Acoustic monitoring and response of katydids (Orthoptera: Tettigoniidae) to the landscape mosaic in a Biosphere Reserve

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Declaration

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Abstract

A charismatic group within the Orthoptera, katydids can be found in a variety of habitat types world-wide due to their excellent bark and leaf mimicry skills. Most male katydids produce species-specific calls to attract female mates. If katydids, like their close relatives the grasshoppers, can function as effective biological indicators, then acoustic monitoring of katydid songs may result in a novel and non-invasive method to rapidly assess local biodiversity. Furthermore, information regarding threat statuses, distributions and life history traits can be inferred for all South African katydid species, leading to the development of a Katydid Biotic Index (KBI) based on the highly effective Dragonfly Biotic Index. If proven effective, the KBI would allow for biodiversity assessments to account for detailed aspects of katydid species composition in addition to the diversity measures normally utilized for biodiversity assessment (e.g. species richness and abundance). In this thesis, I provide the first steps towards determining whether the KBI could be an effective assessment technique. First I assess the utility of the KBI at a coarse-scale by determining its ability to identify regions of high conservation priority. Secondly, I conduct a fine scale study to determine the response of the katydid assemblage to habitat quality. And lastly, the first two aims are combined to determine whether the KBI is an appropriate method to assess habitat quality at a fine-scale.

In Chapter 2, by using a subset of museum records, I investigate the distribution of the katydids within the Cape Floristic Region (CFR), a global biodiversity hotspot. The katydids found within the CFR follow the same trends with regards to threat status, endemism and life history traits to the overall South African katydid assemblage. The KBI assessment method was able to select, at this coarse-scale, the ecosystems of conservation priority.

For Chapters 3 and 4, the Kogelberg Biosphere Reserve (KBR) was selected as a study area as it allowed for the acoustic monitoring and direct comparison of katydid assemblages and responses across the core, buffer and transition zones through the use of passive recordings. In Chapter 3 I found that the katydids of the KBR are not complementary across the zones. However, they respond positively in terms of abundance to measured habitat quality when the entire assemblage is considered. In Chapter 4 I found that katydids responded towards coarse-scale habitat quality and they were not as sensitive towards habitat change as was expected. By including abundances of the katydid species in to the KBI calculations, the sensitivity of the KBI as an assessment method was improved.

For this reason, katydids in the fynbos biome are likely to not be effective indicators of habitat change on a small scale, likely due to the surprisingly low diversity of katydid species in the KBR. However, if the KBI were to be tested out in forest patches or areas with higher diversity, the KBI may prove more promising. For these reasons, a rapid assessment technique based on the KBI is likely to be more appropriate for some habitat types over others.

Opsomming

‘n Charismatiese groep binne die Orthoptera, sabel sprinkane, kan gevind word in 'n verskeidenheid van habitat tipes wêreldwyd as gevolg van hul uitstekende bas en blaar nabootsing vermoë. Die meeste manlike sabel sprinkane produseer spesie-spesifieke geluide om wyfies te lok. Indien sabel sprinkane, soos hul naasbestaandes die sprinkane, effektief as biologiese aanwysers funksioneer, kan akoestiese monitering van sabel sprinkaan geluide lei tot 'n unieke en nie-indringende metode om plaaslike biodiversiteit vinnig te evalueer. Verder, kan inligting rakende bedreiging statusse, verspreiding en lewensgeskiedenis eienskappe afgelei word vir alle Suid-Afrikaanse sabel sprinkaan spesies, wat kan lei tot die ontwikkeling van 'n Sabel Sprinkaan Biotiese Indeks (SBI) gebaseer op die hoogs doeltreffende Naaldekoker Biotiese Indeks (NBI). Indien dit as doeltreffend bewys word, sou die SBI voorsiening maak vir ‘n biodiversiteit assesseringsmetode om rekenskap te gee aan gedetailleerde aspekte van sabel sprinkaan spesiesamestelling bykomend tot die diversiteit maatreëls wat normaalweg gebruik word vir biodiversiteit assessering (bv. spesierykheid en volopheid). In hierdie tesis, wend ek die eerste poging aan om te bepaal of die SBI 'n effektiewe assessering tegniek kan wees. Ek het aanvanklik die gebruiklikheid van die SBI op 'n growwe skaal beoordeel deur die bepaling van die indeks se vermoë om areas van hoë prioriteit vir bewaring te identifiseer. In die tweede plek, doen ek 'n studie op ‘n fyn skaal om die reaksie van sabel sprinkaan spesiesamestelling tot habitat kwaliteit te bepaal. Laastens, is die eerste twee doelwitte gekombineer om te bepaal of die SBI 'n geskikte metode is om habitat kwaliteit te evalueer op 'n fyn skaal.

In Hoofstuk 2, met die gebruik van ‘n gedeelte van museum rekords, ondersoek ek die verspreiding van sabel sprinkane binne die Kaapse Floristiese Streek (KFS), 'n globale biodiversiteit brandpunt. Die sabel sprinkane in die KFS volg dieselfde tendense met betrekking tot bedreiging status, endemisme en lewensgeskiedenis eienskappe in vergelyking met die algehele Suid-Afrikaanse sabel sprinkaan versameling. Die SBI assesseringsmetode was in staat, op hierdie growwe skaal, om die ekosisteme van prioriteit vir bewaring te selekteer.

Vir Hoofstukke 3 en 4, is die Kogelberg Biosfeerreservaat (KBR) as studiegebied gekies omdat dit akoestiese monitering en direkte vergelyking van sabel sprinkaan spesiesamestelling en reaksies oor die kern, buffer en oorgang sones met gebruik van passiewe opnames toegelaat het. In Hoofstuk 3 het ek gevind dat die sabel sprinkane van die KBR nie aanvullende is oor die sones nie, maar hulle reageer positief in terme van volopheid gemeet teenoor habitat kwaliteit wanneer die hele spesiesamestelling in ag geneem word. In Hoofstuk 4 het ek bevind dat sabel sprinkane gereageer het teenoor growwe skaal habitat kwaliteit en hulle was nie so sensitief teenoor habitat verandering as wat verwag is nie. Deur die insluiting van volopheid van die sabel sprinkaan spesies in die SBI berekeninge is die sensitiwiteit van die SBI as 'n assesseringsmetode verbeter.

Vir hierdie rede, is sabel springkane in die fynbos bioom geneig om nie doeltreffende aanduidings van habitat verandering op 'n klein skaal, waarskynlik as gevolg van die merkwaardige lae diversiteit van sabel sprinkaan spesies in die KBR. Maar, indien die SBI getoets sou wees in bos fragmente of gebiede met hoër diversiteit van sabel springkane, kan die SBI as meer belowend bewys word. As gevolg van laasgenoemde redes, is 'n vinnige assessering tegniek gebaseer op die SBI geneig om meer gepas vir sommige tipes habitat teenoor ander te wees.

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Chapter 1

General Introduction

Katydid Biology

The orthopteran story starts 250 million years ago when the orthopterans started to evolve with the very first insects (Song et al. 2015). Today, orthopteran species can be found in a wide variety of habitats, from alpine meadows (Guido & Gianelle 2001) to swarms of desert locust, *Schistocerca gregaria* emerging after sufficient rainfall in the Northern Sahel (Sánchez-Zapata et al. 2007). Variation of physical characteristics is common both between (Gangwere et al. 1997; Song et al. 2015) and within species (Chapman & Joern 1990; Tanaka 2006), resulting in different species being able to adapt to the wide variety of habitats in which they are found (Chapman & Joern 1990; Reinhardt et al. 2005; Vandergast et al. 2007). Often, for this reason, orthopteran species are considered worldwide pests as several species can successfully live on agricultural crops and in the process destroy the yield for an entire year (Abate et al. 2000; Lomer et al. 2001).

Within the Orthoptera are the Tettigoniidae or katydids (Gangwere et al. 1997). Katydids are the most diverse and speciose orthopteran family (Song et al. 2015). Widely distributed, katydids can be found in the tropical forests of India (Nityananda & Balakrishnan 2006; Balakrishnan et al. 2014) down to several isolated caves in the Cederberg Mountains of South Africa (Bazelet & Naskrecki 2014). As observed within the greater orthopteran grouping, katydids display a diverse range of physical characteristics that enable them to inhabit a wide variety of habitats (Brown 1983; Bailey & Rentz 1990; Gangwere et al. 1997; Gwynne 2001). Katydids are highly charismatic species and experts in camouflage, many species are excellent leaf and bark mimics (Bailey & Rentz 1990; Gwynne 2001),and can be active both during the day but predominantly at night (Gwynne 2001). These factors, combined with the fact that katydids are notoriously difficult to sample (Blanton 1990; Schirmel et al. 2010) have previously deemed them as an unfavourable taxon for ecological studies.

However, male katydids produce species-specific calls (Nityananda & Balakrishnan 2006), either within the audible or ultrasonic ranges (Bailey & Rentz 1990), to advertise their location as well as communicate their breeding potential to females (Brown 1983). The sound production organs are situated on the wings. The left forewing bares the file, a hardened tooth-baring vein, while the hind right wing edge forms the scraper. By rapidly rubbing the wings together, the scraper rubs up and down the file thereby producing a recognisable call. The size and patterns of the teeth on the file as well as the frequency with which the file is rubbed, vary across species and result in the production of the species-specific calls (Brown 1983). Even apparently flightless males such as those belonging to the *Ceresia* genus and Hetrodinae subfamily have reduced wings, situated beneath the pronotum, which are equipped with a file and scraper mechanism and are able to produce loud audible calls (Rentz 1988; Kowalski & Lakes-Harlan 2013). Some species have specialised enlarged areas on their wings, known as the mirror, which further amplifies the songs produced. In some species, where males do not produced sounds, males still communicate with females by a process called drumming. Drumming involves stamping their hind leg on a suitable substrate, either the ground or a leaf (Brown 1983). Though thought to be silent, there is evidence of females of *Onomarchus uninotatus* tremulating in phase to the males call as a response (Rajaraman et al. 2015).

Although it is known that songs aid in mate choice, very little work has been done on the courtship of katydids since the mid 1990's. It is known that males produce a nuptial gift for the females during mating. This gift, a spermatophore, contains both the sperm and a nutritional food source for the female (Bailey & Rentz 1990). This gift is thought to encourage the females to actively seek out the singing males as the food reward outweighs the predation risk (McCartney et al. 2012). Supporting this hypothesis is the finding that female katydids are eaten significantly more by bats than their male counterparts. Even though the females are acoustically silent, it is thought that the noise produced while flying and travelling to the males is enough to increase their risk of predation. The males, although producing obvious sounds, remain stationary and are able to quickly seek refuge should a bat approach, reducing their risk of predation (Raghuram et al. 2015).

Following copulation and the presentation of the spermatophore, female katydids are required to find a suitable location in which to lay their eggs (Brown 1983). Unlike acridids, female katydids have external ovipositors comprised of three valves, the dorsal, anterior and inner. Tongue and groove joints enable the valves to palpate and pass eggs from the body to the tip of the ovipositor (Brown 1983). There is great variation in the appearance of ovipostors, from long and straight to short and sharply curved, with or without serrated teeth on the tip, they can be a useful diagnostic character for the identification of the various species (Naskrecki et al. 2008; Naskrecki & Bazelet 2009, 2011). The ovipositors are used to lay eggs deep within suitable substrates, either soil or plant tissue (Brown 1983; Bailey & Rentz 1990). Sensilla cover the ovipositors and aid the female to determine whether the substrate is suitable as well as to help her monitor the position of the egg within the ovipositor (Brown 1983).

The development of katydid eggs is temperature-dependent, as is the case with many insects. The developmental temperature ranges of the eggs tend to differ between the different species. Many species also employ various stages of diapause during egg development (Bailey & Rentz 1990). This then results in various emergence periods for the different species. Much is known about the general biology and ecological requirements of the different species of katydids, although this information is mainly obtained through laboratory-based observations (Brown 1983). Very little information has been collected in field with regards to their mating, egg laying and egg development, contributing to their reputation of being notoriously difficult study organisms.

Katydids of South Africa

Currently there are an estimated 169 species of katydids in South Africa. Of these, 129 species have relatively stable taxonomy. Although detailed, species-specific information is not available for every species, much can be inferred about their distributions as well as their general life history traits from knowledge of closely-related species or museum records (Naskrecki 2008). This knowledge is evident in the fact that all 129 described species were assessed in accordance to the IUCN Red List criteria and in 2014 had the appropriate threat statuses assigned. Traditionally, the warm and humid subtropical regions of South Africa are thought to be the most suitable habitats for katydids as the presence of forests and trees (Rebelo et al. 2006) in the natural landscapes provide more suitable habitats for katydids due to their excellent mimicry of leaves and bark (Bailey & Rentz 1990; Gwynne 2001).

The Cape Floristic Region (CFR) is known for its very high floral diversity, high endemism and high degree of threat faced due to anthropogenic land use change (Myers et al. 2000; Mittermeier et al. 2004). It is for this reason that the CFR is one of the three biodiversity hotspots within the megadiverse South Africa (Thuiller et al. 2006), and one of only 35 global biodiversity hotspots (Mittermeier et al. 2004). Based on museum records contained within the MANTIS database (Naskrecki 2008), only 38 of the Of the 129 valid South African species are thought to occur within the CFR. In comparison with the rest of South Africa, the CFR seems to have relatively low katydid diversity, the species present have high levels of endemism as an estimated 49% of the species are thought to be endemic to the area (see Chapter 1 for analysis). This suggests that parts of the CFR can be thought of as a katydid hotspot (Bazelet et al. 2016) as there are high levels of endemism, as well as the species facing high levels of threat as the threat of land-use change threatens insects and plants alike (Hahn et al. 2005; Bomhard et al. 2005).

The interest in katydids does not only stem from the fact that they communicate acoustically, but it is also hypothesised that they may be potential indicators of habitat quality. Acrididae in grasslands to the north of South Africa are effective indicators of grassland health (Bazelet & Samways 2011a, 2011b). Therefore due to the close relationship between katydids and grasshoppers it stands to reason that katydids may function as indicator species. For this reason, the benefits of studying katydids in the fynbos outweigh the logistical challenges encountered while working on them.

Potential threats to the katydids of the Cape Floristic Region

In short, the katydids of the CFR are significantly understudied. Much is known and information available with regards to the plants of the CFR (Cowling et al. 2003) but very little information is available for the katydids. There are a few taxonomic descriptions (Naskrecki et al. 2008; Naskrecki & Bazelet 2009, 2011) and only one study focusing on how the katydid assemblage changes along elevation gradient (Grant 2014). There is a significant lack of knowledge regarding the fine scale ecological requirements of the species and how their fine scale distributions will be effected in response to habitat transformation, fragmentation and agricultural activities.

Natural habitats within the CFR are under considerable threat due to fragmentation and landscape transformation in the form of urbanisation and agricultural activities (Bomhard et al. 2005; Forest et al. 2007). These combined factors have resulted in an estimated 37% loss in area of natural vegetation (Richardson et al. 1996). These are not the only threats faced by the CFR nor are they endemic to the CFR (Myers et al. 2000). Urbanisation and agricultural activities are the leading causes of global biodiversity loss (Pimm & Raven 2000; Butchart 2011; Hooper et al. 2012). In an attempt to bridge the divide between conservation and the impact of humans on the environment, the Man and the Biosphere Programme (MAB) was established by UNESCO (UNESCO 2016).

In a novel approach to reserve management, the MAB delineates biosphere reserves (BR) into three zones depending on the extent of socio-economic activities that take place in the area. The three zones are 1) the core zone in which no socio-economic activities occur apart from conservation and research, 2) the buffer zone in which light but sustainable activities may occur such as ecotourism and 3) the transition zone which allows for the greatest degree of human activity and incorporates both agricultural land and urban centres. To date, 669 BRs have been established in 120 countries globally (UNESCO 2016).

One of the major goals of the MAB programme is to facilitate the generation of ecological knowledge that can feed directly into the decision-making surrounding the management of the BRs. This feedback loop is aimed at benefiting both the environment and the people living in and around the BRs (UNESCO 2016). Currently, very little empirical research is actually done in this regard and results in a knowledge gap between the environment and management decisions. South Africa is home to eight BRs (UNESCO 2016), yet the relative value and contribution of the three zones is unclear. It is not known whether or not the three zones integrate with each other across this varied landscape, for example do management practices employed in the transition zones have direct effects on the species and habitats found within the core and buffer zones and vice versa. BRs offer an invaluable and unique opportunity to study the response of biodiversity to changing habitat quality relating to increasing levels of human activity.

The Kogelberg Biosphere Reserve (KBR), designated in 1998, is the oldest of eight biosphere reserves in South Africa (UNESCO 2016). Even after almost 20 years, the worth of the KBR is unknown even though it is situated within one of the most biodiverse regions of the world (Boucher 1978; Grant & Samways 2007; Müller 2008). The flora of the area is understood and for this reason the KBR is considered to be a hotspot within a hotspot of biodiversity (the CFR) (Grant & Samways 2011). The beta-diversity of the region is unrivalled anywhere else in the CFR (Boucher 1978). Yet research has focused on the plants in the region, and as a result, there is paucity of information with regards to the invertebrates. So far, the only published study of invertebrates originating from the CFR is on dragonflies. From this work it is apparent that the dragonfly assemblages within the buffer and transition zones were complementary to that of the core, suggesting that the core zones and species therein are buffered against anthropogenic changes in the transition zones by the presence of the buffer zone (Grant &

Samways 2011). The KBR is therefore an ideal study site in which to evaluate the value of BRs, for conservation of a taxon rich in endemics, such as the katydids.

Global approaches to acoustic monitoring

Acoustic monitoring of terrestrial environments focuses on taxa that produce recognisable and identifiable calls, namely birds, bats, frogs and insects such as Orthoptera (Fischer et al. 1997; Sueur et al. 2008). With many sampling techniques being available (Ganchev et al. 2007; Gasc et al. 2013), acoustic surveys are able to be carried out in a rapid and cost effective manner. Much research has been done in linking acoustic monitoring of ecosystems to conservation in order to develop habitat assessment methods. In Australia, for example, the acoustic monitoring of bird species in eucalypt forests has enabled researchers to effectively determine ecological condition of the forests (Tucker et al. 2014). Furthermore acoustic monitoring on a nocturnal bittern has enabled researchers to accurately monitor the response, recovery and recolonisation of a restored wetland by these bitterns over the course of 5 years (Frommolt & Tauchert 2014). In the Amazonian rainforests, acoustic monitoring is helping to determine reliable estimates of species diversity in regions where the actual number of living species far outweigh the number of described species. This then enables managers of these richly biodiverse regions to accurately map the distribution of various acoustic species (Riede 1993; Laiolo 2010). Orthopteran species are known to be sensitive towards habitat condition and therefore are indicative of the quality of various habitat types and so acoustic monitoring of various orthopteran species facilitates the monitoring of various tropical forest systems for conservation purposes (Riede 1993; Depraetere et al. 2012).

