<2	0,2	91,5	10,2	470	8,19	0,071	0,118
Site 2 p3	10,5	<2	0,2	91,5	10,2	470	8,19	0,071	0,118
Site 2 p4	10,5	<2	0,2	91,5	10,2	470	8,19	0,071	0,118
Site 2 p5	10,5	<2	0,2	91,5	10,2	470	8,19	0,071	0,118
Site 2 p6	10,5	<2	0,2	91,5	10,2	470	8,19	0,071	0,118
Site 2 p7	10,5	<2	0,2	91,5	10,2	470	8,19	0,071	0,118
Site 2 p8	10,5	<2	0,2	91,5	10,2	470	8,19	0,071	0,118
Site 2 p9	10,5	<2	0,2	91,5	10,2	470	8,19	0,071	0,118
Site 2 p10	10,5	<2	0,2	91,5	10,2	470	8,19	0,071	0,118
Site 3 p1	14,3	<2	0,2	103,1	10,58	440	8,54	0,112	0,112
Site 3 p2	14,3	<2	0,2	103,1	10,58	440	8,54	0,112	0,112

	Temp (°C)	Nitrate (mg/L)	Soluble Phosphate (mg/L)	DO (% saturation)	Turbidity (ppm)	EC (µS/cm)	pH	Discharge (m^3/s)	Maximum flood peak (m)
Site 3 p3	14,3	<2	0,2	103,1	10,58	440	8,54	0,112	0,112
Site 3 p4	14,3	<2	0,2	103,1	10,58	440	8,54	0,112	0,112
Site 3 p5	14,3	<2	0,2	103,1	10,58	440	8,54	0,112	0,112
Site 3 p6	14,3	<2	0,2	103,1	10,58	440	8,54	0,112	0,112
Site 3 p7	14,3	<2	0,2	103,1	10,58	440	8,54	0,112	0,112
Site 3 p8	14,3	<2	0,2	103,1	10,58	440	8,54	0,112	0,112
Site 3 p9	14,3	<2	0,2	103,1	10,58	440	8,54	0,112	0,112
Site 3 p10	14,3	<2	0,2	103,1	10,58	440	8,54	0,112	0,112

Site ID	**Patch descriptors**				
	Embeddedness and substrate			**Flow variables**	
	Largest clast	Code	Area occupied by fine material (%)	Velocity (m/s)	Depth (mm)
Site 1 p1	S. Cobble	5	70	0,01	43
Site 1 p2	M. Gravel	3	80	0,01	72
Site 1 p3	S. Cobble	5	20	0,23	104
Site 1 p4	S. Boulder	8	3	0,1	80
Site 1 p5	S. Boulder	8	5	0,22	153
Site 1 p6	M. Cobble	6	30	0	64
Site 1 p7	S. Boulder	8	70	0,02	161
Site 1 p8	L. Boulder	9	25	0,11	159
Site 1 p9	M. Cobble	6	10	0,2	134
Site 1 p10	S. Gravel	2	95	0	223
Site 2 p1	L. Gravel	4	0	0,12	165
Site 2 p2	S. Boulder	8	80	0,32	271
Site 2 p3	S. Boulder	8	2	0,41	64
Site 2 p4	S. Boulder	8	40	0,11	104
Site 2 p5	S. Boulder	8	35	0	187
Site 2 p6	S. Gravel	2	80	0	101
Site 2 p7	S. Boulder	8	10	0,1	204
Site 2 p8	M. Gravel	3	2	0,24	177
Site 2 p9	L. Boulder	2	70	0,04	43
Site 2 p10	S. Boulder	8	40	0,21	252
Site 3 p1	S. Cobble	5	40	0,1	120

	Largest clast	Code	Area occupied by fine material (%)	Velocity (m/s)	Depth (mm)
Site 3 p2	S. Gravel	2	60	0,01	98
Site 3 p3	S. Boulder	8	20	0,31	73
Site 3 p4	S. Boulder	8	15	0,14	182
Site 3 p5	M. Cobble	6	55	0	253
Site 3 p6	L. Cobble	7	0	0,27	88
Site 3 p7	S. Boulder	8	10	0,2	172
Site 3 p8	S. Boulder	8	90	0,1	86
Site 3 p9	S. Boulder	8	10	0,27	152
Site 3 p10	S. Boulder	8	45	0,13	265

APPENDIX 1:

Raw data used in CANOCO RDA (October 2014)

Site ID	**Ecological data set (1 = present, 0 = absent)**												
	Leech	Truefly	Flatworm	Worm	Damselfly	Snail	Bug/ beetle	Minnow mayfly	Crab/ shrimp	Dragonfly	Caddisfly	Other mayfly	Stonefly
Site 1 p1		0	1	1	0	0	0	1	0	1	1	0	0
Site 1 p2	0	0	0	1	0	0	0	0	0	0	0	0	0
Site 1 p3	1	0	1	1	1	0	1	0	1	0	1	1	0
Site 1 p4	0	0	1	1	0	1	0	1	1	1	1	1	0
Site 1 p5	0	0	0	0	0	0	1	0	1	1	1	0	0
Site 1 p6	0	0	0	1	0	1	1	0	1	0	0	0	0
Site 1 p7	0	1	1	1	0	1	0	1	0	0	1	1	0
Site 1 p8	0	0	1	1	1	1	1	1	0	0	1	1	0
Site 1 p9	0	0	0	0	0	1	0	0	0	0	0	0	0
Site 1 p10	0	0	0	1	0	1	0	1	0	0	1	1	0
Site 2 p1	0	0	0	1	0	0	1	0	0	1	0	0	0
Site 2 p2	0	0	1	1	1	1	1	1	0	1	1	0	0
Site 2 p3	0	0	0	1	0	1	0	1	1	0	0	1	0
Site 2 p4	0	0	0	0	1	0	1	0	0	0	1	0	0
Site 2 p5	0	0	0	1	0	1	0	1	1	0	0	0	0
Site 2 p6	0	0	0	1	0	0	1	0	0	1	0	0	0
Site 2 p7	0	0	0	1	1	1	1	1	0	0	1	1	0
Site 2 p8	0	0	0	0	0	1	1	1	0	0	1	1	0
Site 2 p9	0	0	0	0	0	0	0	1	0	1	0	1	0
Site 2 p10	0	1	1	1	0	0	0	0	1	1	0	1	0