Acoustic monitoring of the katydids in the CFR

South Africa has set a precedent for rapid biodiversity assessment methods, namely the South African Scoring System (SASS) and the Dragonfly Biotic Index (DBI). SASS utilises the larvae of benthic macro-invertebrates to determine the quality of flowing freshwater (Dickens &

Graham 2002), while the DBI employs the adult dragonfly assemblage to determine the quality of both flowing and still freshwater bodies (Samways & Simaika 2016). The inclusion of species-specific information into the assessments makes the DBI a powerful tool which is sensitive to changes in habitat quality. Each individual dragonfly species has been assessed in accordance to their threat statuses, distributions as well as sensitivity to habitat quality (Samways & Simaika 2016).

Acoustic monitoring of the katydids of the KBR would allow for the direct comparison of the katydid assemblages present in the three zones without interfering with the individuals behaviour. Although not a new technique (Busnel 1963), advances in technology have enabled the field of acoustic monitoring to grow dramatically over the last few years, with advancements enabling a wide range of sampling techniques. For example, techniques range from programmable passive recorders to hand-held microphones connected to a recorder as well as heterodyne detectors to identify ultrasonic species. Perhaps the simplest method is simply listening to the soundscape. Humans are able to recognise different species calls as well as the direction and number of calls (Diwakar et al. 2007). As acoustic monitoring eliminates the need to catch individuals, biodiversity assessments can therefore become streamlined and less time-consuming. As well as streamlining assessments, the acoustic sampling does not interfere with the animal's behaviour, resulting in more natural observations.

The dense and sclerophyllous nature of the fynbos renders traditional invertebrate sampling methods impractical and unsuccessful (Pryke & Samways 2008). It is for this reason that acoustic sampling of invertebrates within the fynbos appears more suitable. A recorder can be placed alongside a dense stand of vegetation and still record the species within the stand. This eliminates the need to manoeuvre through dense vegetation yet the patch is still sampled.

As the threat statuses, distributions and life histories of the South African katydids are known, it is therefore possible to assess the katydid species in a manner very similar to that of the DBI. This Katydid Biotic Index (KBI) could then be used in conjunction with acoustic monitoring to determine terrestrial habitat quality. This could therefore lead to the establishment of a new rapid biodiversity assessment method.

Thesis outline

In this thesis, in which Chapters 2 to 4 have been written as stand-alone scientific publications, the following stepwise approach has been taken to establish a katydid-based rapid biodiversity assessment technique. In Chapter 2, a metadata analysis of museum records was conducted to illustrate how the KBI may be useful as coarse spatial-scale across the CFR. In Chapter 3, extensive field-collected data of KBR katydids are analysed to determine how the katydid assemblages differ across the BR zones and whether there is a correlation between the katydid assemblages and habitat quality. In Chapter 4, the results of Chapters 2 and 3 are combined and the KBI technique is applied to the field-collected data in order to determine how the KBI technique performs on a smaller spatial scale, as smaller scales are more realistic for habitat management. Finally, Chapter 5 summarises the finding of all three data chapters and conclusions are drawn regarding the suitability of katydids and the KBI as a rapid assessment technique in a South African context. Recommendations and the next steps for the adoption of this method are also made.

Aims and Objectives

The overall objective of this study was to determine whether katydids can be acoustically monitored across the CFR as well as to determine their potential to become biological indicators. The main aims of the thesis are:

1. To adapt the DBI scoring system to assess katydids.
2. To conduct a case study to illustrate the efficiency of the KBI method in assessing habitat quality of the CFR in order to highlight areas in need of conservation.
3. To determine whether the zones of the KBR are complementary in terms of katydid species composition.

4. To determine if any particular species stand out as accurate indicators of habitat quality.
5. To propose a standardized method for the semi-quantitative assessment of fynbos habitat quality
6. To determine whether the inclusion of species abundances into the KBI calculations helps to improve the sensitivity of the scoring system by examining the correlation between the habitat quality and zones of the KBR to the KBI scores.


Core Zone


Buffer Zone


Transition Zone

Chapter 2

Developing a katydid (Tettigoniidae) rapid assessment technique of ecosystem vulnerability: case study of a biodiversity hotspot, the Cape Floristic Region, South Africa

Abstract

Global biodiversity faces many challenges, with the conservation of invertebrates among these. South Africa is megadiverse and home to three global biodiversity hotspots. It also employs two invertebrate-based rapid assessment techniques to evaluate habitat quality of freshwater ecosystems. While grasshoppers (Acrididae) are known indicators of terrestrial habitats, katydids (Tettigoniidae) could be as well. Here, we adapt a South African freshwater invertebrate-based rapid assessment method, the Dragonfly Biotic Index (DBI), for the terrestrial katydid assemblage, and propose a new assessment approach using katydids: the Katydid Biotic Index (KBI). KBI assigns each katydid species a score based on its IUCN Red List threat status, geographical distribution, mobility, and trophic level. This means that the rarer, more localized, specialized and threatened katydid species receive the highest score, and the common, geographically widespread and Least Concern species the lowest. As a case study, we calculated KBI across one of South Africa's global biodiversity hotspots, the Cape Floristic Region (CFR). We then correlated KBI/Site scores of individual ecosystems with their ecosystem threat scores. The CFR's katydid assemblage did not differ significantly from that of the overall South African katydid assemblage in terms of its species traits, threat statuses, or distribution among tettigoniid subfamilies. Likewise, KBI/Site scores did not differ significantly among ecosystem threat statuses. This may be explained by the coarse spatial scale of this study or by the lack of specialization of the CFR katydid assemblage. Nevertheless, the KBI holds promise as it is a relatively simple and non-invasive technique for taking invertebrate species composition into account in an assessment of habitat quality. In

regions where katydid assemblages are well-known, acoustic surveys and KBI may provide an efficient means for assessing habitats.

Introduction

Global biodiversity is facing many challenges, resulting in the extinction of species at rates estimated to be 100 to 1000 times faster than the background extinction rate (Rockström et al. 2009). Biodiversity is often measured, or assessed, to guide conservation planning. These assessments involve the measurement of various vertebrate or plant taxa. Although invertebrates are often not included in these assessments owing to their high numbers of species, it is sometimes assumed that due to the great numbers of insect-plant interactions that insect diversity may mirror that of the plants (Myers et al. 2000). Also, biodiversity assessments usually overlook species-specific information, so ignoring the intrinsic value of each species, and compromising the economic viability and conservation value of biodiversity assessments (Samways 2002).

South Africa currently employs two robust and rapid biodiversity assessment methods targeting fresh water and riparian habitats: the South African Scoring System (SASS) (Dickens & Graham 2002) and the Dragonfly Biotic Index (DBI) (Samways & Simaika 2016). Both of these methods are simple yet effective ways in which stream condition can be assessed based on the resident aquatic larvae of invertebrates (SASS) or on the adult dragonfly assemblages (DBI). The DBI uses three sub-indices to indicate the quality of a freshwater system: geographical distribution, habitat sensitivity and Red List status of each species at a focal locality. Based on these three sub-indices, each species is individually assessed and assigned a score of 0 to 9. The higher a species score, the higher the sensitivity of the species and the lower its tolerance to habitat disturbance. This results in dragonfly assemblages being directly comparable in terms of their conservation value and allows for the ranking of different habitats according to their level of disturbance (Samways & Simaika 2016).

Grasshoppers (Orthoptera: Acrididae) in South Africa are good bioindicator group within the grassland ecosystems (Bazelet & Samways 2011a, 2011b) and the Cape Floristic Region (CFR) (Matenaar et al. 2015). However, katydids (Orthoptera: Tettigoniidae) have not yet been explored in the region, and could potentially also be good biological indicators, especially in more woody environments. There are an estimated 169 katydid species in South Africa and, of these, two thirds are thought to be endemic to the country (Picker et al. 2004). So far, 129 species have been described and, as of December 2014, the threat statuses of these species have been assessed and uploaded onto the IUCN Red List (Bazelet et al. 2016; IUCN 2016). Coupled with the threat statuses, a wealth of coarse-scale additional information is available, such as estimates of species distributions and life history information. In-depth studies on the biology of individual species are almost entirely lacking, but confident predictions can be made on the basis of trends among species and within higher taxa. Most notably, mature male katydids produce characteristic species-specific songs enabling non-invasive species detection in an environment by listening alone (Bailey & Rentz 1990). Combined, these characteristics make katydids an attractive taxon upon which an acoustic rapid assessment method could be based for assessing the quality of terrestrial habitats in South Africa (Grant & Samways 2016).

Rapid assessment techniques are vital tools for detecting biodiversity, particularly in areas which have high species diversity and/or experience extreme threat, such as the biodiversity hotspots (Myers et al. 2000; Alonso et al. 2011). Global biodiversity is not homogenous in its distribution (Gaston 2000), with biodiversity hotspots covering only 2% of Earth's surface. Yet 50% of all plant species and 42% of terrestrial vertebrate species exist in this 2% of land (Mittermeier et al. 2004). These "traditional" biodiversity hotspots do not take into account invertebrate diversity, as it was assumed that insect diversity mirrors that of the plants based on the high numbers of observed insect-plant interactions (Orme et al. 2005). The CFR, one of three biodiversity hotspots in mega-diverse South Africa (Mittermeier et al. 2004), is an example of insect diversity mirroring plant diversity (Procheş & Cowling 2006), although these patterns do vary among insect taxa, with some having significantly higher diversity than others (Wright & Samways 1998; Procheş & Cowling 2006).

Here, a new biodiversity assessment method that employs katydids for monitoring terrestrial habitat quality based on an adaptation of the DBI is outlined. The calculation of the Katydid Biotic Index (KBI) is described, and a subset of museum records is used to conduct a case study to illustrate the efficacy of the KBI for assessing biodiversity and habitat quality across a biodiversity hotspot, the CFR, in South Africa. Ultimately, the KBI is evaluated with regards to its possible use in highlighting ecosystems in need of conservation action.

Materials and methods

Data collection

In 2014, the Red List threat status of 133 katydid species were assessed in 2014 by Dr Bazelet, using records obtained from Piotr Naskrecki's MANTIS database. Distribution ranges of species and species endemism were calculated using the collection localities of the records. Published taxonomic descriptions as well as expert knowledge were used to assess various life history traits of the individual species (Rentz 1988; Naskrecki et al. 2008; Naskrecki & Bazelet 2009, 2012) see Bazelet et al. 2016 for methods description.

Development of the Katydid Biotic Index

The KBI allows for individual species to be ranked and compared. Based on similar criteria to that of the DBI, katydids were assessed based on three sub-indices: 1) geographical distribution, 2) life history traits (which consist of mobility and trophic level), and 3) Red List status. Each sub-index is scored out of three, with the life history category being a combination of individual scores for mobility and trophic level. These sub-indices (each of which ranges from 0-3) are added together to give the KBI score for a species. These species KBI scores range from 0 for a widespread, habitat tolerant, Least Concern (LC) species to 9 for a narrow-range, highly habitat sensitive and Red Listed species (Table 2.1; Bazelet *et al*. 2016).

The sum of the scores in any specified region or at any particular site is the total KBI score. When the site score is divided by the number of species recorded, it gives the KBI/Site score. The KBI/Site score is thus an average value calculated from all the individual KBI species scores, and allows for the ranking of sites based on their katydid assemblages.

Katydids in the Cape Floristic Region

Globally renowned for its botanical diversity, the CFR includes 122 different vegetation types or ecosystems (Government Gazette 2011) and covers < 4% of southern Africa or an area of ±90 000 km^2. Within this relatively small area, an estimated 8640 species of plants occur, of which 65% are considered endemic to the CFR. The total number of species within the CFR is disproportionate to its small size as the observed number of species is comparable to that of tropical regions (Goldblatt & Manning 2002).

A subset of geo-referenced katydid collection localities (n = 207 and accurate to 8 decimal places) for the CFR was extracted from the MANTIS (Naskrecki 2008) database. Using QGIS (Quantum GIS Development Team 2015a) katydid records were associated with the CFR ecosystem in which they were found, the threat statuses of the individual ecosystems was available in the list of threatened terrestrial ecosystems (available through the Biodiversity GIS programme of the South African National Biodiversity Institute, map scale was 1:250 000). Duplicate records of the same species were removed from the ecosystems so that there was only one record per species per ecosystem. Average KBI values for each individual ecosystem were calculated. The threat scores and average KBI scores were then mapped using QGIS.

Statistical analysis

A Chi-square contingency table was used to determine whether the distribution of species among threat statuses and level of endemism were significantly correlated for South African and CFR katydid species. A Kruskal-Wallis test in R (R Development Core Team 2015a) was used to assess differences in mean KBI scores of the katydid assemblages of the individual ecosystems and the threat categories to which the ecosystems belong (LC, VU, EN and CR). Kruskal-Wallis was selected as it is suitable for non-parametric data, as KBI scores were not normally-distributed (Shapiro-Wilk's W = 0.95, $p < 0.001$). Post-hoc Nemenyi-Tests were then conducted using the package PMCMR in R (Pohlert 2014a) to assess pairwise differences among katydid threat statuses, ecosystem threat status and average KBI. After mapping the threat scores and average KBI scores of the ecosystems these two maps were then visually assessed in order to identify any emergent patterns.

Results

Of the 133 katydid species which were assessed for IUCN Red List threat status, 16 (12%) were Data Deficient (DD) and were therefore excluded here from further analyses. Across all South African katydid species, over 50% are considered to be LC, while 31% of species were assessed as threatened (Vulnerable (VU), Endangered (EN), or Critically Endangered (CR); Fig. 2.1A). Within the CFR, of the non-DD species, almost three-quarters (73%) of species are LC, and 27% of species are threatened (Fig. 2.1B).

The CFR katydids did not differ significantly from all South African katydids in terms of the number of species assigned to each threat status, endemism level, mobility class or trophic level ($\chi^2_{(df=3,\ =134)} = 0.88$, $p > 0.05$; $\chi^2_{(df=3,\ =38)} = 0.25$, $p > 0.05$; $\chi^2_{(df=2)} = 0.9$, $p > 0.05$ and $\chi^2_{(df=3)} = 0.07$, $p > 0.05$ respectively; Fig. 2.1).

Within the total katydid assemblage of South Africa, all species considered to be threatened (VU, EN or CR) were also endemic to the country, this is also true for the CFR katydid species (Fig. 2.2A, B). In total, 34.4% (n = 11) of all species within the CFR are flightless, and within the entire South African assemblage 28.9% (n = 11) are flightless (Fig. 2.1G, H). No South African flighted species were assessed as either EN or CR (Fig. 2.2C), and all flighted species in the CFR were assessed to be LC (Fig. 2.2D). Among the South African katydid assemblage, species with varying trophic levels were evenly spread across the threat status categories

(Fig. 2.2E). However, within the CFR katydid assemblage, all omnivorous species were classified as LC, while 25% of species (n = 8) are monophagous herbivores and these were relatively more prevalent in the threat classes (VU, EN and CR) than in LC (Fig. 2.2F).

The distribution of species in each subfamily maintains similar patterns in the CFR as in South Africa as a whole, with Phaneropterinae the most abundant subfamily overall, and Pseudophyllinae the least common (Fig. 2.3).

LC katydids have significantly lower median species-specific KBI scores than the threatened katydids (VU, EN and CR), which do not differ from each other (χ^2= 44.18, df = 9, $p < 0.05$). There were no significant differences in the mean KBI scores among the ecosystem threat status categories (χ^2= 3.28, df = 3, $p > 0.05$; Fig. 2.4). Through visual inspection, the western section of the CFR, there appears to be a slight but non-significant inverse correlation between the KBI score with ecosystem threat status, such that the lower the KBI score, the more threatened the ecosystem threat status. In the eastern section of the CFR, this relationship is not evident (Fig. 2.5).

Discussion

Although no significant differences were observed among the ecosystem threat statuses in terms of their KBI/Site values (i.e. average KBI value), the aim was rather to show how the KBI could be employed in the future once more thorough sampling has been conducted. When mapped, patterns do start to emerge in KBI/Site values among ecosystems. Ecosystems with low KBI/Site scores (mean KBI 0 – 4) tend to be those which are threatened (CR, EN and VU ecosystems) in the western CFR while the LC ecosystems tend to score higher KBI/Site values (mean KBI 5 – 8). This relationship is to be expected, as the more common and less sensitive species will be able to persist in ecosystems that have been anthropogenically altered from the original state. Whereas the more sensitive and threatened species (those with higher species-specific KBI values) are expected to prefer the natural habitats and not to persist in the transformed systems. However, in the eastern CFR, where the ecosystems appear to be less threatened overall, there seems to be little correlation between the threat status of the ecosystems and their KBI/Site values. The LC ecosystems score relatively low KBI/Site values between 0 and 4. These discrepancies could be due to numerous factors.

Among the possible explanations for the lack of correlation between ecosystem threat status and KBI/Site value, the small sample size is the most likely. With only 162 unique katydid records being present in 54 of the 122 CFR ecosystems (or 44% of ecosystems), the area is under-sampled. Furthermore, the scale of this study was very coarse and the KBI/Site values were calculated according to ecosystem threat polygons which is not a relevant biological spatial scale for katydids. Future work would need to determine the spatial scale at which the KBI/Site would be an accurate measure, as has been discussed for the DBI (Samways & Simaika 2016).

Furthermore, the CFR is an arid biome characterized by a matrix of agriculturally transformed landscapes and the native fynbos vegetation, which is characterized by evergreen plants in the Ericaceae, Restionaceae and Proteaceae. Large trees are naturally almost absent from the CFR (Rebelo et al. 2006). In turn, katydids are known to be most diverse and abundant in tropical forest habitats and some subfamilies, like the Pseudophyllinae, show a strong degree of adaptation to tree environments, often bearing a strong cryptic resemblance to their tree habitats. Understandably, Pseudophyllinae are extremely rare in the CFR and in South Africa in general, of which only 1% is native forest habitat (Mucina & Rutherford 2006).

South African katydids are relatively well-documented (Naskrecki, unpublished data). Information regarding the ecology and habitat requirements of the species is relatively well-known, and where information is lacking it is possible to infer biologies based on well-documented related species. Indeed, most species could be assessed for the IUCN’s Red List (Bazelet et al. 2016; IUCN 2016). Although some habitats and katydid groups are more diverse than others, katydids are found in nearly all terrestrial ecosystems in South Africa and thus present themselves as a favourable taxon upon which to base a rapid assessment method. All threatened South African katydids (VU, EN and CR) are either national endemics or are localised endemic species. Similar patterns are seen in the effective mobility of a species, with the less mobile species featuring more prominently in the threat classes. These patterns are also then maintained within the CFR katydids. Katydid traits are shown here to correlate with threat status, thus providing further evidence that the KBI will be an effective way to monitor habitat quality through the resident katydids.