	Leech	Truefly	Flatworm	Worm	Damselfly	Snail	Bug/ beetle	Minnow mayfly	Crab/ shrimp	Dragonfly	Caddisfly	Other mayfly	Stonefly
Site 3 p1	0	0	0	1	0	0	0	0	0	0	0	0	0
Site 3 p2	0	0	1	0	0	0	0	0	0	1	0	1	0
Site 3 p3	0	0	1	1	0	0	1	1	0	1	0	0	0
Site 3 p4	0	0	1	1	0	0	0	1	0	1	1	1	0
Site 3 p5	0	0	0	1	0	0	0	0	0	1	0	1	0
Site 3 p6	0	0	0	1	1	0	0	0	1	1	1	1	0
Site 3 p7	0	0	1	1	0	0	0	1	0	0	1	1	0
Site 3 p8	0	0	1	1	0	0	1	1	0	0	1	0	0
Site 3 p9	0	0	1	1	1	0	0	0	0	1	1	1	0
Site 3 p10	0	0	0	1	0	0	0	0	0	1	1	1	0

Site ID	**Site descriptors**								
	Temp (°C)	Nitrate (mg/L)	Soluble Phosphate (mg/L)	DO (% saturation)	Turbidity (ppm)	EC (µS/cm)	pH	Discharge (m^3/s)	Maximum flood peak (m)
Site 1 p1	15,2	<2	0,5	95,6	8,93	530	8,62	0,016	0,094
Site 1 p2	15,2	<2	0,5	95,6	8,93	530	8,62	0,016	0,094
Site 1 p3	15,2	<2	0,5	95,6	8,93	530	8,62	0,016	0,094
Site 1 p4	15,2	<2	0,5	95,6	8,93	530	8,62	0,016	0,094
Site 1 p5	15,2	<2	0,5	95,6	8,93	530	8,62	0,016	0,094
Site 1 p6	15,2	<2	0,5	95,6	8,93	530	8,62	0,016	0,094
Site 1 p7	15,2	<2	0,5	95,6	8,93	530	8,62	0,016	0,094
Site 1 p8	15,2	<2	0,5	95,6	8,93	530	8,62	0,016	0,094
Site 1 p9	15,2	<2	0,5	95,6	8,93	530	8,62	0,016	0,094
Site 1 p10	15,2	<2	0,5	95,6	8,93	530	8,62	0,016	0,094
Site 2 p1	14,1	<2	0,5	90,1	9,38	572	8,66	0,144	0,118
Site 2 p2	14,1	<2	0,5	90,1	9,38	572	8,66	0,144	0,118
Site 2 p3	14,1	<2	0,5	90,1	9,38	572	8,66	0,144	0,118
Site 2 p4	14,1	<2	0,5	90,1	9,38	572	8,66	0,144	0,118
Site 2 p5	14,1	<2	0,5	90,1	9,38	572	8,66	0,144	0,118
Site 2 p6	14,1	<2	0,5	90,1	9,38	572	8,66	0,144	0,118
Site 2 p7	14,1	<2	0,5	90,1	9,38	572	8,66	0,144	0,118
Site 2 p8	14,1	<2	0,5	90,1	9,38	572	8,66	0,144	0,118
Site 2 p9	14,1	<2	0,5	90,1	9,38	572	8,66	0,144	0,118
Site 2 p10	14,1	<2	0,5	90,1	9,38	572	8,66	0,144	0,118
Site 3 p1	20,2	<2	0,3	108	10,12	547	8,92	0,059	0,112
Site 3 p2	20,2	<2	0,3	108	10,12	547	8,92	0,059	0,112

	Temp (°C)	Nitrate (mg/L)	Soluble Phosphate (mg/L)	DO (% saturation)	Turbidity (ppm)	EC (µS/cm)	pH	Discharge (m^3/s)	Maximum flood peak (m)
Site 3 p3	20,2	<2	0,3	108	10,12	547	8,92	0,059	0,112
Site 3 p4	20,2	<2	0,3	108	10,12	547	8,92	0,059	0,112
Site 3 p5	20,2	<2	0,3	108	10,12	547	8,92	0,059	0,112
Site 3 p6	20,2	<2	0,3	108	10,12	547	8,92	0,059	0,112
Site 3 p7	20,2	<2	0,3	108	10,12	547	8,92	0,059	0,112
Site 3 p8	20,2	<2	0,3	108	10,12	547	8,92	0,059	0,112
Site 3 p9	20,2	<2	0,3	108	10,12	547	8,92	0,059	0,112
Site 3 p10	20,2	<2	0,3	108	10,12	547	8,92	0,059	0,112

Site ID	**Patch descriptors**				
	Embeddedness			**Flow**	
	Largest clast	Code	Area occupied by fine material (%)	Velocity (m/s)	Depth (mm)
Site 1 p1	S. Boulder	8	15	0,31	65
Site 1 p2	S. Boulder	8	80	0,01	189
Site 1 p3	S. Gravel	2	80	0,11	120
Site 1 p4	S. Boulder	8	20	0,25	154
Site 1 p5	S. Boulder	8	40	0,02	267
Site 1 p6	S. Boulder	8	46	0,01	287
Site 1 p7	M. Cobble	6	10	0,3	76
Site 1 p8	L. Boulder	9	5	0,29	234
Site 1 p9	S. Cobble	5	99	0	161
Site 1 p10	S. Cobble	5	70	0,01	152
Site 2 p1	M. Gravel	3	60	0,01	55
Site 2 p2	S. Boulder	8	2	0,4	150
Site 2 p3	S. Boulder	8	50	0,11	187
Site 2 p4	Fine	1	50	0	76
Site 2 p5	S. Boulder	8	50	0	190
Site 2 p6	S. Gravel	2	70	0,01	265
Site 2 p7	S. Boulder	8	5	0,36	75
Site 2 p8	M. Cobble	6	20	0,09	304
Site 2 p9	S. Boulder	8	40	0,12	65
Site 2 p10	S. Boulder	8	90	0,25	43
Site 3 p1	S. Cobble	5	95	0,02	100

	Largest clast	Code	Area occupied by fine material (%)	Velocity (m/s)	Depth (mm)
Site 3 p2	L. Boulder	9	50	0,03	89
Site 3 p3	L. Boulder	9	5	0,42	80
Site 3 p4	L. Boulder	9	5	0,33	164
Site 3 p5	S. Boulder	8	40	0,01	260
Site 3 p6	S. Boulder	8	30	0,11	298
Site 3 p7	S. Boulder	8	80	0,25	171
Site 3 p8	S. Boulder	8	18	0,57	57
Site 3 p9	S. Boulder	8	40	0,12	165
Site 3 p10	S. Boulder	8	10	0,09	251

APPENDIX 1:

Raw data used in CANOCO RDA (March 2015)

Site ID	**Ecological data set (1 = present, 0 = absent)**												
	Leech	Truefly	Flatworm	Worm	Damselfly	Snail	Bug/ beetle	Minnow mayfly	Crab/ shrimp	Dragonfly	Caddisfly	Other mayfly	Stonefly
Site 1 p1	0	0	0	0	0	0	0	0	0	0	0	0	0
Site 1 p2	0	0	0	0	0	0	0	0	0	0	0	0	0
Site 1 p3	0	0	0	0	0	1	1	0	0	0	0	0	0
Site 1 p4	0	0	0	1	0	1	1	0	0	0	0	1	0
Site 1 p5	0	0	0	1	0	1	0	0	0	0	0	1	0
Site 1 p6	0	0	0	1	0	1	0	0	0	1	0	1	0
Site 1 p7	0	0	0	0	0	1	0	0	0	0	1	1	0
Site 1 p8	1	1	0	1	0	1	1	0	0	0	0	1	0
Site 1 p9	0	0	0	1	0	0	1	0	0	0	0	0	0
Site 1 p10	1	0	0	1	0	1	1	0	1	0	0	1	0
Site 2 p1	0	0	0	1	0	0	1	0	0	0	0	0	0
Site 2 p2	0	1	0	1	0	0	0	0	1	0	1	0	0
Site 2 p3	0	0	0	1	0	0	1	0	0	0	0	1	0
Site 2 p4	0	0	0	1	1	1	1	0	0	0	1	1	0
Site 2 p5	0	0	0	1	0	1	1	0	0	0	0	0	0
Site 2 p6	0	1	0	1	0	1	1	0	1	0	0	0	0
Site 2 p7	0	1	0	0	0	1	0	1	0	0	0	1	0
Site 2 p8	0	0	0	1	0	0	1	0	0	0	0	0	0
Site 2 p9	0	0	0	1	0	1	0	1	1	1	1	1	0
Site 2 p10	0	0	0	0	0	0	0	0	0	0	0	0	0

	Leech	Truefly	Flatworm	Worm	Damselfly	Snail	Bug/ beetle	Minnow mayfly	Crab/ shrimp	Dragonfly	Caddisfly	Other mayfly	Stonefly
Site 3 p1	0	1	0	0	0	0	1	0	0	0	0	1	0
Site 3 p2	0	0	0	1	0	0	1	0	1	0	0	0	0
Site 3 p3	0	0	0	1	0	0	0	0	1	0	1	1	0
Site 3 p4	0	0	0	1	0	1	1	0	0	1	1	1	0
Site 3 p5	0	0	0	1	0	1	1	0	0	0	0	1	0
Site 3 p6	0	0	0	1	0	1	1	0	0	1	0	1	0
Site 3 p7	0	1	0	1	1	0	1	0	1	0	1	1	0
Site 3 p8	0	0	0	1	0	0	1	0	1	0	1	1	0
Site 3 p9	0	0	0	1	0	0	1	0	1	0	1	1	0
Site 3 p10	0	0	0	1	1	1	0	0	1	1	0	1	0