Katydids are known to be highly cryptic and notoriously difficult to locate in the wild due to their predominantly nocturnal habits. This means that they are not a popular taxon for assessment in comparison with other charismatic invertebrate groups such as dragonflies and butterflies. For this reason, museum records of katydids become a very important source of information. The MANTIS database contains records of all 126 valid species of katydids in South Africa so allowing for the individual species to be assessed for KBI assessments as accurately as possible.

Although cryptic and difficult to locate, katydids are perhaps best known for the species-specific songs produced by mature adult males (Bailey & Rentz 1990). There has been considerable research into monitoring and tracking of katydid species, as well as other acoustically communicating insects, through acoustic monitoring (Riede 1993, 1998; Diwakar & Balakrishnan 2007; Grant & Samways 2016). Acoustic monitoring can be conducted using a variety of techniques, ranging from simple listening exercises (Diwakar et al. 2007) to complex microphone arrays (Blumstein et al. 2011; Marques et al. 2013; Stevenson et al. 2015). In South Africa, acoustic monitoring of katydids is an attractive option as the acoustic environment in which they sing is not such a complex chorus as in tropical forests (Jain et al. 2014). The CFR, in particular, has a simple acoustic community, but very complex Mediterranean-type vegetation structure consisting of a majority of thorny and difficult to access bushes and shrubs. This provides ample hiding space for katydids, and increases the need to detect singing individuals.

In view of these conditions, South African katydids can be monitored using inexpensive and simple equipment. A well-trained listener is able to distinguish between the different calls of both katydid and gryllid species (Diwakar et al. 2007). It is not possible for these listeners to pick up any ultrasonic calls, yet by using a bat detector to scale down ultrasonic katydid calls, real-time identification of these species is possible in the field. Although time is required for the listener to learn the various calls, time will be saved in the long-term as, once a reliable voucher collection with associated song library has been constructed, there will be no need to locate the individual insect to correctly identify it. Simple and relatively cheap recorders are also available for long-term deployment, allowing for passive, non-invasive monitoring that, once an operator is well trained, provides an effective way in which to remotely monitor katydid distributions.

Conclusions

With improved monitoring of katydids, perhaps on a smaller scale and with controlled measuring of environmental parameters, it could be possible to demonstrate the further value of this scoring system as a monitoring technique. This is a preliminary study aimed only to introduce the idea of a rapid assessment method for terrestrial habitats based on katydid song. It has identified some of the advantages of the approach but has emphasized that much more data gathering is required. However, it does appear as if the KBI may be a promising method, particularly for regions where katydids are abundant and diverse, but relatively well-known.

Species Score	Threat (T)	Distribution (D)	Life History Traits (LH)[†]		
			Mobility (M)	*Trophic Level (Tr)*	*M+Tr Sum*
0	LC	Very common: *> 75% coverage of SA and sA*	*Fully-flighted*	*Omnivorous*	0
1	VU	Localized across a wide area in SA, and localized or common in sA: *> 66% in SA and > 66% sA* -OR- Very common in 1-3 provinces of SA and localized or common in sA: *0 - 33% SA and >66% sA*	*Only one sex flighted* *-OR-* *One or both sexes partially flighted*	*Predatory*	1 - 2
2	EN	National SA endemic confined to 3 or more provinces: *> 33% SA* -OR- Widespread in sA but marginal and very rare in SA *< 33% SA and > 66% sA*	*Flightless*	*Herbivorous, polyphagous*	3
3	CR	Endemic or near-endemic and confined to only 1 or 2 SA provinces *< 33% in SA alone*		*Herbivorous, monophagous*	4 - 5

SA=South Africa, Lesotho, and Swaziland and sA = southern Africa (South Africa, Lesotho, Swaziland, Namibia, Botswana and Zimbabwe).

[†] To calculate LH score, M (range 0 - 2) + Tr (range 0 -3) are summed. The sum is assigned a logical species score.

Table 2.1: Katydid Biotic Index calculation method

Figure 2.1: Proportion of South African (a, c, e, g) and Cape Floristic Region (b, d, f, h) katydid assemblages as characterised by the KBI assessment criteria (Threat Status, Distribution, Trophic level and Mobility).

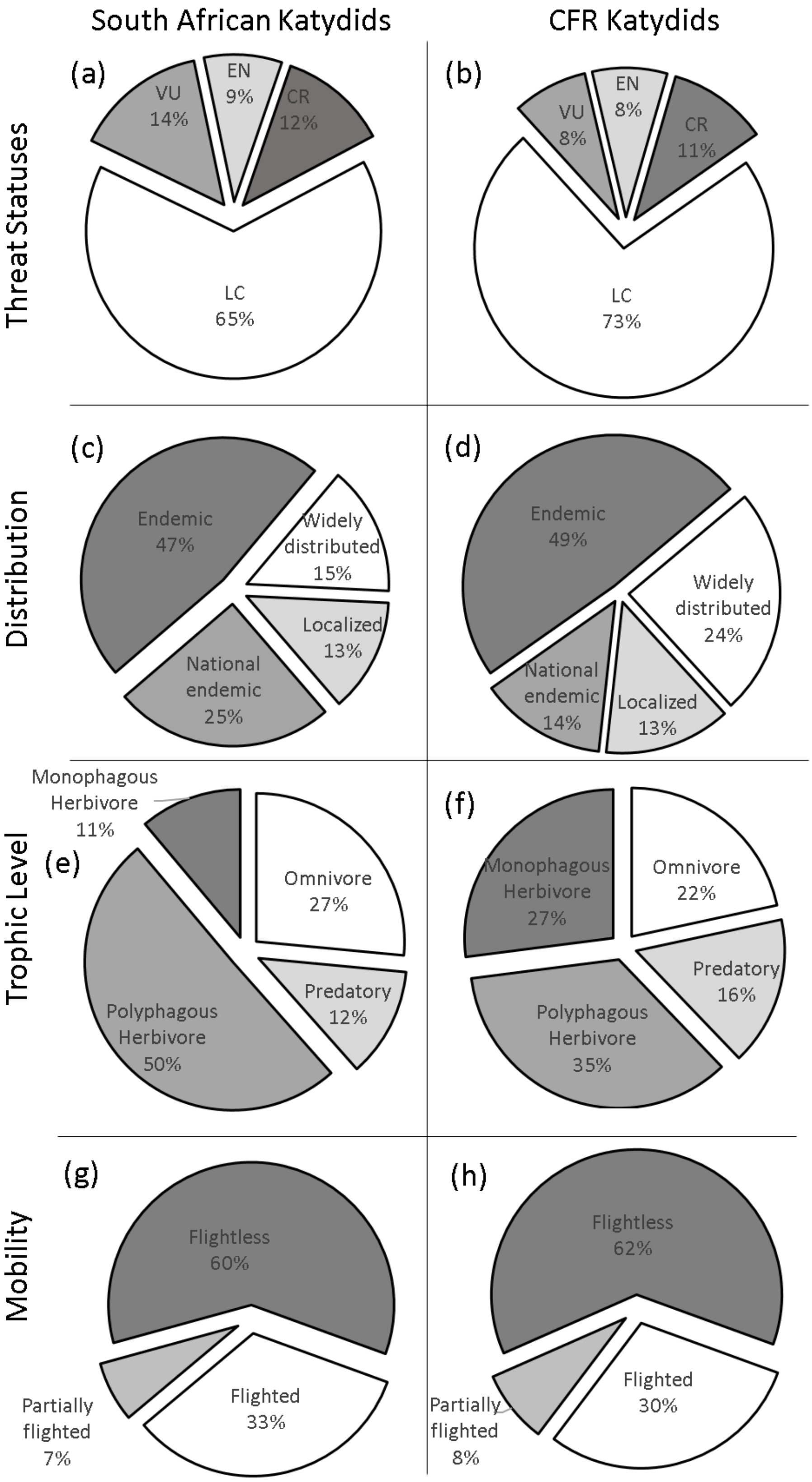


Figure 2.2: Composition of South African (a, c, e) and Cape Floristic Region (b, d, f) katydid assemblages as characterised by their distribution (a, b), mobility (c, d), and trophic level (e, f) relative to their IUCN threat status

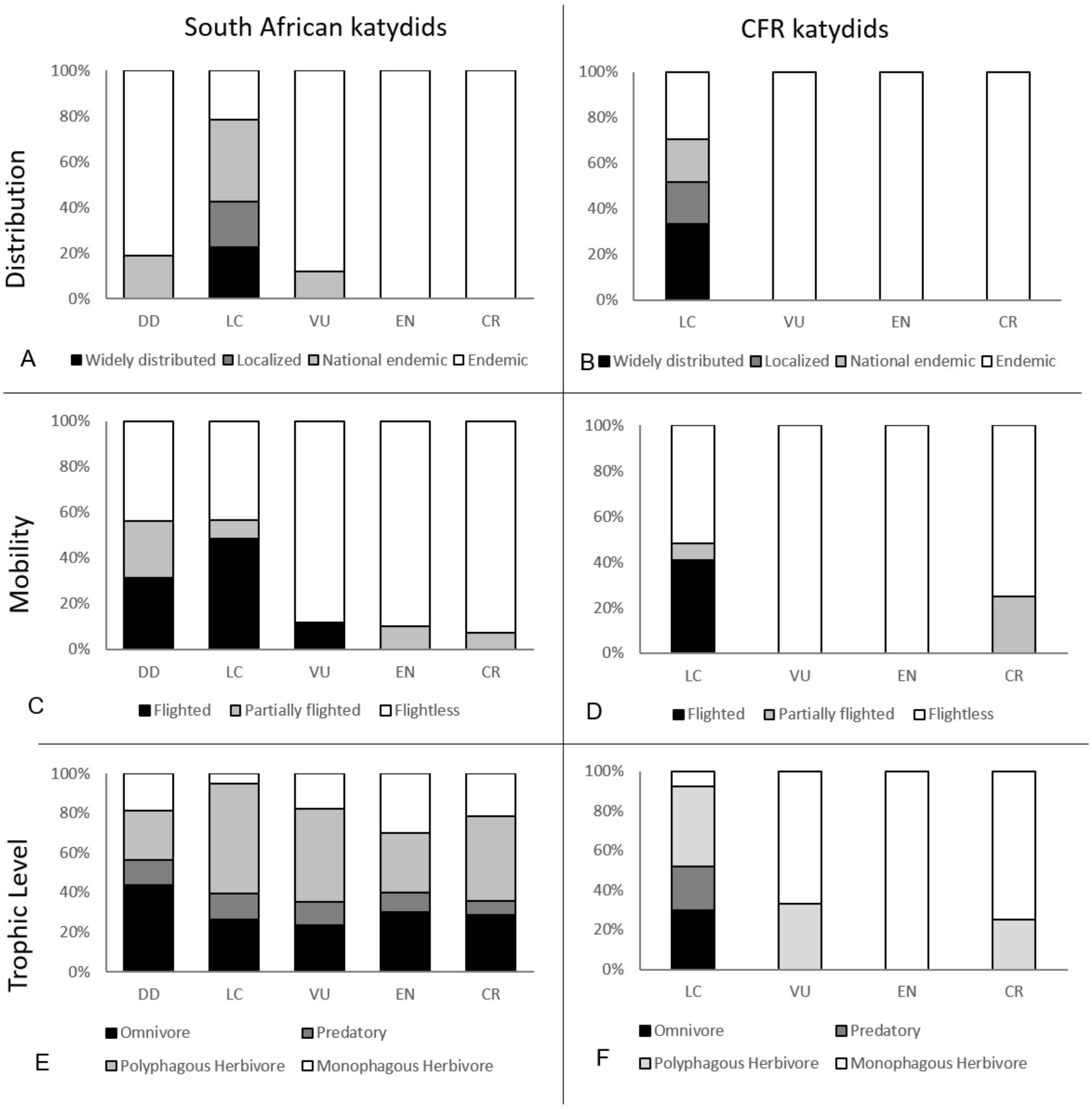


Figure 2.3: Distribution of South African and Cape Floristic Region katydid species among Tettigoniidae subfamilies

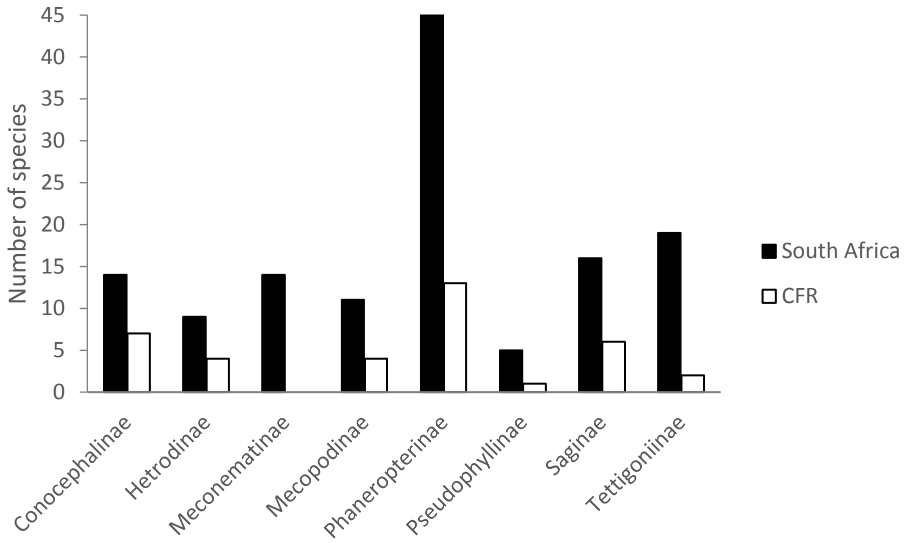


Figure 2.4: Distribution of Katydid Biotic Index (KBI) among ecosystem threat statuses (mean ± s.e.)

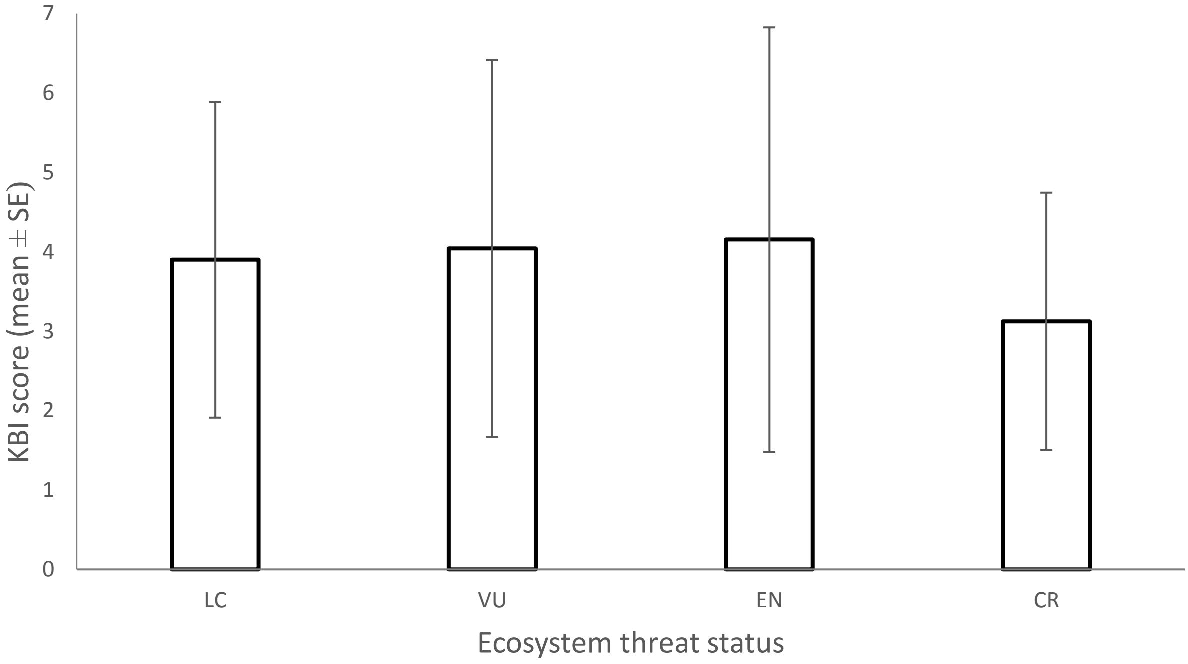
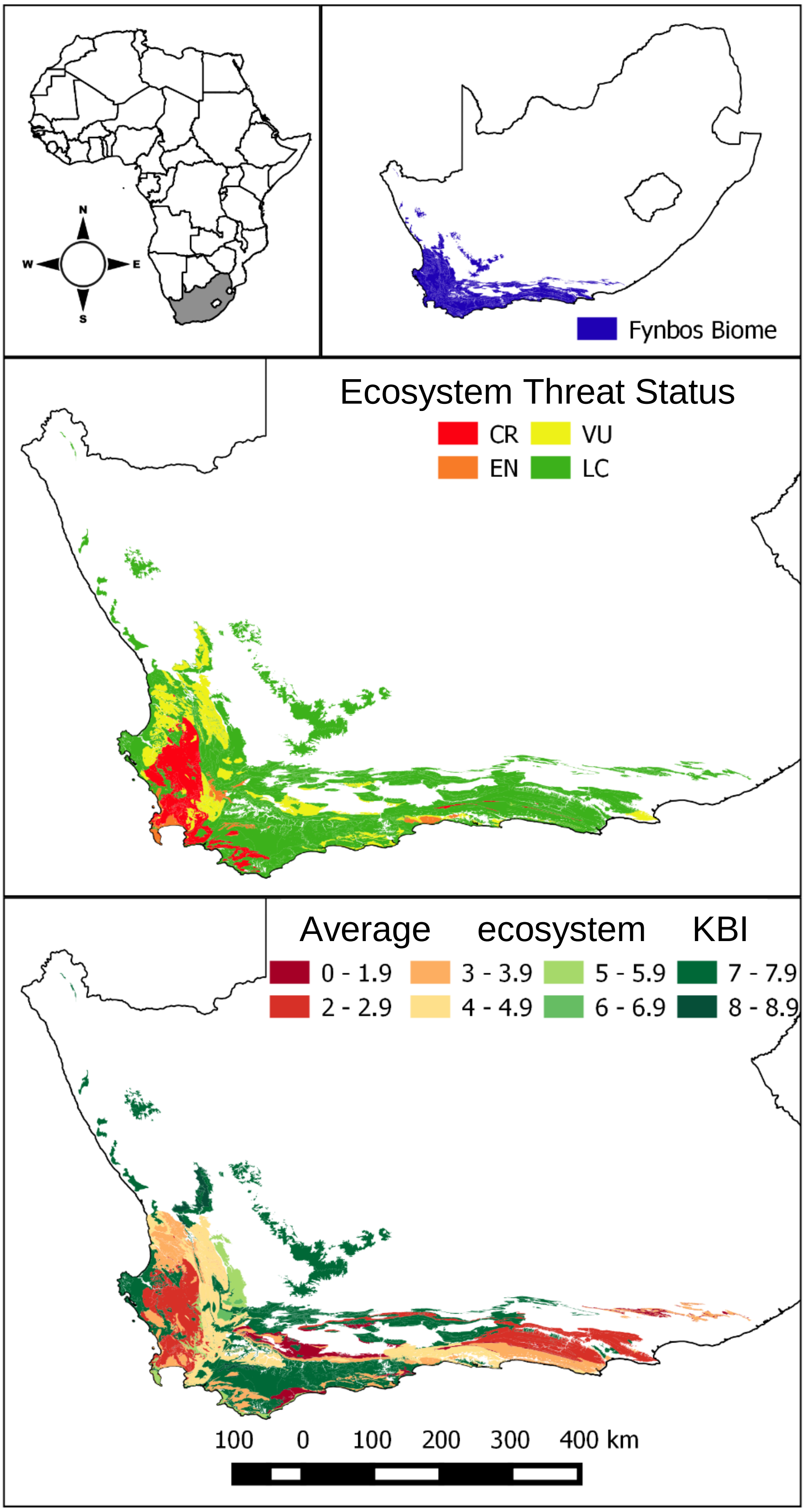


Figure 2.5: Map of ecosystem threat statuses and the average KBI scores (i.e. KBI/Site) of each ecosystem



Chapter 3

Katydids (Orthoptera: Tettigoniidae) respond to habitat quality and not to the zones of a biosphere reserve

Abstract

Biodiversity hotspots are globally renowned for being areas of exceptional biodiversity facing high levels of threat. They are often seen as the best areas for developing new approaches to conservation. The Cape Floristic Region (CFR) covers only 4% of South Africa and yet is one of the most biodiverse areas in the world, but also faces many challenges. The Man and the Biosphere Programme addresses some of the challenges facing global biodiversity by establishing biosphere reserves (BRs), which consist of three zones with sequential increases of permitted human activity and perceived disturbance in each zone – core, buffer, and transition zones. However, little research has compared biodiversity of the zones and evaluated efficient ways to assess this biodiversity. New assessment approaches are required, with non-invasive acoustic monitoring being one possibility, especially using the highly vociferous katydids (Orthoptera: Tettigoniidae). Katydid song profiles were compiled across the three BR zones of the Kogelberg Biosphere Reserve in the CFR over five months. Vegetation quality was also measured to allow for direct comparison of the response of katydids to changing habitat quality. An index of Acoustic Activity was used to determine abundances from recordings. Only 8 species were recorded over this period with no changes in the mean species richness or abundances across the zones. Vegetation quality had a greater effect on abundance of species than on species richness, and the timing of sampling had a significant effect on measured richness and abundance. No individual katydid species can be identified as an indicator species, yet when considered as a whole assemblage, katydids were responsive to habitat quality. This lays the foundation upon which a sensitive and non-invasive tool for habitat assessment can be built.

Introduction

To improve the relationship between humans and the environment, as well as to understand the various factors driving natural processes, the United Nations Educational, Scientific and Cultural Organisation (UNESCO 2016) initiated the Man and the Biosphere (MAB) Programme in 1971. There are both eco-centric and anthropocentric goals that the MAB programme aims to achieve. Of the eco-centric goals, the understanding of interactions between humans and ecosystems is paramount as well as to maintain healthy ecosystems to support both biodiversity and humans(Di Castri et al. 1981).

Currently there are 669 BRs in 120 countries, with each BR comprised of three zones: core, buffer and transition. The purpose of these zones is to delineate the reserves into complementary parts with various functions. The core zone is highly protected and aims to conserve ecosystems, species and associated genetic variation. The buffer zone surrounds and protects the core, and provides a space in which scientific research as well as eco-friendly socioeconomic activities can occur. The transition zone is where the most human activity occurs. This zone can span towns as well as agricultural land, yet all activities within the transition zones must remain ecologically sustainable (UNESCO 2016). If the zones are functioning as intended, one would expect to see increased biodiversity from the transition to buffer to core. However, little empirical research has investigated whether biodiversity responds as expected in the biosphere reserve zones.

Biosphere reserves in South Africa are of particular interest because South Africa is a megadiverse country (Thuiller et al. 2006) and has three biodiversity hotspots, or areas with exceptionally high levels of endemism and that face equally high levels of threat (Mittermeier et al. 2004). Since there are only 35 biodiversity hotspots globally, 10% of all biodiversity hotspots are found within South Africa. Often promoted as the best way in which to choose areas in urgent need of conservation (Forest et al. 2007; Mittermeier et al. 2011), biodiversity hotspot assessments have mostly been based entirely on vertebrate and plant taxa (Myers et al. 2000). It has been assumed that invertebrates are to follow the same trend as plants in these hotspots with high diversity due to the sheer number of host specific insect-plant relationships (Procheş et al. 2009).

Of South Africa's three biodiversity hotspots – Succulent Karoo, Maputoland-Pondoland-Albany, and the Cape Floristic Region (CFR) – the CFR is perhaps the most well-known as it is the smallest of the six globally recognised floral kingdoms (Goldblatt 1997). Covering only 4% of South Africa, the CFR supports an estimated 42% of the country's vascular plants, of which 5622 species are endemic to the region(Goldblatt 1978). However, the CFR is under significant threat due to alien invasive plant species (Higgins et al. 1999; Yelenik et al. 2004; Van Wilgen 2009), habitat fragmentation (Cowling & Bond 1991; Heijnis et al. 1999; Kemper et al. 1999) and anthropogenic climate change (Midgley et al. 2002, 2003; Williams et al. 2005).

The Kogelberg Biosphere Reserve (KBR), the first of eight biosphere reserves in South Africa, was proclaimed in 1998. Although only covering a small portion of the CFR, the KBR is considered a hotspot within the CFR global biodiversity hotspot (Grant & Samways 2011), as there are an estimated 1600 plant species present, of which 150 are endemic to the area (UNESCO 2016). Due to the high floral species richness and endemism in the CFR, and especially within the KBR, it could be expected that similar levels of species richness and endemism of invertebrates might occur there (Procheş & Cowling 2006).

The threats facing the CFR, such as fragmentation, invasion, habitat loss and climate change, are not specific to the CFR flora alone, with invertebrate (French & Major 2001; Donaldson et al. 2002; Pryke & Samways 2010) and vertebrate species also being affected (Macdonald 1992; Richardson & van Wilgen 2004; van der Mescht et al. 2012). The impact of these various threats can be observed in the dragonfly assemblage of the KBR, where the assemblages of the buffer and transition are complementary (in terms of species composition) to that of the core. This suggests that the buffer and transition zones play a role in protecting the core sites and mitigating the effect of the current threats (Grant & Samways 2007, 2011).

Dragonflies have also been shown to be accurate indicators of riparian habitat quality and are used extensively in the Dragonfly Biotic Index (Samways & Simaika 2016). It is therefore possible that other invertebrate taxa of the KBR could be sensitive towards changes in habitat quality (Stork & Eggleton 1992; Foote & Hornung 2005) and thus be complementary across the zones as is the case with the dragonflies (Grant & Samways 2007, 2011). Through the direct observation of terrestrial invertebrate assemblages across a BR it would be possible to determine the existence of complementarity or lack thereof within the species assemblages.

Orthoptera are effective indicators of grassland habitat quality in South Africa's grassland biome (Bazelet & Samways 2011a, 2011b). However, this work focused on Acridoidea in the day-time, which in this region are mostly mute. In contrast, Tettigoniidae in the region are conspicuous singers in the landscape at night and have much merit as bioindicators of landscape quality (Grant & Samways 2016). Traditional biodiversity sampling and assessments overlook katydids as they are nocturnal, highly cryptic (Bailey & Rentz 1990) and difficult to sample as they often avoid light traps, vacuum sampling (Blanton 1990) and pitfall traps (Schirmel et al. 2010) making them difficult organisms to study and collect. Yet their song provides a great opportunity for simple assessments worldwide, with much progress in recent years (Riede 1998; Diwakar et al. 2007; Bormpoudakis et al. 2013).

The success of acoustic monitoring of katydids is due to the males individual species producing species-specific songs (Bailey & Robinson 1971; Nityananda & Balakrishnan 2006) which can then be recorded using a variety of methods, such as microphone arrays (Blumstein et al. 2011), passive recorders (Roca & Proulx 2016), bat detectors for ultrasonic calls (Grant & Samways 2015), as well as through active searching and song recording by hand held microphones (Diwakar & Balakrishnan 2007). Passive recording, perhaps the simplest of the methods, relies on pre-programmed recorders positioned at a site which in turn record the soundscape (the variety of songs emitted across the landscape over a specified period of time). In turn, active searching and identification of katydid species is also possible notwithstanding limitations of the human ear to detect ultrasonic wavelengths (Diwakar et al. 2007).

Acoustic sampling of katydids can be done without having to locate the singing individual. This streamlines the sampling process and allows for both cost-effective and relatively timely sampling (Acevedo & Villanueva-rivera 2006) of katydids in comparison to traditional insect collection methods. Furthermore, acoustic sampling in the CFR seems promising for acoustically communicating taxa, especially as the natural vegetation is often dense, woody and inaccessible which renders traditional sampling methods, such as sweep netting, ineffective (Pryke & Samways 2008).

To date, there are 129 valid species of katydids in South Africa, which is probably not rich in comparison with other areas of the world (Nickle & Castner 1999). Information regarding threat status, life histories and distributions are known for some of these. Many of the species' calls are known through previous acoustic studies conducted within the CFR (Grant & Samways 2011). Katydids are therefore an appropriate study taxon to assess complementarity of the KBR zones for terrestrial insects.

The primary aim of this study is to determine whether the KBR zones are complementary in terms of species composition for a sensitive terrestrial insect group – the katydids. We hypothesize that katydids will respond more strongly to an independent measure of habitat quality than to KBR zones. A Habitat Quality Index is suggested here for the fynbos, as there is no standardized method at present for assessing fynbos quality. Secondly, it aims to determine whether any particular species of katydid within the KBR can be used as accurate indicators of habitat quality. Finally, we make recommendations for the future assessment and conservation of biosphere reserves.

Materials and methods

Study site

Fieldwork was conducted within the KBR (34° 12' 59.43" S, 19° 1' 24.92" E). The KBR covers 103 629 ha of terrestrial and marine habitats. The terrestrial component comprises 30 000 ha core area, 17 000 ha buffer area and 32 000 ha transition area (KBRC 2016; UNESCO 2016). The area is characterised by a Mediterranean-type climate, with hot, dry summers and cool, wet winters. Due to the mountainous terrain of the KBR, orographic rain is common, as well as mists and clouds off the ocean (Boucher 1978).

Katydid sampling

Sampling was conducted from November 2015 to April 2016, in November 2015, mid-January, late February and mid-April 2016. Sampling sessions were separated by roughly six weeks to ensure that all katydid emergence peaks would be sampled. Twenty-six sites within natural vegetation were selected. Ten sites were originally identified in the core but a fire and the burning down of the vegetation in January 2016 resulted in the need to identify three replacement core sites. There were eight sites within the buffer and transition zones respectively (Fig. 3.1). Sites were selected on the relative safety and accessibility, as sites were visited during the night as well as the day. Closely positioned sites were separated by >300 m to prevent overlaps in recorded species in adjacent sites as previous studies found that separation of sites by 100m increased the effective sample area and prevented overlap of recordings across sites (Grant & Samways 2016). The sphere of recording is dependent on amplitude of the signal. For example, a road can be heard from many kilometres away (although road sound interference was not an issue here) and an insect just a few meters. It was therefore decided that 300m was an effective distance between recorders to isolate individual insect calls from each site.

One SM2+ song meter (Wildlife Acoustics, USA) fitted with an omnidirectional microphone was placed at each site. As only five song meters were used for the duration of sampling, it was necessary to move the song meters to new sites every two days to sample all 26 sites. The sample rate of the recordings was set to 48 kHz with the microphone gain set to 6 to enable even the faintest of calls to be recorded. Files were saved as uncompressed wave audio files (.wav files). For each sampling session, the song meters were placed upon a rock or raised feature in the landscape to be higher than the immediate surrounding vegetation and to prevent excessive attenuation of the katydid songs (Forrest 1994; Mankin et al. 2011).

The song meters were programmed to record for 3 min every 30 min between 20h00 and 23h00. Half an hour time intervals were chosen as they proved adequate to capture the entire soundscape, as the nocturnal chorus of katydids does not vary significantly within one evening (Grant & Samways 2016) and was short enough to prevent severe weather changes, such as temperature decreases and windy gusts, from disrupting the species assemblages (R. Balakrishnan pers. comms.). Each site was recorded for two consecutive evenings. Each sampling session therefore resulted in 42 min of recordings per site per session. These sessions made up a total of 72.8 hours of recordings. Ambient temperature was measured at the time of each recording by the song meter, while wind speed was measured using a Testo 410-1 air velocity meter (Testo, Inc. Germany), once per site per session. During each of these site visits, a Pettersson D200 Ultrasound Detector (Pettersson Elektronik, Sweden) was used to locate the ultrasonic species.

In conjunction with the song meters, spot counts were conducted for 10 min at each site, between 20h00 and 21h00, as katydids stay calling from the same site for up to 30 min (Diwakar et al. 2007)**.** A trained listener would sit at a point close to the recorder and note the number of individuals heard singing of each species, and the direction from which they were heard calling. It is not possible to determine the number of individuals calling in single channel recordings (R. Balakrishnan pers. comms.), as individuals cannot be identified so if there are five calls from one species it is impossible to determine whether it id five calls from on individual of the species of five individuals of the same species calling once. Therefore the spot counts were conducted to provide an estimate of species abundances.

Vegetation sampling

Vegetation at each site was surveyed in November 2015. At each site, four 25 m-long transects were laid out at right angles to each other to form an “X”. By placing the song meter in the middle, transects aimed to cover the approximate area that would be recorded by the song meter. The length covered by various plant structural classes was measured. At the same time, 30 random vegetation heights were recorded and averaged. To measure the height, an assistant walked a random directionless route around the site and stopped when the recorder, who was not watching, called “stop”. The nearest plant was then measured at its tallest point with a graduated stick with increments of 10 cm.

Five characteristics related to the vegetation and disturbance of the sites were also recorded: 1) Extent to which all the typical components of fynbos were present (Proteacae, Ericaceae, and Restionaceae); 2) Type and level of anthropogenic disturbance present in the immediate surrounds; 3) Presence of any invasive alien plant species, and their identity; 4) Vegetation diversity and plant dominance; and 5) General fynbos condition. Each characteristic was scored from 0 to 3, with higher scores indicating a more natural habitat condition (Table 3.1). The scores for the five characteristics were then added together and divided by 15, resulting in each site being awarded a value between 0 and 1 that reflects the quality of the habitat at the site. Here this method has been called the Habitat Quality Index (HQI). Sites of higher quality habitat had HQI values nearing 1, while the sites of lower quality had values closer to 0.

Signal analyses

All recordings were analysed using the program Raven Pro 1.5 (Cornell Laboratories, USA). Spectrograms, between 0 kHz and 24 kHz, were produced of each recording to enable the identification of the various acoustic species and guilds. The individual calls were identified, and various acoustic parameters were measured. Voucher specimens were collected of the individual species in the field and kept in an enclosure mimicking the natural environment within the laboratory setting. This encouraged the males to call as they would in the field and enable the human listener to learn the calls of the individual species as well as to record these calls with a hand held microphone for further identification practice. Calls were also identified on the observable patterns within the spectrograms as well as by listening to the individual calls. In total, 1456 recordings were created.

Data analyses

Estimate S (University of Connecticut, USA) was used to calculate a species accumulation curve. This was followed by calculating the Acoustic Activity index (AI), this was done so that an estimate of species abundances could be determined. The method relies on the presence of species in individual time windows and therefore is was possible to adapt the method to the data. As katydids occurrences in each recording were available it is possible to treat each recording as a time window and to then calculate the proportion of samples that contained calls (see Miller 2001 for detailed explination of the calculations). Although this method was originally developed for use with bats, it proved successful for estimating katydid acoustic activity, and provided a more realistic estimation of species abundances from the individual recordings while also reducing the amount of pseudo-replication within the data by preventing one individual from being counted more than once in a recording. The calculated AI values correlated with katydid abundances observed during the spot counts and are a realistic reflection of the number of katydid males observed calling during site visits.

All further statistical analyses were conducted using R (R Development Core Team 2015b). Generalised linear mixed effects models (GLMMs) were run, using the 'glmer' function in the R package Lme4 (Bates et al. 2015), for the species richness and AI of the katydids at each site at each session. The following tests were run to test the hypothesis that katydids respond positively, both in species richness and abundance, to increasing habitat quality. Kruskal-Wallis ANOVAS were run, followed by the nemenyi post hoc test, where appropriate, were conducted with the package PCMR in R (Pohlert 2014b) to test whether HQI differed significantly across the three zones. This was also done for vegetation heterogeneity (the total number of different plant structural types at a site) and average plant height across the zones. ‘Zone’ and ‘site’ were chosen as random variables as it was found that HQI was a more accurate reflection of habitat quality than Zone and site, while HQI, vegetation heterogeneity, average height, session (Nov 2015, Jan 2016, Feb 2016 and Apr 2016) and dominant plant type (the plant structural type that had the highest percentage calculated from the four transects) were chosen as the fixed effects.

To ensure that these analyses were conducted on complete datasets, data from the three sites that burned in January 2016 and the three replacement sites sampled from January 2016 onwards were excluded. Moran’s I and a Mantel test using a Bray-Curtis dissimilarity matrix were used to test for spatial autocorrelation. As there was no indication of spatial autocorrelation, the GLMMs did not need to account for this. A GLMM was run using species richness as the response variable and were calculated with a Poisson distribution and Laplace approximation. A second GLMM using AI as the response variable was run with a negative binomial distribution, with a calculated theta value and Laplace approximation. Tests for significance between the fixed effects of the various models were conducted using an ANOVA which then guided the selection of the appropriate fixed effects. This involved creating an inclusive model and follow up models where successive fixed effects were removed and reinserted. The inclusive model was then tested against each successive model with an ANOVA. When there was a significant difference between the inclusive model and another, the fixed effect that was left was then included in the final model as it had a statistically significant relationship with the response variable.