Site ID	**Site descriptors**								
	Temp (°C)	Nitrate (mg/L)	Soluble Phosphate (mg/L)	DO (% saturation)	Turbidity (ppm)	EC (µS/cm)	pH	Discharge (m^3/s)	Maximum flood peak (m)
Site 1 p1	18,2	<2	0,7	121	20,2	532	9	0	0,094
Site 1 p2	18,2	<2	0,7	121	20,2	532	9	0	0,094
Site 1 p3	18,2	<2	0,7	121	20,2	532	9	0	0,094
Site 1 p4	18,2	<2	0,7	121	20,2	532	9	0	0,094
Site 1 p5	18,2	<2	0,7	121	20,2	532	9	0	0,094
Site 1 p6	18,2	<2	0,7	121	20,2	532	9	0	0,094
Site 1 p7	18,2	<2	0,7	121	20,2	532	9	0	0,094
Site 1 p8	18,2	<2	0,7	121	20,2	532	9	0	0,094
Site 1 p9	18,2	<2	0,7	121	20,2	532	9	0	0,094
Site 1 p10	18,2	<2	0,7	121	20,2	532	9	0	0,094
Site 2 p1	18,9	<2	1,5	81,5	10,5	455	8,24	0,035	0,118
Site 2 p2	18,9	<2	1,5	81,5	10,5	455	8,24	0,035	0,118
Site 2 p3	18,9	<2	1,5	81,5	10,5	455	8,24	0,035	0,118
Site 2 p4	18,9	<2	1,5	81,5	10,5	455	8,24	0,035	0,118
Site 2 p5	18,9	<2	1,5	81,5	10,5	455	8,24	0,035	0,118
Site 2 p6	18,9	<2	1,5	81,5	10,5	455	8,24	0,035	0,118
Site 2 p7	18,9	<2	1,5	81,5	10,5	455	8,24	0,035	0,118
Site 2 p8	18,9	<2	1,5	81,5	10,5	455	8,24	0,035	0,118
Site 2 p9	18,9	<2	1,5	81,5	10,5	455	8,24	0,035	0,118
Site 2 p10	18,9	<2	1,5	81,5	10,5	455	8,24	0,035	0,118
Site 3 p1	16,6	<2	0,7	96,2	14,23	545	8,86	0,015	0,112
Site 3 p2	16,6	<2	0,7	96,2	14,23	545	8,86	0,015	0,112

	Temp (°C)	Nitrate (mg/L)	Soluble Phosphate (mg/L)	DO (% saturation)	Turbidity (ppm)	EC (µS/cm)	pH	Discharge (m^3/s)	Maximum flood peak (m)
Site 3 p3	16,6	<2	0,7	96,2	14,23	545	8,86	0,015	0,112
Site 3 p4	16,6	<2	0,7	96,2	14,23	545	8,86	0,015	0,112
Site 3 p5	16,6	<2	0,7	96,2	14,23	545	8,86	0,015	0,112
Site 3 p6	16,6	<2	0,7	96,2	14,23	545	8,86	0,015	0,112
Site 3 p7	16,6	<2	0,7	96,2	14,23	545	8,86	0,015	0,112
Site 3 p8	16,6	<2	0,7	96,2	14,23	545	8,86	0,015	0,112
Site 3 p9	16,6	<2	0,7	96,2	14,23	545	8,86	0,015	0,112
Site 3 p10	16,6	<2	0,7	96,2	14,23	545	8,86	0,015	0,112

Site ID	Patch descriptors				
	Embeddedness			Flow	
	Largest clast	Code	Area occupied by fine material (%)	Velocity (m/s)	Depth (mm)
Site 1 p1	L. Cobble	7	10	0	0
Site 1 p2	S. Cobble	5	90	0	0
Site 1 p3	Fine	1	100	0	44
Site 1 p4	S. Boulder	8	50	0,01	108
Site 1 p5	S. Boulder	8	0	0,01	266
Site 1 p6	S. Cobble	5	10	0,09	29
Site 1 p7	S. Boulder	8	38	0,01	207
Site 1 p8	S. Boulder	8	15	0	55
Site 1 p9	S. Boulder	8	30	0,01	399
Site 1 p10	S. Boulder	8	5	0,01	531
Site 2 p1	Fine	1	100	0,01	134
Site 2 p2	S. Gravel	2	23	0,04	60
Site 2 p3	S. Boulder	8	5	0,15	108
Site 2 p4	S. Boulder	8	40	0,05	277
Site 2 p5	S. Boulder	8	90	0,01	285
Site 2 p6	S. Boulder	8	30	0,02	369
Site 2 p7	S. Boulder	8	10	0,21	72
Site 2 p8	S. Boulder	8	80	0	111
Site 2 p9	M. Cobble	6	20	0,39	43
Site 2 p10	S. Gravel	2	90	0,19	51
Site 3 p1	S. Boulder	8	65	0,02	26

	Largest clast	Code	Area occupied by fine material (%)	Velocity (m/s)	Depth (mm)
Site 3 p2	S. Gravel	2	30	0,04	102
Site 3 p3	S. Boulder	8	40	0,15	23
Site 3 p4	S. Boulder	8	50	0,04	162
Site 3 p5	S. Boulder	8	50	0,01	239
Site 3 p6	S. Boulder	8	50	0,01	245
Site 3 p7	S. Boulder	8	45	0,07	215
Site 3 p8	S. Boulder	8	10	0,27	56
Site 3 p9	S. Boulder	8	10	0,3	56
Site 3 p10	S. Boulder	8	30	0,1	97

APPENDIX 1:

Raw data used in CANOCO RDA (May 2015)

Site ID	**Ecological data set (1 = present, 0 = absent)**												
	Leech	Truefly	Flatworm	Worm	Damselfly	Snail	Bug/ beetle	Minnow mayfly	Crab/ shrimp	Dragonfly	Caddisfly	Other mayfly	Stonefly
Site 1 p1	0	1	0	1	0	1	0	1	0	1	0	1	1
Site 1 p2	0	1	0	1	1	1	0	0	1	0	1	1	0
Site 1 p3	0	0	0	0	0	1	0	0	0	1	0	1	1
Site 1 p4	1	1	1	1	1	1	0	0	0	0	1	1	1
Site 1 p5	0	0	0	1	1	1	0	0	1	0	0	1	1
Site 1 p6	0	0	0	1	1	1	0	0	0	0	0	1	1
Site 1 p7	0	1	0	1	0	1	1	0	0	1	0	0	0
Site 1 p8	0	1	1	1	1	1	0	0	0	0	1	1	1
Site 1 p9	0	0	0	0	1	1	1	1	1	0	1	1	0
Site 1 p10	0	0	0	1	1	0	0	0	0	0	0	1	0
Site 2 p1	0	0	0	0	0	0	1	0	0	0	0	0	0
Site 2 p 2	1	1	1	1	1	1	1	0	0	1	0	1	0
Site 2 p3	0	0	0	1	1	1	1	1	1	1	1	1	0
Site 2 p4	0	1	1	0	1	1	0	0	0	0	1	1	0
Site 2 p5	0	0	0	1	0	1	0	0	0	0	0	1	0
Site 2 p6	0	0	0	0	0	0	1	0	0	0	0	0	0
Site 2 p7	0	0	0	1	1	1	0	0	0	0	0	1	0
Site 2 p8	0	1	0	1	1	1	1	0	0	1	1	1	0
Site 2 p9	1	0	0	0	1	1	1	0	0	1	0	1	0
Site 2 p10	0	1	0	1	1	0	1	0	1	1	0	1	0