A complete community matrix consisting of the species present at each site, as well as their respective abundances, was $\log_{10}(x+1)$ transformed and used throughout in order to investigate the species composition across the three zones. First an analysis of similarity (ANOSIM) was conducted. The ANOSIM returns an *R* statistic which explains the difference between groups of samples (Clarke 1993), in this case the katydid assemblage across the three zones. As the *R* statistic approached one, the difference between groups increased. In contrast, an *R* statistic close to zero indicates no difference between the groups. This was then followed by a similarity percentage analysis (SIMPER) to determine which species contribute the most to the similarity and dissimilarity between the zones of the KBR. The differences between the katydid assemblages of the biosphere zones were graphically displayed with a non-metric multidimensional scaling (nMDS) plot. A Bray-Curtis similarity matrix was calculated and used for both the ANOSIM and nMDS. Both the burned sites and replacement core sites were included in these analyses as excluding them had no effect on the results.

Finally, to determine whether any specific katydid species could be used as indicator species, the three most dominant species across all three zones were selected. This would help to determine if a particular species prefers sites dominated by native fynbos or invaded sites by determining the abundances observed on the different plant types. Initially, species-specific GLMMs, with a negative binomial distribution and a calculated theta value, were run. This was then followed by testing the species response to the most dominant grouped plant types with the use of a Kruskal-Wallis ANOVA. Once again the burned and replacement sites were excluded from these analyses.

Results

Across the entire four fieldwork sessions, 10 876 calls were identified belonging to katydids, crickets, frogs, grasshoppers and birds. Of these, 2596 calls were attributed to katydids. Across all sites and zones, the species *Ceresia* sp. 1 had the highest number of calls at 1174 calls, *Plangia graminea* was the second most call abundant at 871 calls, and *Ruspolia* sp. was third with 30 calls. These three species were spread across most sites and across all three zones. The remainder of the calls were attributed to the following species: *Clonia melanoptera* (6), while the following species were only recorded from one site each: Tettigoniidae sp. 1 (425), *Ceresia* sp. 2 (73), Tettigoniidae sp. 2 (19) and *Hetrodus pupus* (2) (Appendix 3.1). Of the 1456 possible recordings, 32.2% (469 recordings) were not used for a variety of reasons such as there being no calls present in the recordings, or wind and rain noise creating a significant amount of interference which rendered calls unidentifiable. Species were successfully identified based on call structures visualised in the spectrograms (Appendix 3.2) as well as by listening to the individually measured calls. The species accumulation curve (Fig. 3.2) reached an asymptote of eight species, suggesting that adequate sampling was conducted.

Ruspolia sp. is the only species whose AI was not correlated to the spot count abundances due to it only being observed in the recordings and was not heard during the spot counts. As a result of the correlation between the AI values and the observed numbers of the actual katydids singing in the field, it was decided that the AI values would be used as an abundance measure rather than the number of calls counted in each recording as the counted values would be highly inflated.

There were no significant differences between the mean species richness and abundance of the katydids across the core, buffer and transition zones ($X^2_{\text{species richness}} = 0.26$; $p_{\text{species richness}} > 0.05$ and $X^2_{\text{abundance}} = 1.67$; $p_{\text{abundance}} > 0.05$) (Fig. 3.3). When the fieldwork session was considered, there was a slight but insignificant increase in species richness between November and January, which then decreased towards April. This trend was not as apparent in the abundance as the sampling progressed from November 2015 to April 2016. There was a slight increase over January and February, with November and April being slightly lower (Fig. 3.4). From preliminary investigations it appears as if there are no significant differences between the abundances as well as species richness across the different HQI's of the sites nor across the vegetation heterogeneity of the sites.

From Moran's I (Observed I = - 0.061, p = 0.75) and the simulated p-value of the Mantel test (R = - 0.14, p = 0.99) for spatial autocorrelation, the observed spatial distribution of the katydids across the sites did not differ from the expected, and therefore spatial autocorrelation was not present. The GLMMs could therefore be run without correcting for spatial autocorrelation.

Since the zones of the KBR had little effect on the katydid assemblage, zone was considered a random effect and HQI was considered as a fixed effect. This was further supported by the results of the Kruskal-Wallis ANOVA and posthoc nemenyi test for HQI across the three zones (Fig. 3.5). The median HQI of the transition sites was significantly lower than that of the core sites ($X^2{}_2$= 14.9, p = 0.001). The core and buffer sites did not differ significantly from each other. Vegetation heterogeneity did not differ significantly across the three zones (X^2= 0.72; p > 0.05), although average height did (X^2= 9.43; p < 0.05). The average height of the core sites was significantly lower than that of the transition sites and was almost significantly lower than that of the buffer, while the average height of the transition and buffer did not differ significantly.

Katydid species richness was significantly affected by session ($X^2{}_3$= 14.61, p = 0.002), while HQI was nearing significance ($X^2{}_1$= 2.96, p = 0.08). Although not a statistically significant relationship ($X^2{}_1$= 1.52, p = 0.2), average height was included into the final model as other previously tested models showed average height to be significant for katydid species richness. The inclusion of these three variables into the final model improved the model fit as the AIC value of the original model was higher than that of the final model (original model: $AIC_{\text{HQI + Veg heterogeneity + AveHeight + Session + Dominant plant type}}$ = 222.28; final model: $AIC_{\text{HQI + AveHeight + Session}}$ = 214.5).

Katydid abundance responded in a similar way to katydid species richness: HQI, average height and fieldwork session all had a statistically significant relationship ($X^2{}_1$= 4.64, p = 0.03; $X^2{}_1$= 7.71, p = 0.005 and $X^2{}_3$= 12.12, p = 0.007 respectively). Again, inclusion of the same three variables resulted in the best fit of the model (original model: $AIC_{\text{HQI + Veg heterogeneity + AveHeight + Session + Dominant plant type +}}$ = 455.19; final model: $AIC_{\text{HQI + AveHeight + Session}}$ = 451.3).

From the ANOSIM, it was apparent that there were no significant differences in the katydid species composition between the zones (R= 0.009; p = 0.383; Fig. 3.6) and therefore the assemblages were not complementary. Across the zones, *Ceresia* sp. 1, *P. graminea* and *Ruspolia* sp., were selected repeatedly by the SIMPER analysis and in the same order, implying that these species contribute the most towards similarity between zones (Table 3.2). The nMDS showed that most of the points clustered around 0 and no other patterns emerged.

The only significant environmental variable for the katydid assemblage composition to emerge from the nMDS test was that of vegetation heterogeneity (Fig. 3.7).

The mean abundances of *Ceresia* sp. 1, *P. graminea* and *Rusopolia* sp. did not differ across the zones of the KBR (Fig. 3.8). Also, there were no significant differences between the mean abundances of these three species across the dominant plant groups at each site (Fig. 3.9). For all three species, the inclusion of HQI and average height improved the fit of the models yet it was only for *Ceresia* sp. 1 where there was a positive statistically significant relationship between abundance and average vegetation height ($Z_1 = 2.46$, $p = 0.014$).

Discussion

Only eight katydid species were identified from the recordings spanning five months. This low number was not expected as the Kogelberg Biosphere Reserve is in one of the most florally diverse areas of the world (Myers et al. 2000; Orme et al. 2005). When planning this study, it was estimated that there would be >25 katydid species, based upon records from the MANTIS database (Naskrecki 2008), containing collection localities for katydids within South Africa. Although a low number of species, the accumulation curves nevertheless suggested sufficient sampling (Gotelli & Colwell 2001), spanning the summer months and sampling all emergence peaks. Therefore, the presence of only eight species of katydids across all the recordings can be considered an accurate assessment of the species richness for the area.

Another reason for there being fewer than the expected species is that only the males of the various species call to attract mates and so other species may have been overlooked if protogyny is present in the species. This is unlikely as it has been shown that sperm competition in males of *Requana verticalis* drives male mate selection, and protandry rather than protogyny is established (Simmons et al. 1993). Katydid species call at various frequencies (Morris & Beier 1982; Schul & Patterson 2003) and it is possible that some species were present yet called at frequencies higher than the microphone was able to record (24 kHz). However, this is unlikely as the bat detector would have located these truly ultrasonic species. The bat detector did not pick up any additional calls over those recorded by the microphone and song meter. This suggests that all species within the KBR are easy to locate and identify without the help of additional (ultrasonic) equipment.

The most common survey methods used to determine species abundances are count-based methods relying on sightings of the different individuals of a species or on mark-recapture methods (Marques et al. 2013). As katydids are generally small, cryptic, difficult to collect, as well as being predominantly nocturnal (Bailey & Rentz 1990, Balakrishnan 2016, applying these traditional methods to obtain a robust estimate of abundance is a time consuming process. Passive acoustic monitoring of katydids allows for easy identification of the different species present at a site, yet determining the abundances of the individual species from these data remains a substantial challenge (Marques et al. 2013). The human ear can reliably determine the different species and number of calls when in the field (Diwakar et al. 2007). Although this is not the case when analyzing single channel recordings, as neither the human ear nor a computer algorithm can triangulate the direction and number of calls in this one-dimensional sound space (R. Balakrishnan pers. comms.). By considering each recording as a time window, it is possible to apply the methods outlined in Miller (2001) to calculate a relative Acoustic Activity Index for each species at each site each season. Although not a true abundance measure, it does correlate to the abundances counted during the spot counts, indicating that it is a reliable estimate of abundance.

It may be argued that spot counts are the best method to determine species abundance of katydids and other acoustic species as it is proven that a trained human ear is excellent at resolving the number and direction of calls in the field (Diwakar et al. 2007). Due consideration was given to human fatigue: it was only possible to visit <4 sites/night to conduct the spot counts before become overly fatigued. In this case, relying on song meters is the best method to employ since over the two night sampling rotations of this study, five song meters were able to sample for a total of 3.5 hours while the observer was only able to sample for 50 min over the two night period. Passive recordings conducted with song meters are therefore the most time saving and cost-effective method for sampling field acoustic signals (Sueur et al. 2008) as it requires few human-hours to set up and many recorders can be deployed across multiple sites. In comparison, if a listener were to visit five sites for a total of 42 min over two consecutive nights across four fieldwork sessions it would be an expensive, time-consuming and complex logistical challenge. By employing passive recordings and the AI method, the time taken to sample five sites is greatly reduced and allows for many more sites to be sampled so increasing replication and size of area sampled.

The zones which comprise a biosphere reserve are a unique feature of these reserves, yet there is no evidence for the existence of complementarity between the katydid assemblages of the three zones. This was unexpected as dragonflies show complementarity as a result of changing managerial practices across the three zones (Grant & Samways 2007, 2011). This may also be as a result of the very small numbers of species that were found across the KBR, which could prevent finer resolution differences emerging from the dataset (Magurran 2003). The evidence points to there being little turnover between the katydid species even though this study was conducted in a biodiversity hotspot.

It is assumed that the habitat quality would decrease from core to buffer to transition as a direct result of the decreasing protection levels of the zones (UNESCO 2016). Habitat Quality Index does just that, although no significant difference was found between core and buffer, the core differed significantly from the transition zone. Average height was significantly lower in the core compared to the transition zone, yet it is unknown whether it is the result of changing habitat types or vegetation quality. There were no discernable patterns in the vegetation heterogeneity of the various zones and instead this could be as a direct result of turn over between plant species across the sites. The typical fynbos vegetation of the CFR is characterized by extremely high levels of beta-diversity, resulting in the complete change of plant species composition yet the maintenance of plant richness across very closely related sites (Cowling 1990). As only plant structure was measured at each site, it is not possible to say that turnover as a result of beta-diversity was responsible for the lack of patterns. Since the patterns observed did not follow the expected trend for the three zones, it is possible to disregard the effect of zone. This is especially valid in view of the fact that the boundaries of these zones are artificial and generally arbitrarily delineated (UNESCO 2016) and do not take into regard natural processes or habitat quality at the different sites.

The HQI is a standardized method by which all sites were assessed. It consisted of five categories that were independently assessed and therefore provided an accurate reflection of the actual habitat quality at each site. The Habitat Quality Index is therefore better suited than the coarse scale and non-standardized biosphere reserve zones for comparison with any patterns emerging within the katydid assemblage.

The month of sampling had an effect on the katydid assemblage, with phonological succession well known within the group, and with activity peaking during the summer months (September to May in the southern hemisphere) (Grant & Samways 2016). Preliminary sampling that was conducted in September 2015 with the aim of sampling the emergence peak of early spring species of *Brinckiella*, only resulted in the collection of nymphs of *Clonia melanoptera* and *Ceresia* sp. 1. Even though the increases in species richness and abundance across the sessions were not significant, they still followed the same pattern that other South African katydids show: a peak during the middle of summer (December to February) and then a gradual decrease with the approach of winter. These changes would possibly be more apparent in an assemblage comprised of >8 species as greater refinement between emergence peaks would be possible (Magurran 2003).

The apparent lack of spatial autocorrelation in the dataset can be explained by the presence of only eight species in total, with the three most common species being found at virtually all sites. As the KBR covers a small area of the CFR, and as all sites were relatively close together, it was not expected that there would be no spatial autocorrelation in the data (Legendre 1993).

It is interesting that species richness and the AI of the various species across the KBR all responded to the same variables, not just the fieldwork session. As the HQI of a site increased, so too did the AI of the various species present at the individual sites also increase. It is likely that better quality habitat can support a larger number of individuals when all habitat requirements and resources are present (Bieringer & Zulka 2003; Unsicker et al. 2010). Species richness only takes into account the number of individual species and not the fluctuations in the numbers of individuals of each species at each site (Magurran 2003). Assemblage composition is a better source of information, as it refers to the actual species present, which is especially important when investigating an assemblage with few species, as in the case of the katydid species here in the KBR. The more generic approaches overlook smaller changes in composition and response to environmental variables and can lead to incorrect conclusions (Magurran 2003).

When comparing the entire katydid assemblage and taking into account the species richness as their various abundances at each sites, vegetation heterogeneity is the only factor that showed any trend. This was not expected as the study sites covered a range of habitat and plant assemblage types and so there should have been recognizable differences between the katydid assemblages at the different sites (Kemp et al. 1990). It was also expected that the different plant structural assemblages, not just number of plant types, would have some effect on the katydids here. Although there were some notable exceptions where several species were unique to particular sites. In particular *Ceresia* sp. 2 was found in three seasons at one core site. This species has a distinctly different call pattern to the similar *Ceresia* sp. 1 but is not as widely spread.

Katydids are known to respond positively towards vegetation structure rather than the individual plant species. For example, members of the morphologically similar genus *Platycleis* katydids in Western Europe have been shown to all have very specific habitat preferences (Samways 1976). This means that by simply identifying the local vegetation present, it is possible to deduce, with relative accuracy, the different katydid species that would be present at a site. Even when sites are categorized broadly by the dominant plant structural types, the katydid species in the KBR did not select for structural composition as is normally found among katydids. If this were the case, *Ruspolia* sp. would only have been found in grassy sites and *Ceresia* sp. 1 in dense shrubby sites. Yet no apparent preferences for vegetation structure among the sampled katydids were found.

Perhaps the climate is playing a role as well as a long history of natural selection. It is known that *Betiscoides* grasshoppers, which are CFR endemics, are very susceptible to weather conditions with the cool, wet, windy winters and the hot, dry summers apparently being stressful (Matenaar et al. 2014). Interestingly, the low species richness, fairly low abundance yet generalized habitat selection seems to be emerging for Orthoptera in general in the area (Matenaar et al. 2014; Pronk 2016), which may also be interrelated with the high fire frequencies in the CFR (about every 12 years) which temporarily removes all vegetation equally (leading to temporal biotope homogenization), and even leads to fire melanisim in some acridids (Schlettwein and Giliomee 1987).

Ceresia sp. 1, *P. graminea* and *Ruspolia* sp. were not effective indicators of habitat quality. If they were sensitive towards changing habitat quality it could be expected that the HQI of a site would have a positive significant effect on the abundances. Yet the results indicated that no particular factors influenced the abundances of the species at the different sites. Nor did these three species respond to any particular plant structural type. Since the effect of habitat quality and other factors are only apparent when the entire katydid assemblage is considered, it is not possible to say that one particular species is a better or more accurate indicator than the other species or that the entire katydid assemblage is a successful indicator of habitat quality.

Conclusions

Katydids are not complementary across the zones of the Kogelberg Biosphere Reserve but rather respond as an entire assemblage to habitat quality. For future studies on katydids to be conducted, habitat quality is essential and needs to be measured in a standardised manner. Overall abundance of katydids has proven to be a more inclusive way in which to analyse the data as it allows for fluctuations in the individual populations to be taken into account. Patterns only start to emerge with regards to katydid responses to habitat quality when the assemblages are monitored for the entire summer season. As katydids emerge at different times, sampling over shorter temporal windows will result in in abundances varying and inaccurate conclusions being drawn as a result of the varied abundances observed.

Little research has been done in Biosphere Reserves and it for this reason that managers of these areas should rather approach conservation in line with the precautionary principle: when scientific knowledge is lacking, decision-making becomes a social responsibility to ensure that the environment and people involved are safeguarded from harm. This approach may not seem to be applicable to the katydids of the KBR as they appear to be resilient to habitat transformation. Although there is no adequate information to support the low diversity of species in the reserve. Perhaps this is a result of poor past management decisions and we are actually observing an extinction debt being repaid.

Score	**Characteristic 1:** Presence of fynbos components	**Characteristic 2:** Extent of disturbance	**Characteristic 3:** Extent of alien invasion	**Characteristic 4:** Plant community dominance	**Characteristic 5:** Observed fynbos condition
0	No components present	Three or more sources of disturbance* in a 150 m radius	Complete invasion of the site	Monospecific site	Poor quality fynbos, unhealthy
1	One component present	Two sources of disturbance in a 150 m radius	Dense patches of alien species or more than one species present	Almost dominated but with a few small diverse patches	Fynbos mixed with other vegetation components alien to the fynbos. Does not appear to be healthy.
2	Two components present	One source of obvious disturbance in a 150 m radius	Scattering of alien species or only one species present	Diverse with patches dominated by other species	Average fynbos, not all components present but still indigenous and appears healthy
3	All components present	Apparent lack of anthropogenic disturbance	No alien species present	Diverse	Good quality fynbos, diverse and robust

*Disturbance is defined here as anything resulting from human activity, i.e. hiking paths, roads, farmland, plantations etc.

Table 3.1: Assessment rubric for the Habitat Quality Index

	Ceresia sp. 1	*Plangia graminea*	*Ruspolia* sp.
Core x Buffer	0.37	0.65	0.85
Core x Transition	0.38	0.63	0.81
Buffer x Transition	0.34	0.68	0.8

Table 3.2: Cumulative contributions of the most influential species across the various assemblages within the KBR zones, calculated using the Bray-Curtis dissimilarities. Values represent the average contributions of each species towards the overall Bray-Curtis dissimilarity from the simper analysis.

Figure 3.1: Map of the Kogelberg Biosphere Reserve and sampling sites

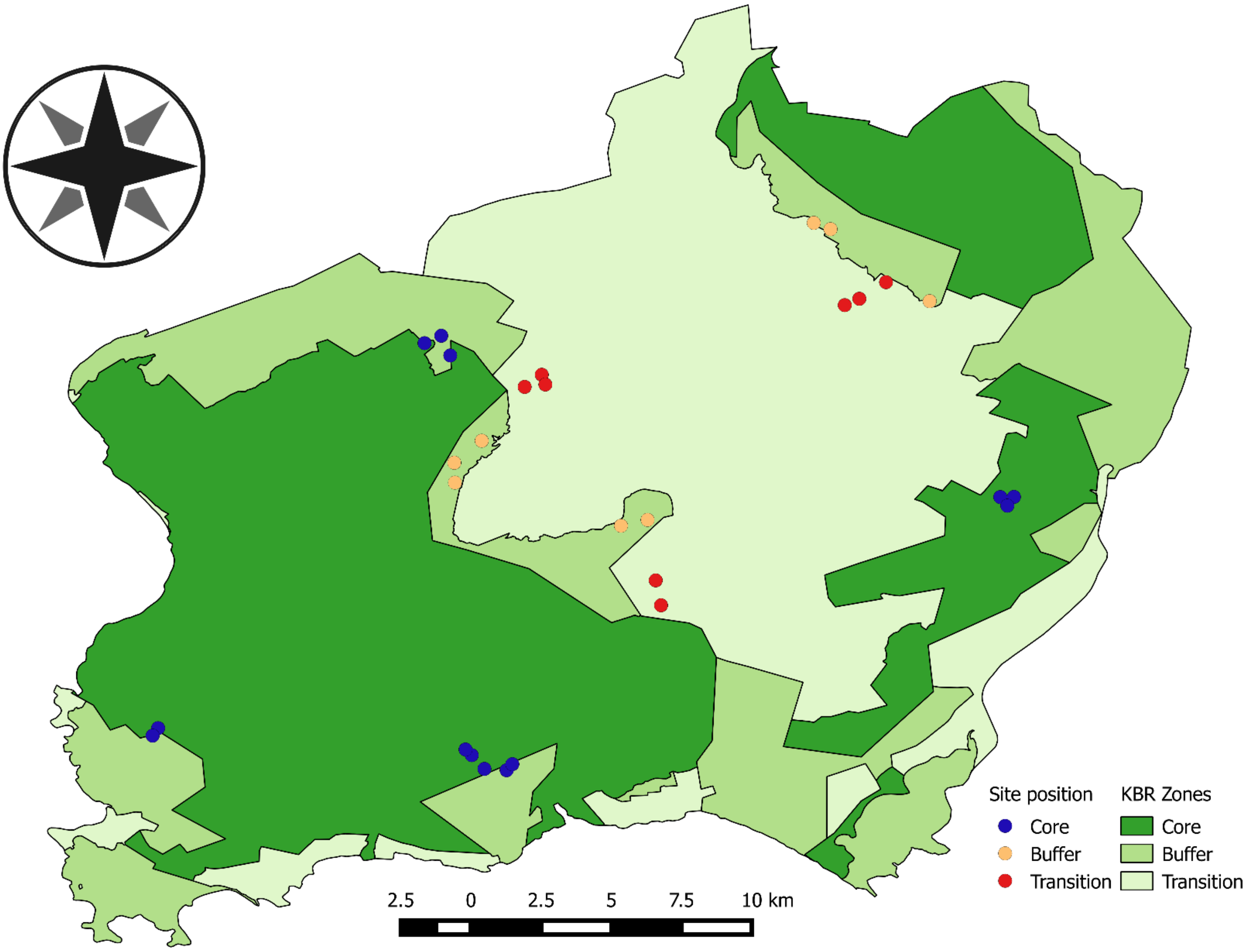


Figure 3.2: Species accumulation curve based on the katydid species observed in each night at each site

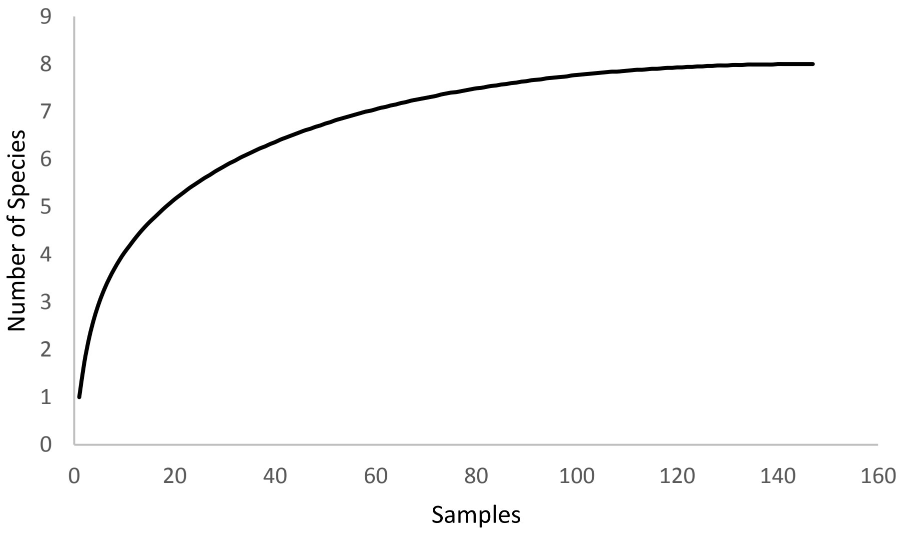


Figure 3.3: Mean abundance and species richness of the katydids across the three zones of the KBR

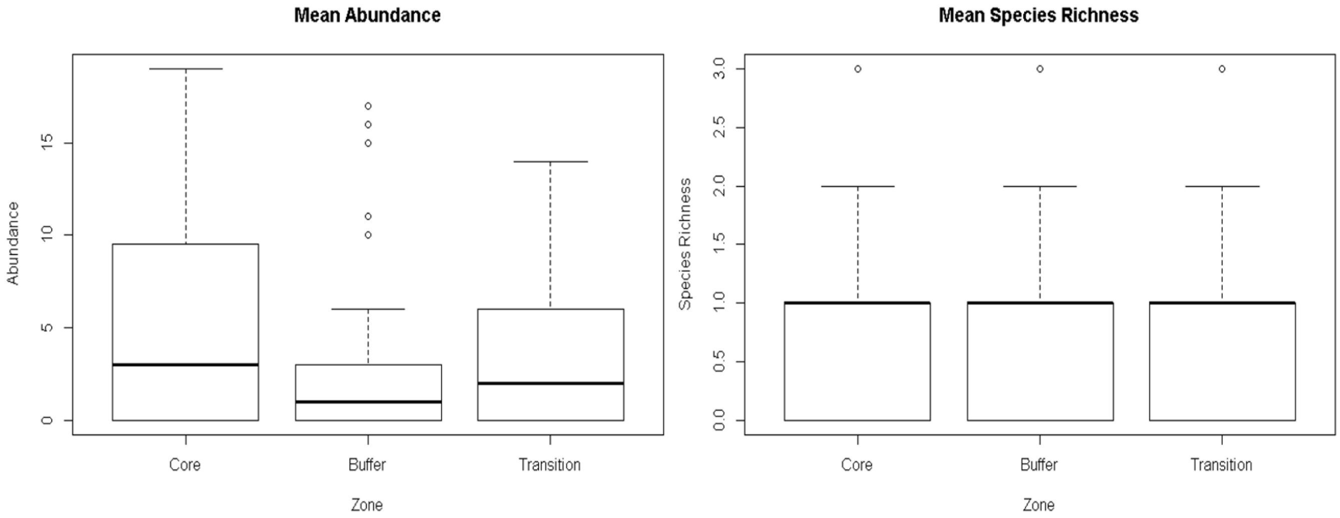


Figure 3.4: Mean abundance and species richness of katydids across the sampling sessions

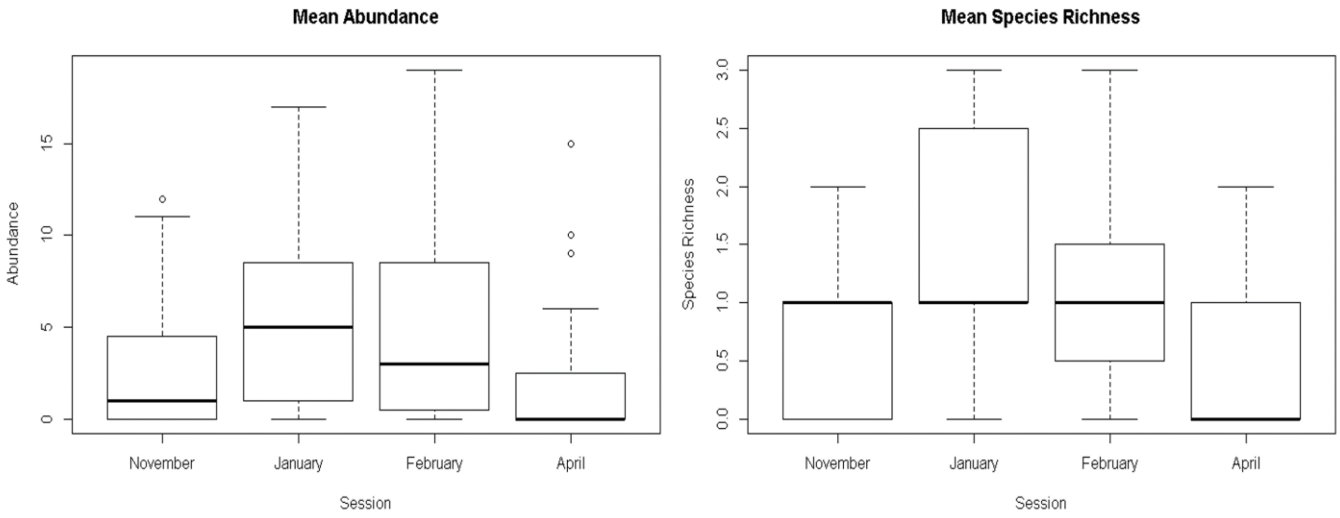


Figure 3.5: Mean HQI, plant height and vegetation heterogeneity of the three zones of the Kogelberg Biosphere Reserve. * p< 0.05; ** p< 0.01; ***p<0.001

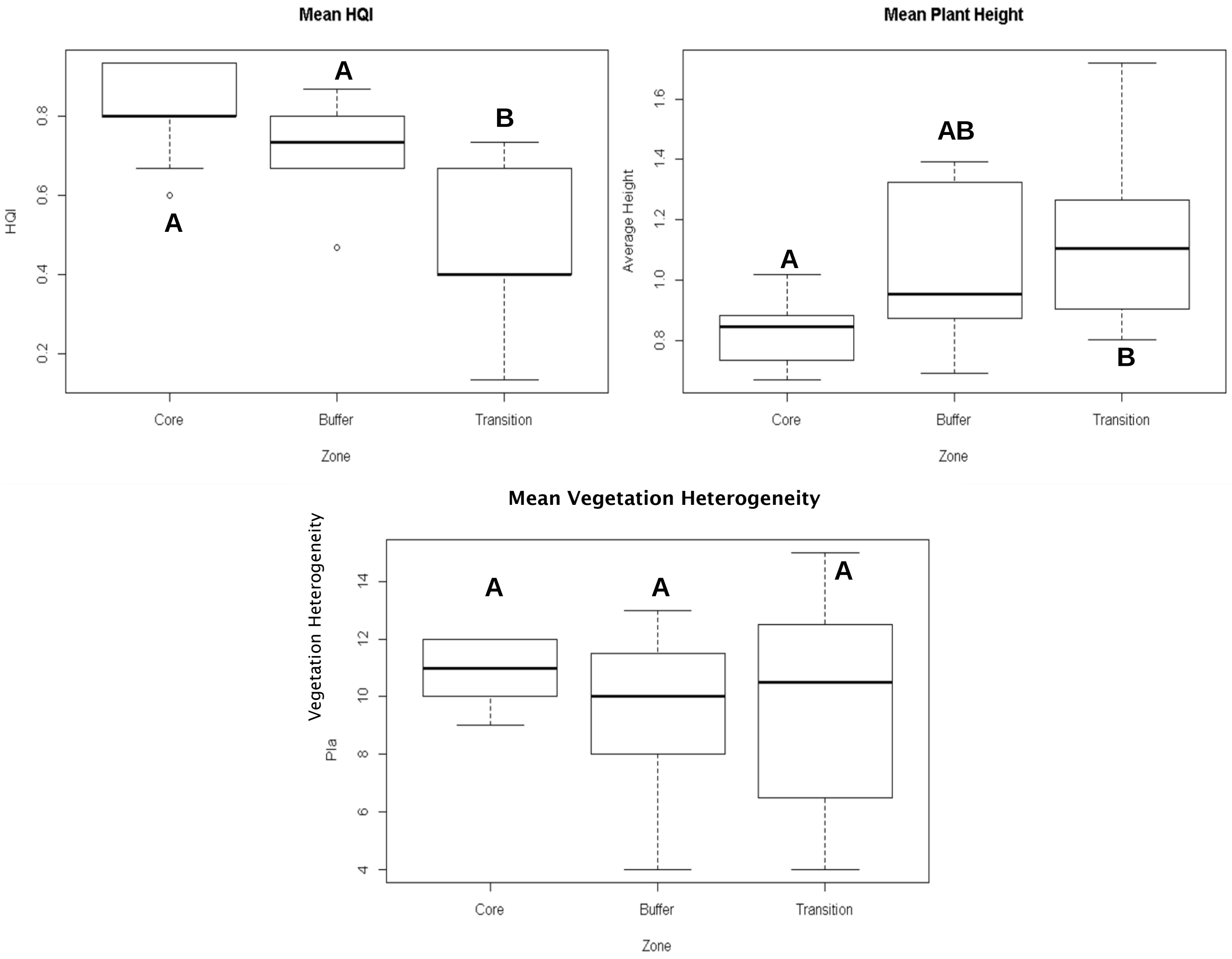


Figure 3.6: Variance between the species composition of the three zones from ANOSIM and nMDS

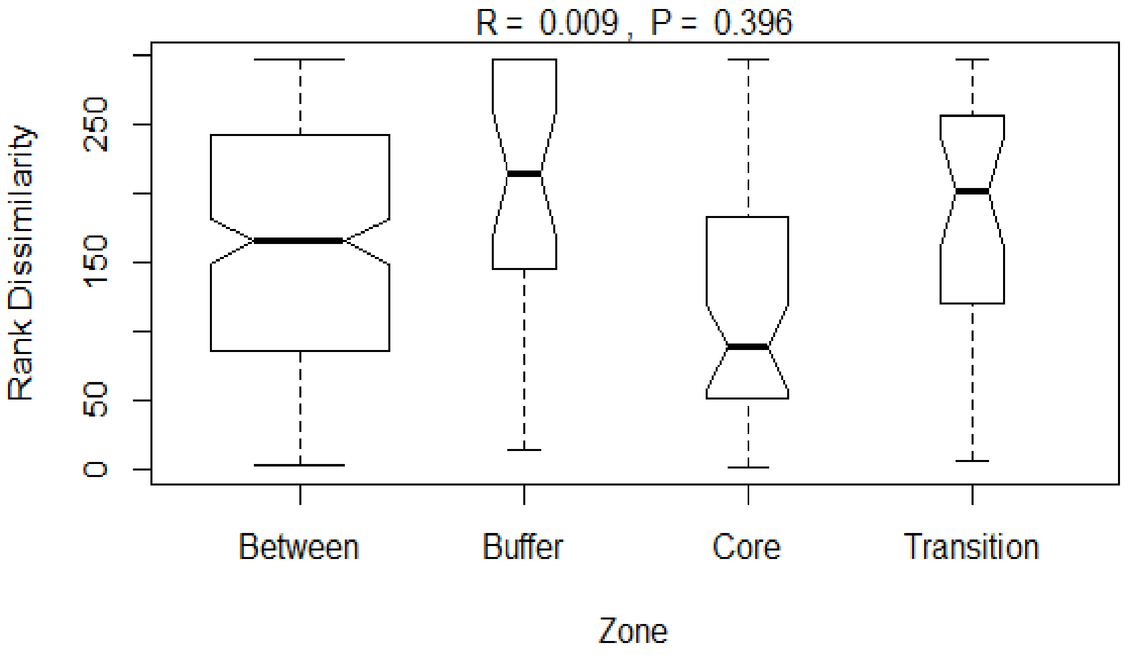


Figure 3.7: nMDS plot for the katydid community composition with zone as the grouping factor. Black circles indicate the sites and red dots the species. VegHet: vegetation heterogeneity; AveH: average vegetation height; HQI: Habitat Quality Index

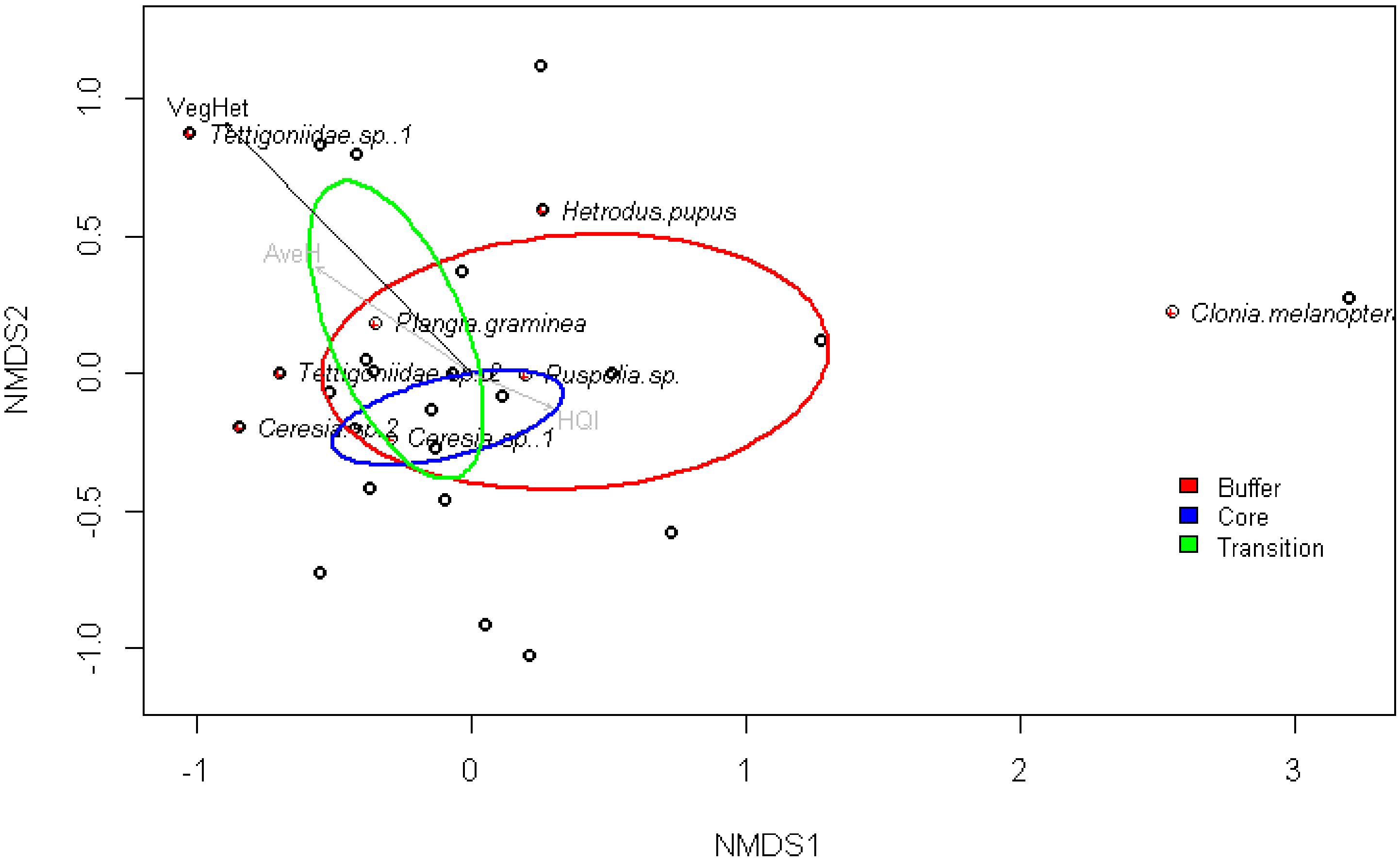


Figure 3.8: Mean abundances of the three most common katydid species across the three zones of the KBR