	Leech	Truefly	Flatworm	Worm	Damselfly	Snail	Bug/ beetle	Minnow mayfly	Crab/ shrimp	Dragonfly	Caddisfly	Other mayfly	Stonefly
Site 3 p1	1	0	1	0	1	1	0	0	1	1	0	1	0
Site 3 p2	0	0	0	1	0	0	1	0	0	1	0	0	0
Site 3 p3	0	1	1	0	1	0	0	1	0	0	1	1	0
Site 3 p4	0	0	0	1	0	0	1	0	0	0	0	0	0
Site 3 p5	0	0	0	1	1	0	1	1	0	0	0	1	0
Site 3 p6	1	1	1	1	1	0	1	0	1	0	0	1	0
Site 3 p7	0	0	0	0	1	0	0	0	0	0	1	0	0
Site 3 p8	1	0	0	1	1	0	0	0	0	0	0	1	0
Site 3 p9	1	0	1	1	1	0	0	0	0	0	0	1	0
Site 3 p10	0	0	0	0	1	0	1	0	0	0	0	1	0

Site ID	**Site descriptors**								
	Temp (°C)	Nitrate (mg/L)	Soluble Phosphate (mg/L)	DO (% saturation)	Turbidity (ppm)	EC (µS/cm)	pH	Discharge (m^3/s)	Maximum flood peak (m)
Site 1 p1	10,4	<2	0,2	92,1	3,11	632	7,62	0,006	0,094
Site 1 p2	10,4	<2	0,2	92,1	3,11	632	7,62	0,006	0,094
Site 1 p3	10,4	<2	0,2	92,1	3,11	632	7,62	0,006	0,094
Site 1 p4	10,4	<2	0,2	92,1	3,11	632	7,62	0,006	0,094
Site 1 p5	10,4	<2	0,2	92,1	3,11	632	7,62	0,006	0,094
Site 1 p6	10,4	<2	0,2	92,1	3,11	632	7,62	0,006	0,094
Site 1 p7	10,4	<2	0,2	92,1	3,11	632	7,62	0,006	0,094
Site 1 p8	10,4	<2	0,2	92,1	3,11	632	7,62	0,006	0,094
Site 1 p9	10,4	<2	0,2	92,1	3,11	632	7,62	0,006	0,094
Site 1 p10	10,4	<2	0,2	92,1	3,11	632	7,62	0,006	0,094
Site 2 p1	10,9	<2	0,5	96,2	3,23	651	7,95	0,052	0,118
Site 2 p 2	10,9	<2	0,5	96,2	3,23	651	7,95	0,052	0,118
Site 2 p3	10,9	<2	0,5	96,2	3,23	651	7,95	0,052	0,118
Site 2 p4	10,9	<2	0,5	96,2	3,23	651	7,95	0,052	0,118
Site 2 p5	10,9	<2	0,5	96,2	3,23	651	7,95	0,052	0,118
Site 2 p6	10,9	<2	0,5	96,2	3,23	651	7,95	0,052	0,118
Site 2 p7	10,9	<2	0,5	96,2	3,23	651	7,95	0,052	0,118
Site 2 p8	10,9	<2	0,5	96,2	3,23	651	7,95	0,052	0,118
Site 2 p9	10,9	<2	0,5	96,2	3,23	651	7,95	0,052	0,118
Site 2 p10	10,9	<2	0,5	96,2	3,23	651	7,95	0,052	0,118
Site 3 p1	11,7	<2	0,2	92,6	2,97	594	8,39	0,026	0,112
Site 3 p2	11,7	<2	0,2	92,6	2,97	594	8,39	0,026	0,112

	Temp (°C)	Nitrate (mg/L)	Soluble Phosphate (mg/L)	DO (% saturation)	Turbidity (ppm)	EC (µS/cm)	pH	Discharge (m^3/s)	Maximum flood peak (m)
Site 3 p3	11,7	<2	0,2	92,6	2,97	594	8,39	0,026	0,112
Site 3 p4	11,7	<2	0,2	92,6	2,97	594	8,39	0,026	0,112
Site 3 p5	11,7	<2	0,2	92,6	2,97	594	8,39	0,026	0,112
Site 3 p6	11,7	<2	0,2	92,6	2,97	594	8,39	0,026	0,112
Site 3 p7	11,7	<2	0,2	92,6	2,97	594	8,39	0,026	0,112
Site 3 p8	11,7	<2	0,2	92,6	2,97	594	8,39	0,026	0,112
Site 3 p9	11,7	<2	0,2	92,6	2,97	594	8,39	0,026	0,112
Site 3 p10	11,7	<2	0,2	92,6	2,97	594	8,39	0,026	0,112

Site ID	**Patch descriptors**				
	Embeddedness and substrate			**Flow variables**	
	Largest clast	Code	Area occupied by fine material (%)	Velocity (m/s)	Depth (mm)
Site 1 p1	S. Boulder	8	23	0,01	415
Site 1 p2	S. Boulder	8	10	0,06	57
Site 1 p3	S. Boulder	8	95	0	342
Site 1 p4	M. Cobble	6	40	0,13	40
Site 1 p5	L. Boulder	9	10	0	135
Site 1 p6	L. Cobble	7	5	0	170
Site 1 p7	S. Boulder	8	90	0,01	330
Site 1 p8	S. Boulder	8	2	0,14	30
Site 1 p9	S. Boulder	8	10	0,01	236
Site 1 p10	S. Cobble	5	90	0	225
Site 2 p1	Fine	1	100	0,01	170
Site 2 p 2	S. Boulder	8	20	0,13	174
Site 2 p3	S. Boulder	8	2	0,2	271
Site 2 p4	S. Boulder	8	1	0,35	108
Site 2 p5	L. Boulder	9	60	0	197
Site 2 p6	S. Gravel	2	80	0,01	275
Site 2 p7	S. Boulder	8	70	0	206
Site 2 p8	S. Boulder	8	5	0,26	164
Site 2 p9	L. Cobble	7	20	0	434
Site 2 p10	S. Boulder	8	90	0,04	86
Site 3 p1	S. Boulder	8	70	0	205

	Largest clast	Code	Area occupied by fine material (%)	Velocity (m/s)	Depth (mm)
Site 3 p2	Fine	1	100	0,16	200
Site 3 p3	M. Cobble	6	0	0,02	90
Site 3 p4	S. Boulder	8	80	0,45	97
Site 3 p5	L. Boulder	9	40	0,08	165
Site 3 p6	S. Cobble	5	10	0,01	242
Site 3 p7	L. Boulder	9	10	0,01	189
Site 3 p8	S. Boulder	8	20	0,43	91
Site 3 p9	L. Cobble	7	20	0,06	103
Site 3 p10	S. Boulder	8	35	0,06	77

APPENDIX 1:

Raw data used in CANOCO RDA (August 2015)

Site ID	**Ecological data set (1 = present, 0 = absent)**												
	Leech	Truefly	Flatworm	Worm	Damselfly	Snail	Bug/ beetle	Minnow mayfly	Crab/ shrimp	Dragonfly	Caddisfly	Other mayfly	Stonefly
Site 1 p1	0	1	1	0	0	1	0	0	1	0	1	1	0
Site 1 p2	0	0	0	1	0	1	0	0	0	0	0	1	0
Site 1 p3	0	0	0	0	0	0	0	0	0	0	0	1	0
Site 1 p4	0	1	0	1	0	0	1	0	0	0	1	1	1
Site 1 p5	0	1	1	1	0	1	0	1	0	1	1	1	0
Site 1 p6	0	0	0	1	0	1	0	0	0	0	0	1	0
Site 1 p7	0	0	0	0	0	1	0	1	0	0	1	1	0
Site 1 p8	0	0	0	1	0	1	1	0	0	0	0	1	0
Site 1 p9	0	0	0	1	0	0	0	0	0	0	0	0	0
Site 1 p10	0	0	0	1	0	1	0	0	0	0	0	1	0
Site 2 p1	0	0	0	1	0	0	1	0	0	0	0	0	0
Site 2 p2	1	1	0	0	0	1	1	1	0	0	1	1	0
Site 2 p3	1	1	1	0	0	1	0	1	0	0	1	1	0
Site 2 p4	0	0	0	1	0	1	1	0	0	0	0	1	0
Site 2 p5	0	0	0	1	0	1	0	0	0	0	0	1	0
Site 2 p6	0	0	0	1	0	0	0	1	0	0	0	0	0
Site 2 p7	0	0	0	1	0	1	1	1	1	0	0	1	0
Site 2 p8	0	0	1	0	0	1	1	1	0	0	0	1	1
Site 2 p9	0	1	1	0	0	1	1	1	0	0	1	1	0
Site 2 p10	0	0	0	1	0	0	1	0	0	0	0	0	0

	Leech	Truefly	Flatworm	Worm	Damselfly	Snail	Bug/ beetle	Minnow mayfly	Crab/ shrimp	Dragonfly	Caddisfly	Other mayfly	Stonefly
Site 3 p1	0	0	1	1	0	0	0	0	0	1	1	1	1
Site 3 p2	0	0	0	1	0	0	0	0	0	1	0	1	0
Site 3 p3	0	1	0	0	0	0	0	0	0	0	0	0	0
Site 3 p4	0	0	1	1	0	0	0	1	0	0	1	1	0
Site 3 p5	0	1	0	1	0	1	1	1	0	0	1	1	0
Site 3 p6	0	0	0	1	0	1	1	1	0	0	0	1	0
Site 3 p7	0	1	1	1	0	0	0	1	0	0	1	1	0
Site 3 p8	0	0	0	1	0	1	0	1	0	0	0	1	0
Site 3 p9	1	1	0	1	0	0	0	1	0	1	1	1	1
Site 3 p10	1	0	0	0	0	0	1	1	0	0	0	1	0