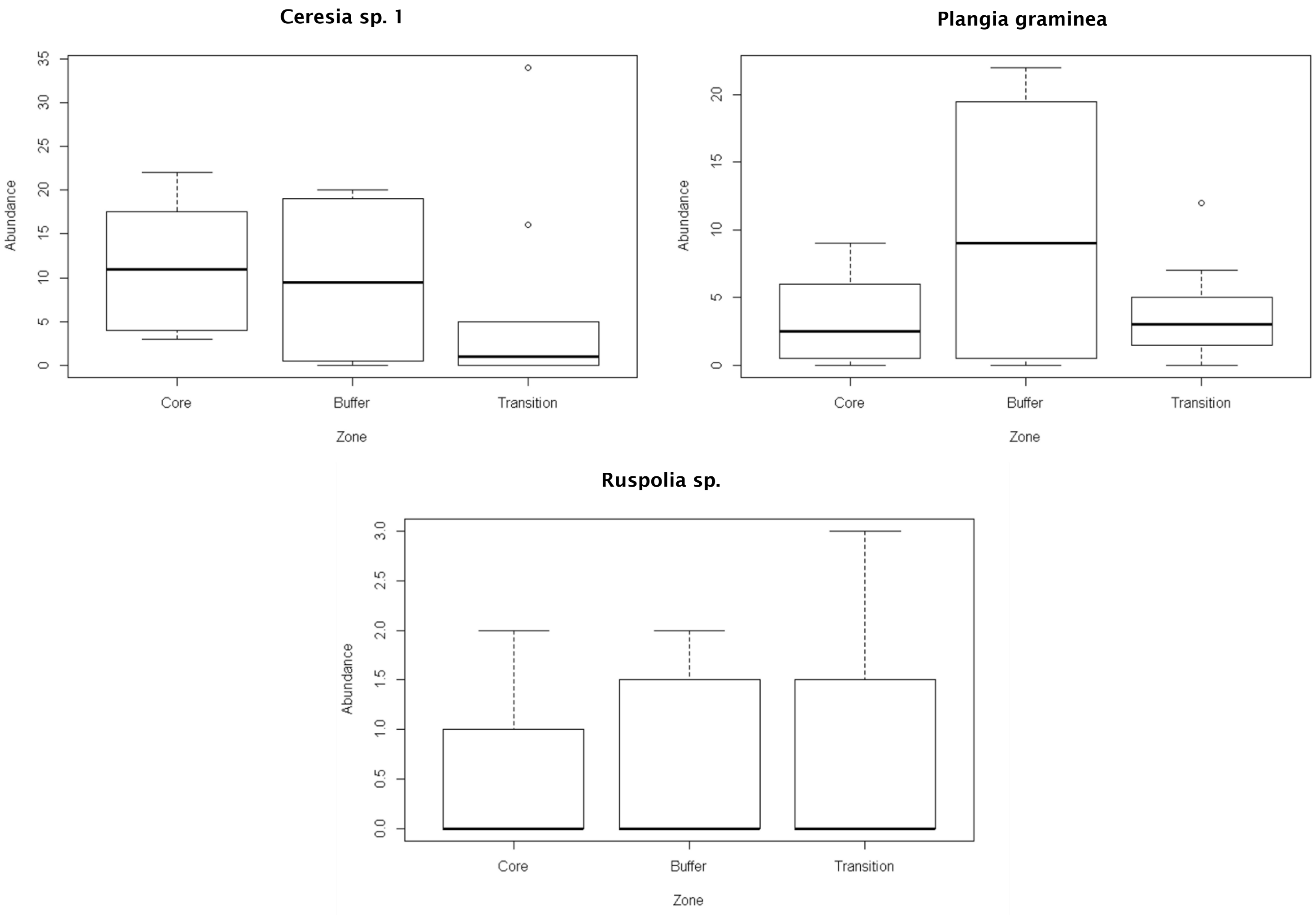
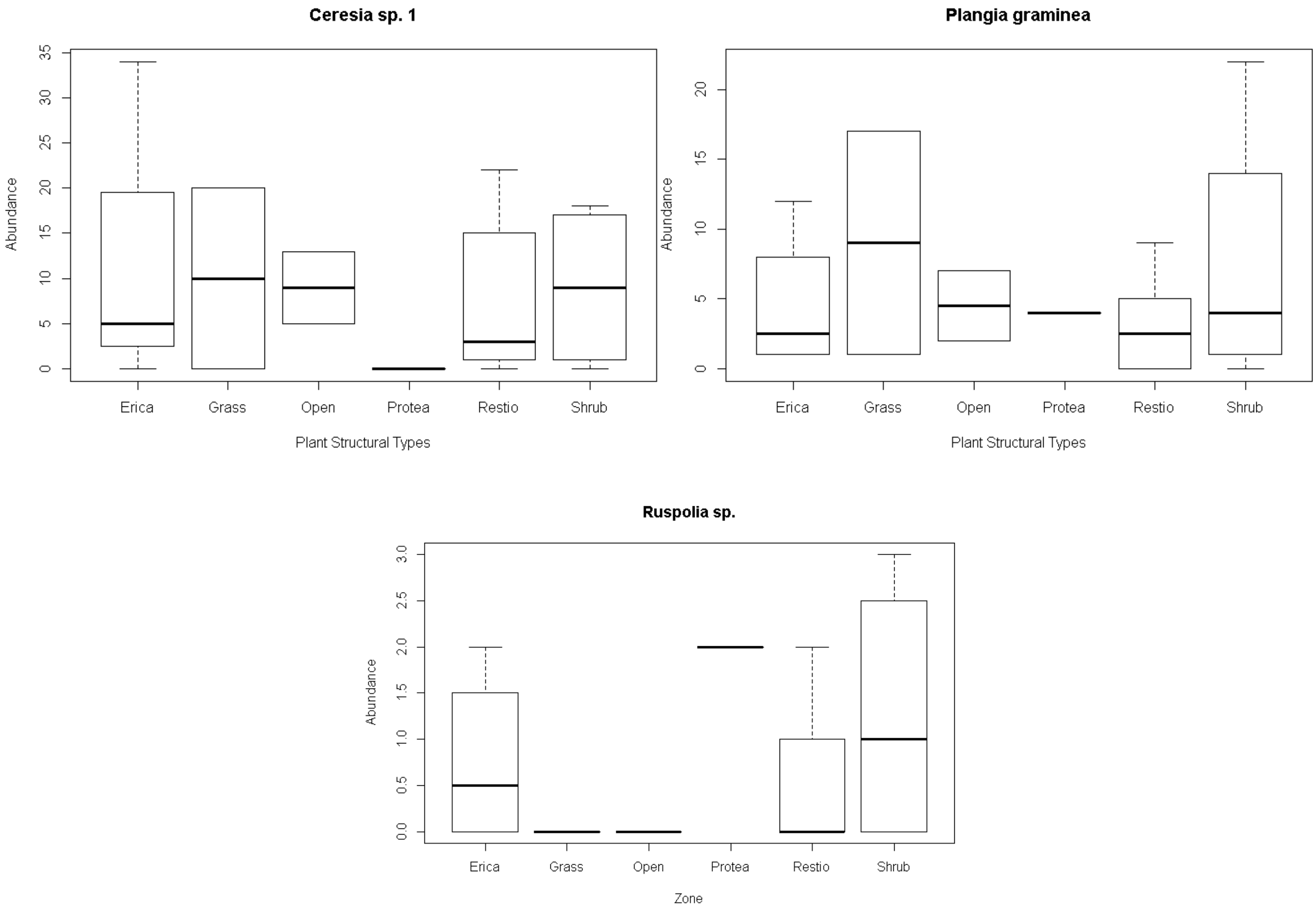


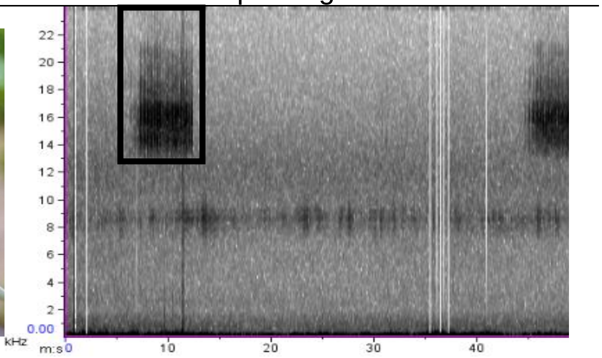
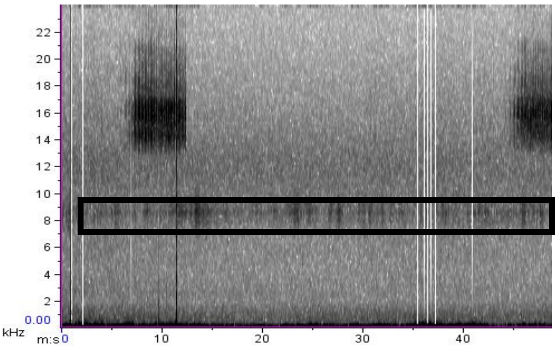
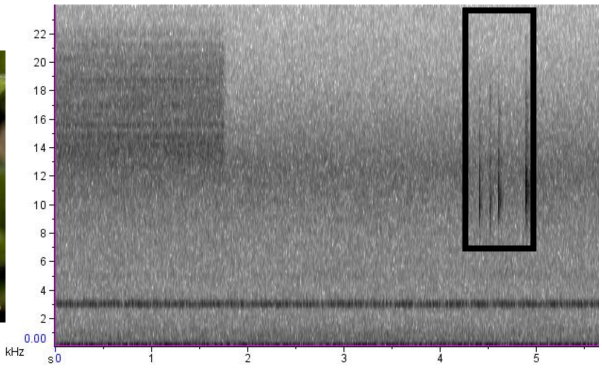
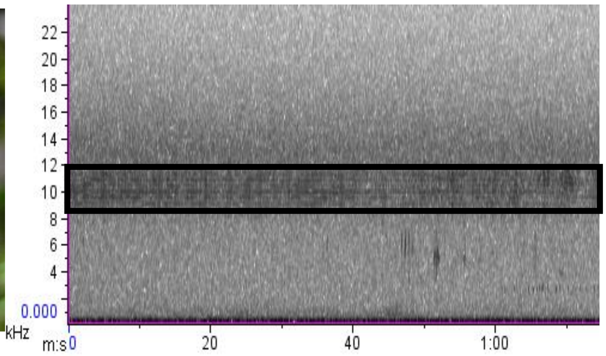
Figure 3.9: Mean abundances of the three most common katydid species across the dominant plant groups at each site

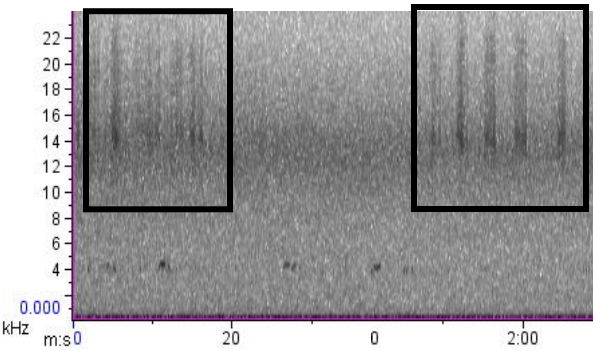
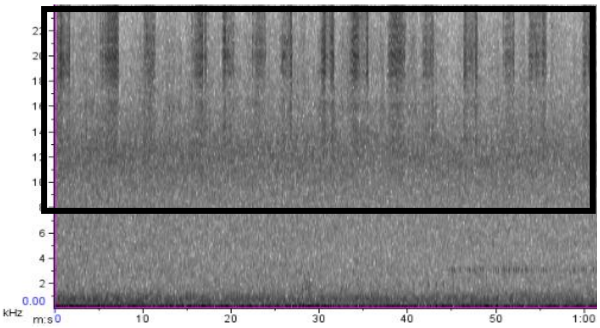
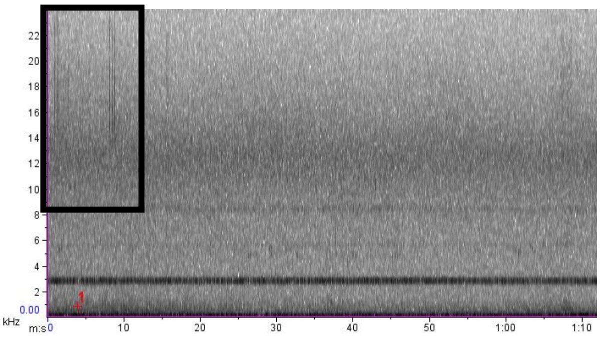
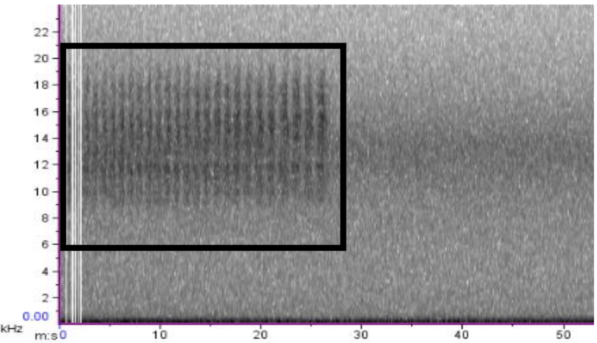


Appendix 3.1: Acoustic Activity estimates for each species across the four field work sessions and three zones of the Kogelberg Biosphere Reserve

	November 2015			**January 2016**			**February 2016**			**April 2016**		
	C	**B**	**T**	**C**	**B**	**T**	**C**	**B**	**T**	**C**	**B**	**T**
Ceresia sp. 1	113	18	77	206	120	98	135	41	136	14	75	
Ruspolia sp.			1	16	5	5	1				2	
Plangia graminea				50	97	195	115	116	190		106	2
Clonia melanoptera					4		2					
Ceresia sp. 2	63						8					
Tettigoniidae sp. 1						125			162			138
Tettigoniidae sp. 2								19				
Hetrodus pupus												2

Appendix 3.2: Photographs and spectrogram of all eight recorded species

Species	Photograph	Spectrogram
Ceresia sp. 1		
Ruspolia sp.	No photograph available	
P. graminea		
C. melanoptera		

Species	Photograph	Spectrogram
Ceresia sp. 2		
Tettigoniidae sp. 1	No photograph available	
Tettigoniidae sp. 2	No photograph available	
H. pupus		

Chapter 4

Katydids (Orthoptera: Tettigoniidae) of the Cape Floristic Region are less specialized than was expected

Abstract

Species are being lost at an unprecedented rate. South Africa has set a precedent with regards to invertebrate based assessment methods and currently employs two such methods to assess riparian health: the South African Scoring System (SASS) and the Dragonfly Biotic Index (DBI). As it is possible to assess the South African katydids in a similar way, katydids may be potential indicators of terrestrial habitat quality. As the Kogelberg Biosphere Reserve (KBR) is situated in one of three biodiversity hotspots in South Africa, and is classified into three zones of relative habitat disturbance, it is ideally suited for examining the potential of a katydid-based method for the assessment of habitat quality. Similarly to SASS and the DBI, the Katydid Biotic Index (KBI) takes into account the species composition of katydids at a site. This study tested whether the inclusion of abundance data improves the sensitivity of the KBI. Between November 2015 and April 2016, four field work sessions were conducted in which recordings, spot counts and active searches were used to determine the numbers and identities of the katydid species across 21 sites in the KBR. By assigning a species-specific score to each species, it was possible to calculate the KBI scores of the zones and sites, with and without the inclusion of species abundance information. After removing an outlier site, the inclusion of abundance information resulted in a more sensitive KBI. However, KBI was less sensitive than originally expected as it was only able to determine differences between the zones of the KBR and did not correlate well with habitat quality. The low diversity of katydid species observed in the CFR may hinder the applicability of the KBI assessment method. More research is necessary to determine whether the KBI would accurately assess habitat quality in environments more traditionally suited to katydids, such as forest patches in South Africa.

Introduction

Species are being lost a rate estimated to be between 100 and 1000 times faster than that of the natural extinction rate (Singh 2002; Rockström et al. 2009). This rapid and unprecedented loss of biodiversity is leading to the reduction of global ecosystem resilience (Chapin et al. 2000; Rockström et al. 2009). Great emphasis is placed on monitoring biodiversity and selecting sites for conservation by determining the species richness as well as endemism at the sites of interest (Myers et al. 2000; Mittermeier et al. 2011). The global biodiversity hotspots, aid in the selection of priority sites for conservation, and are based solely on the levels of threat and endemism of the plant taxa (Myers et al. 2000), greater focus on invertebrates is required for true biodiversity assessments to be made.

South Africa has set a precedent. Currently, there are two widely used rapid biodiversity assessment methods employed in the country. Both methods utilise invertebrates, the first is the South African Scoring System (SASS), which relies on the examination of benthic larvae of macro invertebrates to indicate the quality of the freshwater habitats in which they are found (Dickens & Graham 2002). The second assessment method is the Dragonfly Biotic Index (DBI) which relies on the adult dragonfly assemblages of freshwater and riparian systems to determine the quality of both environments (Samways & Simaika 2016). Both of these methods are used to determine the quality of riparian and freshwater habitats, yet there is only one method employed for terrestrial assessments in South Africa, the Grasshopper Conservation Index (Matenaar et al. 2015).