Site ID	**Site descriptors**								
	Temp (°C)	Nitrate (mg/L)	Soluble Phosphate (mg/L)	DO (% saturation)	Turbidity (ppm)	EC (µS/cm)	pH	Discharge (m^3/s)	Maximum flood peak (m)
Site 1 p1	9,6	<2	0,1	72,5	2,07	419	8,14	0,032	0,094
Site 1 p2	9,6	<2	0,1	72,5	2,07	419	8,14	0,032	0,094
Site 1 p3	9,6	<2	0,1	72,5	2,07	419	8,14	0,032	0,094
Site 1 p4	9,6	<2	0,1	72,5	2,07	419	8,14	0,032	0,094
Site 1 p5	9,6	<2	0,1	72,5	2,07	419	8,14	0,032	0,094
Site 1 p6	9,6	<2	0,1	72,5	2,07	419	8,14	0,032	0,094
Site 1 p7	9,6	<2	0,1	72,5	2,07	419	8,14	0,032	0,094
Site 1 p8	9,6	<2	0,1	72,5	2,07	419	8,14	0,032	0,094
Site 1 p9	9,6	<2	0,1	72,5	2,07	419	8,14	0,032	0,094
Site 1 p10	9,6	<2	0,1	72,5	2,07	419	8,14	0,032	0,094
Site 2 p1	10,2	<2	0	76,2	2,33	466	8,19	0,118	0,118
Site 2 p2	10,2	<2	0	76,2	2,33	466	8,19	0,118	0,118
Site 2 p3	10,2	<2	0	76,2	2,33	466	8,19	0,118	0,118
Site 2 p4	10,2	<2	0	76,2	2,33	466	8,19	0,118	0,118
Site 2 p5	10,2	<2	0	76,2	2,33	466	8,19	0,118	0,118
Site 2 p6	10,2	<2	0	76,2	2,33	466	8,19	0,118	0,118
Site 2 p7	10,2	<2	0	76,2	2,33	466	8,19	0,118	0,118
Site 2 p8	10,2	<2	0	76,2	2,33	466	8,19	0,118	0,118
Site 2 p9	10,2	<2	0	76,2	2,33	466	8,19	0,118	0,118
Site 2 p10	10,2	<2	0	76,2	2,33	466	8,19	0,118	0,118
Site 3 p1	11,2	<2	0,1	83,9	2,27	451	8,44	0,087	0,112
Site 3 p2	11,2	<2	0,1	83,9	2,27	451	8,44	0,087	0,112

	Temp (°C)	Nitrate (mg/L)	Soluble Phosphate (mg/L)	DO (% saturation)	Turbidity (ppm)	EC (µS/cm)	pH	Discharge (m^3/s)	Maximum flood peak (m)
Site 3 p3	11,2	<2	0,1	83,9	2,27	451	8,44	0,087	0,112
Site 3 p4	11,2	<2	0,1	83,9	2,27	451	8,44	0,087	0,112
Site 3 p5	11,2	<2	0,1	83,9	2,27	451	8,44	0,087	0,112
Site 3 p6	11,2	<2	0,1	83,9	2,27	451	8,44	0,087	0,112
Site 3 p7	11,2	<2	0,1	83,9	2,27	451	8,44	0,087	0,112
Site 3 p8	11,2	<2	0,1	83,9	2,27	451	8,44	0,087	0,112
Site 3 p9	11,2	<2	0,1	83,9	2,27	451	8,44	0,087	0,112
Site 3 p10	11,2	<2	0,1	83,9	2,27	451	8,44	0,087	0,112

Site ID	**Patch descriptors**				
	Embeddedness			**Flow**	
	Largest clast	Code	Area occupied by fine material (%)	Velocity (m/s)	Depth (mm)
Site 1 p1	L. Cobble	7	25	0,32	85
Site 1 p2	S. Cobble	5	80	0,07	91
Site 1 p3	Fine	1	100	0,01	280
Site 1 p4	L. Cobble	7	10	0,21	74
Site 1 p5	S. Boulder	8	15	0,31	124
Site 1 p6	S. Boulder	8	20	0	408
Site 1 p7	L. Cobble	7	30	0,04	211
Site 1 p8	M. Cobble	6	90	0,02	145
Site 1 p9	S. Boulder	8	50	0,48	98
Site 1 p10	M. Cobble	6	60	0,01	238
Site 2 p1	Fine	1	100	0,03	281
Site 2 p2	S. Boulder	8	10	0,19	256
Site 2 p3	S. Boulder	8	2	0,34	161
Site 2 p4	S. Boulder	8	90	0,11	71
Site 2 p5	S. Boulder	8	60	0,01	231
Site 2 p6	Fine	1	100	0,01	338
Site 2 p7	L. Boulder	9	40	0,01	136
Site 2 p8	S. Boulder	8	15	0,01	406
Site 2 p9	S. Boulder	8	25	0,32	107
Site 2 p10	Fine	1	100	0,02	226
Site 3 p1	L. Cobble	7	60	0,05	92

	Largest clast	Code	Area occupied by fine material (%)	Velocity (m/s)	Depth (mm)
Site 3 p2	S. Gravel	2	99	0,18	159
Site 3 p3	S. Boulder	8	20	0,29	296
Site 3 p4	S. Boulder	8	2	0,61	84
Site 3 p5	S. Boulder	8	75	0,02	167
Site 3 p6	L. Boulder	9	40	0,06	140
Site 3 p7	S. Boulder	8	5	0,16	75
Site 3 p8	S. Boulder	8	60	0,01	194
Site 3 p9	L. Cobble	7	5	0,14	79
Site 3 p10	L. Boulder	9	60	0,2	210

APPENDIX 2

Raw data used to create HMID (July 2014)

Site 1	Velocity (m/s)	Depth (mm)	**Site 2**	Velocity (m/s)	Depth (mm)	**Site 3**	Velocity (m/s)	Depth (mm)
1	0,01	43	**1**	0,12	165	**1**	0,10	120
2	0,01	72	**2**	0,32	271	**2**	0,01	98
3	0,23	104	**3**	0,41	64	**3**	0,31	73
4	0,10	80	**4**	0,11	104	**4**	0,14	182
5	0,22	153	**5**	0,00	187	**5**	0,00	253
6	0,00	64	**6**	0,00	101	**6**	0,27	88
7	0,02	161	**7**	0,10	204	**7**	0,20	172
8	0,11	159	**8**	0,24	177	**8**	0,10	86
9	0,20	134	**9**	0,04	43	**9**	0,27	152
10	0,00	223	**10**	0,21	252	**10**	0,13	265
Std Dev.	0,096	56,091	**Std Dev.**	0,137	76,872	**Std Dev.**	0,108	68,966
Mean	0,09	119,3	**Mean**	0,155	156,8	**Mean**	0,153	148,9
CV	1,067	0,470	**CV**	0,887	0,490	**CV**	0,704	0,463
HMID	**9,24**		**HMID**	**7,91**		**HMID**	**6,22**	
Taxa	13		**Taxa**	18		**Taxa**	14	

Raw data used to create HMID (October 2014)

Site 1	Velocity (m/s)	Depth (mm)	**Site 2**	Velocity (m/s)	Depth (mm)	**Site 3**	Velocity (m/s)	Depth (mm)
1	0,31	65	**1**	0,01	55	**1**	0,02	100
2	0,01	189	**2**	0,40	150	**2**	0,03	89
3	0,11	120	**3**	0,11	187	**3**	0,42	80
4	0,25	154	**4**	0,00	76	**4**	0,33	164
5	0,02	267	**5**	0,00	190	**5**	0,01	260
6	0,01	287	**6**	0,01	265	**6**	0,11	298
7	0,30	76	**7**	0,36	75	**7**	0,25	171
8	0,29	234	**8**	0,09	304	**8**	0,57	57
9	0,00	161	**9**	0,12	65	**9**	0,12	165
10	0,01	152	**10**	0,25	43	**10**	0,09	251
Std Dev.	0,139	74,96258	**Std Dev.**	0,150794	93,035238	**Std Dev.**	0,191035	83,67563
Mean	0,131	170,5	**Mean**	0,135	141	**Mean**	0,195	163,5
CV	1,061072	0,439663	**CV**	1,116994	0,6598244	**CV**	0,979668	0,511778
HMID	**8,81**		**HMID**	**12,35**		**HMID**	**8,96**	
Taxa	16		**Taxa**	17		**Taxa**	14	

Raw data used to create HMID (March 2015)

Site 1	Velocity (m/s)	Depth (mm)	**Site 2**	Velocity (m/s)	Depth (mm)	**Site 3**	Velocity (m/s)	Depth (mm)
1	0,00	0	**1**	0,01	134	**1**	0,02	26
2	0,00	0	**2**	0,04	60	**2**	0,04	102
3	0,00	44	**3**	0,15	108	**3**	0,15	23
4	0,01	108	**4**	0,05	277	**4**	0,04	162
5	0,01	266	**5**	0,01	285	**5**	0,01	239
6	0,09	29	**6**	0,02	369	**6**	0,01	245
7	0,01	207	**7**	0,21	72	**7**	0,07	215
8	0,00	55	**8**	0,00	111	**8**	0,27	56
9	0,01	399	**9**	0,39	43	**9**	0,30	56
10	0,01	531	**10**	0,19	51	**10**	0,10	97
Std Dev.	0,027162	183,6666	**Std Dev.**	0,126934	116,08426	**Std Dev.**	0,106505	86,80815
Mean	0,014	163,9	**Mean**	0,107	151	**Mean**	0,101	122,1
CV	1,940148	1,120602	**CV**	1,186298	0,7687699	**CV**	1,054506	0,710959
HMID	**38,87**		**HMID**	**14,95**		**HMID**	**12,36**	
Taxa	13		**Taxa**	16		**Taxa**	16	

Raw data used to create HMID (May 2015)

Site 1	Velocity (m/s)	Depth (mm)	**Site 2**	Velocity (m/s)	Depth (mm)	**Site 3**	Velocity (m/s)	Depth (mm)
1	0,01	415	**1**	0,01	170	**1**	0,00	205
2	0,06	57	**2**	0,13	174	**2**	0,16	200
3	0,00	342	**3**	0,20	271	**3**	0,02	90
4	0,13	40	**4**	0,35	108	**4**	0,45	97
5	0,00	135	**5**	0,00	197	**5**	0,08	165
6	0,00	170	**6**	0,01	275	**6**	0,01	242
7	0,01	330	**7**	0,00	206	**7**	0,01	189
8	0,14	30	**8**	0,26	164	**8**	0,43	91
9	0,01	236	**9**	0,00	434	**9**	0,06	103
10	0,00	225	**10**	0,04	86	**10**	0,06	77
Std Dev.	0,055217	135,5007	**Std Dev.**	0,128582	99,485622	**Std Dev.**	0,171062	60,54833
Mean	0,036	198	**Mean**	0,1	208,5	**Mean**	0,128	145,9
CV	1,533798	0,684347	**CV**	1,28582	0,4771493	**CV**	1,336422	0,414999
HMID	**18,21**		**HMID**	**11,40**		**HMID**	**10,93**	
Taxa	16		**Taxa**	17		**Taxa**	16	

Raw data used to create HMID (August 2015)

Site 1	Velocity (m/s)	Depth (mm)	**Site 2**	Velocity (m/s)	Depth (mm)	**Site 3**	Velocity (m/s)	Depth (mm)
1	0,32	85	**1**	0,03	281	**1**	0,05	92
2	0,07	91	**2**	0,19	256	**2**	0,18	159
3	0,01	280	**3**	0,34	161	**3**	0,29	296
4	0,21	74	**4**	0,11	71	**4**	0,61	84
5	0,31	124	**5**	0,01	231	**5**	0,02	167
6	0,00	408	**6**	0,01	338	**6**	0,06	140
7	0,04	211	**7**	0,01	136	**7**	0,16	75
8	0,02	145	**8**	0,01	406	**8**	0,01	194
9	0,48	98	**9**	0,32	107	**9**	0,14	79
10	0,01	238	**10**	0,02	226	**10**	0,20	210
Std Dev.	0,171273	108,1996	**Std Dev.**	0,132351	104,97836	**Std Dev.**	0,177814	71,26352
Mean	0,147	175,4	**Mean**	0,105	221,3	**Mean**	0,172	149,6
CV	1,165123	0,616873	**CV**	1,260481	0,4743712	**CV**	1,033802	0,47636
HMID	**12,26**		**HMID**	**11,11**		**HMID**	**9,02**	