As set out in Bazelet et al. (2016), katydids (Orthoptera: Tettigoniidae) are a suitable taxon to contribute to a biodiversity assessment. In the Katydid Biotic Index (KBI) each katydid species is scored in accordance to its threat status, level of endemism, trophic level and degree of mobility. Therefore, katydid species that are highly threatened, endemic and have highly specialized habitat requirements will obtain higher scores than those species that are not threatened, are widespread and have generalist habitat requirements (Bazelet et al. 2016). The KBI scoring system therefore relies a great deal upon species-specific information, and as the katydids of South Africa are relatively well-understood and documented, this is possible to do (Naskrecki 2008). This method fills a vital need in the rapid assessment methods, as it should enable the quality of terrestrial environments to be determined. Although currently in its development phase, the method is proving promising as it relies on species-specific information therefore allowing species composition and species traits to be taken into account, making it a more sensitive indicator of changes in habitat quality than species richness alone (Bazelet et al. 2016).

Although the katydids in South Africa are vastly under-sampled (Naskrecki 2008), acoustic sampling allows for both quick and cost-effective sampling of these highly cryptic species within a variety of habitat types (Riede 1998; Diwakar et al. 2007; Sueur et al. 2008) in South Africa (Grant 2014; Grant & Samways 2016). By combining the KBI and acoustic sampling of katydids it should be possible to develop a rapid assessment technique that can aid in the assessment of habitat quality in the country. The KBI system shows promise in the Cape Floristic Region (CFR) when only museum records are employed as the sampling effort (see Chapter 2). Even within these under-sampled ecosystems, the KBI method was able to select and highlight those ecosystems that are threatened and experience higher than normal threats. The mean KBI scores of the threatened ecosystems tend towards having lower KBI scores than those of the least concerned ecosystems as all the necessary habitat requirements are met by these more pristine ecosystems. Therefore they are able to support more species with specific habitat requirements.

Within the CFR, the Kogelberg Biosphere Reserve (KBR) is situated in the most speciose and diverse area with regards to the plant and dragonfly species assemblages (Boucher 1978; Grant & Samways 2007, 2011). Little is known about the diversity patterns beyond those of the plants and dragonflies, and so this diverse yet understudied area presents itself as an appropriate area in which to study a relatively under-sampled invertebrate taxon. The characteristic zones of a biosphere reserve are core, buffer and transition zones (UNESCO 2016), which allow for a gradient of habitat transformation and disturbance to be studied. From previous work on dragonflies, it is known that the core supports highly endemic and sensitive species while these same species are absent from the transition and buffer zones which can be a few meters away (Grant & Samways 2007), such is the contrast between the zones. When the DBI is applied to the sites, sites within the core are selected as having many highly threatened, sensitive and range-restricted species (Grant & Samways 2011).

When calculating the DBI scores, only species richness and species composition are considered. The scores of the resident species are summed irrespective of their individual abundances (Samways & Simaika 2016). Katydid response to habitat quality is greater when abundance, and not just species richness, is taken into account (see Chapter 3). Therefore, this study aims to determine whether the inclusion of abundance information will help improve the sensitivity of the KBI scoring system by examining its correlation to measured habitat quality or to the zones of the KBR. This study therefore proposes a terrestrial biodiversity assessment method utilizing the acoustic monitoring of katydid species assemblages in order to determine the habitat quality of a site.

Methods

Study site

Covering 103 629 ha of marine and terrestrial habitats, the KBR was chosen as the study area as the reserve covers a wide range of terrestrial habitat types and land-use practices. Within the buffer and transition zones, natural vegetation is often surrounded by a matrix of agricultural land and other land uses, while the core remains largely protected as a formalised reserve with few human activities permitted within this zone. As the KBR is situated in the Fynbos Biome, it is characterised by hot dry summers and cool wet winters. The annual average rainfall for the area varies between 874 and 1 050 mm, while the average minimum temperature ranges between 4.2°C and 7.1°C, while the average maximum temperatures range between 26.5°C and 29.4°C (Boucher 1978).

Katydid sampling

Sampling was conducted from November 2015 to April 2016, spread over four sampling sessions, separated by roughly six weeks between sessions. This ensured that the entire succession of katydid assemblages of the area would be sampled as the different katydid species are hypothesized to emerge at different times. Therefore, November 2015, January 2016, February 2016 and April 2016 were sampled each for approximately two weeks. Sampling was only conducted in natural fynbos vegetation. Five sites were selected in the core zone, eight in the buffer zone, and eight in the transition zone (n = 21 sites in total). As the distance sound travels varies with amplitude and frequency (Marten & Marler 1977), it was decided that >300m would be sufficient to separate the sites from one another as the site distribution often tended to be clumped with multiple sites situated in the same region of the reserve. This clumped distribution of sites was as a result of the safety and logistical constraints that were considered when selecting sites as field work was conducted both during the day and at night.

At each site, SM2+ song meters (Wildlife Acoustics, USA) fitted with omnidirectional microphones were placed on a rock or raised landscape feature to reduce the attenuation of katydid songs. Song meters were moved between sites every two days. The soundscape was recorded at 48 kHz and the microphone set to a gain of 6, enabling faint insect calls to be recorded. Each site was sampled over two consecutive nights every field work session. The song meters were programmed to record for 3 min every 30 min between 20h00 and 23h00. The short time window was chosen to prevent severe weather changes, such as wind and rain, from disrupting the chorus. Since it has been shown that the katydid chorus does not vary temporally through the night in the area (Grant 2014), the timing of recordings did not affect the quantity or identity of the species heard calling. A total of 42 min of recordings were captured for each site for each session, resulting in a total of 58.8 hrs of recordings, which proved to be an adequate sampling effort as the calculated species accumulation curved reached an asymptote for the six species.

A spot count, coupled with active searching, was also conducted at night at each site in all four sessions. For ten minutes, a trained listener would sit and note down the approximate location, identity and number of calls heard of each katydid species. At the same time, a second observer would walk through the site tracking calling male katydids and actively searching and collecting male and female individuals. After completing the spot count, the listener would then join the second observer with the active searching. This was conducted over 30 min, resulting in a measurable sampling period of 1 hr: 10 min for the spot count and 50 min for the combined active searches. At the same time, a Pettersson D200 Ultrasound Detector (Pettersson Elektronik, Sweden) was used to locate the species calling in the ultrasonic range that could not be detected with the human ear or by the songmeter.

Habitat assessment

All habitats were assessed in November 2015. Assessments were conducted semi-quantitatively by completing a survey of five individual criteria: 1) Presence of typical fynbos components; 2) Sources of anthropogenic disturbance; 3) Presence of alien invasive plant species; 4) Diversity of plant species and 5) An assessment of habitat quality. Each criterion consisted of four levels, scored between 0 – 3, and at each site it was determined which level best suited the focal habitat. The individual scores were then added together and divided by 15 to obtain a Habitat Quality Index (HQI) between 0 - 1. Sites of better quality obtained final values close to 1 while poorer quality sites obtained values close to 0.

Signal analysis

All recordings were processed using Raven Pro 1.5 (Cornell Laboratories, USA). Individual spectrograms of each recording were produced, allowing for both visual and auditory identification of the various acoustic species. All katydid calls were individually identified, measured and counted. Identification of the various species was possible as voucher specimens and calls were collected in the field, enabling accurate identification of the recorded calls.

Data analyses

As it is not possible to determine the abundances of the individual species in the recordings, a method was required that enables an estimate of abundance. Miller (2001) suggested a method to determine the number of bat passes, the Acoustic Activity Index (AI). As the method relies on the presence of species in individual time windows, it was possible to adapt the method to the presence of katydids in each recording, with each recording being treated as a time window. The calculated AI values of each katydid species are thought to be a realistic estimate of the abundances of species in the field as they correlated with the various species abundances determined during the spot counts (see Chapter 3). It was therefore decided that the calculated AI values were to be used as the baseline dataset, as it was more reflective of number of individuals than the total number of calls. If total number of calls were to be used, pseudoreplication would be introduced into the analyses. The counts from the active searching and the spot counts were only added in when a species at a site showed 0 in the AI values as the active searched picked up species that were not located on the recordings. By combining these two methods it allows for a more inclusive sample of species to be used.

All statistical analyses were conducted using R (R Development Core Team 2015b). Initially a rank abundance curve was plotted to determine the most dominant and abundant species. This was followed by analyses run on two data sets, the initial one contained observations from all 21 sites, while the second data set contained data from 20 sites as one site was thought to be an outlier as a species assessed as sensitive and with specific habitat requirements was found at an unsuitable site. For both datasets, the observations across the four sessions were pooled together and the species identified and assigned the relevant KBI scores (Appendix 4.1).

As the aim is to determine whether the inclusion of species abundance improves the accuracy of the KBI, the KBI scores of each site were determined using two methods. The first method, called the KBI method, followed the DBI calculation methods and therefore the final KBI score for each site was determined by simply adding together the KBI scores of each species in accordance with their presence at the site, irrespective of the abundances of the species, and then dividing by the species richness of each site. The second method, called the KBI + AI method, added an extra step. After the scores of each species had been assigned, each species score was then multiplied by the abundance of each species present at the site. These multiplied values were then added together and divided by the total abundance of species at the site instead of species richness of the site. Both the KBI and the KBI + AI methods returned values between 0 - 9. Higher scores therefore indicated sites that had more specific habitat requirements, threatened or range restricted species for the KBI method and individuals for the KBI + AI method. Lower scores thus indicated sites that had less specific requirements, threatened or range restricted species or individuals depending on the method used.

Since KBI data were not normally distributed (Shapiro-Wilk’s $W = 0.86$, $p > 0.05$), Kruskal-Wallis ANOVAs were run for the KBI method data sets, using both the zones that the sites were situated in (core, buffer or transition) as a grouping factor as well as categorical HQI groupings based on natural breaks in the HQI data (0 – 0.5, 0.5 – 0.75 and 0.75 – 1) as the second grouping factor. As the KBI + AI method data were normally distributed (Shapiro-Wilk’s $W = 0.98$, $p < 0.05$), ANOVAs were run using the same two grouping factors. The number of total observations of all katydid species at each site was determined, and were also compared across the biosphere zones and HQI groups using a Kruskal-Wallis ANOVA, since the observations were non-normal (Shapiro-Wilk’s $W = 0.91$, $p > 0.05$). The same analyses were run after removing one site that proved to be an outlier as it is situated in the transition zone, had only three katydid observations and had the lowest HQI score of 0.13. Yet it had the highest KBI score for both the KBI and KBI + AI methods (KBI = 5 and KBI + AI = 4.67) as one

Brinckiella wilsoni individual was found at the site. It was therefore decided to exclude De Rust 4.

After removing the outlier site, the KBI and KBI + AI methods as well as the number of observations maintained the same distribution patterns: the KBI method was nonparametric (W = 0.85, $p < 0.05$), the KBI + AI method was parametric (W = 0.96, $p > 0.05$) and the number of observations was nonparametric (W = 0.91, $p < 0.05$).

Significant differences were only found between the zones in the abundance method once the outlier site was removed, and so the ANOVA was followed by a Tukey post hoc test. The correlation between the numbers of observations of katydid species and the KBI and KBI + AI scores for each site was run before and after the removal of the outlier site.

Finally the correlations between the HQI of the sites and the KBI and KBI + AI values were tested after the outlier site was removed. Spearman's correlation was used for the KBI scores as it is non-parametric while linear regression was used for the KBI + AI scores as they were parametric.

Mapping

To illustrate the patterns of species distributions in the KBR, three maps were produced using QGIS (Quantum GIS Development Team 2015b). The first map illustrated the various katydid species assemblages at each site by calculating what proportion of the assemblage each species comprised. The overall assemblage composition was represented by a pie chart and the diameter of each chart was scaled in accordance to the species richness of each site. The second and third maps illustrated the KBI and KBI + AI values of each site. The KBI and KBI +AI values were placed into continuous classes based on Jenks classification method (Brewer & Pickle 2002). These categories were then both colour coded and sized appropriately. For the KBI and KBI + AI maps the outlier site was removed.

Results

In total 10 876 calls were identified from the recordings. These consisted of katydids, crickets, grasshoppers, frogs and birds. Of these, 2 596 were identified as katydid calls. From these calls it was possible to positively identify six species of katydids: *Ceresia* sp. 1 was the most abundant caller with 1 174 calls being identified. *Plangia graminea* was the second most abundant caller, of which 871 calls were identified. *Ruspolia* sp. was the third most abundant at 30 calls. *Clonia melanoptera* was the fourth most abundant caller with only six identified calls. Two calls of *Hetrodes pupus* were identified at one site as well as a likely new species, *Ceresia* sp. 2, being located at one site and calling 73 times. A total of 67.8% of all the recordings collected were usable. The remainder of the recordings were excluded as there were either no calls present or the presence of wind and rain on the recording masked all other sounds.

All six of the species identified from the recordings were identified during both spot counts and active searching conducted in the field. In addition to these six species, two other species were identified during the active searching period: *Megalotheca montana* and *Brinckiella wilsoni,* yet they were not observed to be singing during the spot counts, nor were they picked up on the bat detector (Fig. 4.1 and Fig. 4.4a).

There were no significant differences between the median KBI scores for the zones or the HQI classes ($X_2 = 1.59$, p = 0.45 and $X_2 = 0.91$, p = 0.64). This was also the case with the KBI + AI method as there were no significant differences between the mean scores for the zones or HQI classes (F = 1.19, p = 0.33 and F = 0.29, p = 0.7, Fig. 4.2a and b). As with both scoring methods, there were no significant differences in the number of observations of katydids across the zones or HQI classes ($X_2 = 4.73$, p = 0.094 and $X_2 = 4.95$, p = 0.084, Fig. 4.2c), although they were close to having significant differences. There was no correlation between the number of observations and the KBI scores of each site when using either the KBI method ($r^2 = 0.12$) or the KBI+AI method ($r^2 = -0.039$).

After removal of the outlier site, the median KBI scores derived from the KBI method did not differ significantly across the zones or the HQI classes (X_2 = 2.86, p = 0.24 and X_2 = 2.86, p = 0.23, Fig. 4.3a). Neither did the mean KBI + AI scores differ across the HQI classes (F = 0.18, p = 0.87, Fig. 4.3b) yet the mean KBI + AI scores did differ significantly across the zones of the biosphere reserve with the core having a significantly higher KBI+AI score than the transition zone. The core and the buffer were not significantly different and the buffer and the transition zone were very nearly significantly different. There were no significant differences between the number of observations across the zones or HQI classes (X_2 = 4.42, p = 0.11 and X_2 = 4.74, p = 0.093, Fig. 4.3c). Once again there was no correlation between the number of observations at a site and the KBI score of the site for both the KBI and KBI + AI method (r^2 = 0.061, r^2 = -0.048). There were no correlations between the KBI or the KBI + AI methods and the HQI of the sites (Spearman's rho = 0.31, p = 0.15 and r^2= 0.026, p = 0.48, Fig. 4.4).

Figure 4.5 illustrates the katydid assemblages (Fig. 4.5a) across the sites as well as the KBI (Fig. 4.5b) and KBI + AI (Fig. 4.5c) scores across all sites. There were no differences in the distribution and spread of species across the KBR. *Ceresia* sp. 1, *Plangia graminea* and *Ruspolia* sp. dominate most of the assemblages across the reserve.

Discussion

The KBI species assessment method is a powerful method with which to assess the individual katydid species of South Africa, resulting in the individual species being directly comparable. A biodiversity assessment technique that is based on individually assessed species will by virtue of this fact be more sensitive to changes in the habitat required by the individual species (Bieringer & Zulka 2003; Rodrigues et al. 2006). Although highly promising in its ability to assess the individual katydid species, the KBI assessment method requires further work to determine whether or not it is a feasible terrestrial biodiversity assessment method. In line with this, it was determined that the inclusion of abundance information into the calculations did improve the sensitivity of the method and allow for the KBI assessments to differentiate between the zones of the KBR yet not between the habitat quality. The inclusion of abundance information allows for the higher scoring and rarer species to have a greater impact on the overall KBI score of a site.

Traditional biodiversity measures focus mainly on the species richness of a site (Myers et al. 2000; Mittermeier et al. 2004; Clausnitzer et al. 2009), but this cannot be the case with katydids. As previously shown in chapter 2, katydids respond towards habitat quality but this response is only noticeable when the entire assemblage is considered, including both species richness and abundance. As shown in figure 4a, the species richness of the sites does not change, and as a result, the KBI scoring system is ineffective when only species richness is taken into account. Therefore, the accuracy and sensitivity of the KBI assessments are improved when both the species richness and the abundances of the individual species are taken into account.

Even after taking the abundances of the different species into account and using them in the calculations, differences were only observed across the zones of the KBR and not the habitat quality ranges which can be thought of as a more accurate measure of habitat quality rather than the arbitrary boundaries of the KBR (UNESCO 2016). The lack of correlations between habitat qualities of a site and the KBI or KBI + AI scores shows that katydid species are not as sensitive as was previously thought.

Perhaps katydids in the CFR are more sensitive towards land use change instead of habitat quality. If katydids were more sensitive towards changing habitat quality it would be expected that there would be discernable differences between the assemblages of the core, buffer and transition zones, as well as the overall KBI and KBI + AI scores of these zones. Yet the only difference was found when the mean KBI + AI score of the transition zone was compared to those of the core and buffer zones. This suggests that the katydids only respond to coarse scale land use changes that occur in the transition zones, as the impact of land use change is minimized in the buffer and negligible in the core zones (UNESCO 2016). The total number of observations of katydids across the sites can also not be used as indicator on its own as there are no significant differences between either the number of observations at sites in the three zones or between sites across the HQI ranges.

Excluding the specialist species *Brinckiella wilsoni,* only two other katydid species were identified at the outlier site, De Rust 4: *Plangia graminea* and *Ceresia* sp. 1. Both of these species are widespread and have no specific habitat requirements (Rentz 1988; Hemp et al. 2015), and have relatively low KBI scores (see Appendix 4.1), *P. graminea* obtains a score of 3 and *Ceresia* sp. 1 a score of 4**.** In fact, *P. graminea* is a vineyard pest in the Western Cape and thrives in disturbed environments (Ferreira & Venter 1996; Allsopp 2012). *Ceresia* p. 1 here was heard singing in both urban gardens and pine plantations. These observations reinforce the KBI species assessments, both these two species are habitat generalists and occur in a wide range of disturbed habitats. It would therefore be expected that this particular site should obtain an average KBI score ranging between 3 and 4. As these were the only other two species observed at the site.

It is essential to understand the life histories of any taxon to be used as an assessment tool as this will enable the end user to prevent inconsistencies from obscuring the more essential patterns. From this study, the inclusion of a single site, De Rust 4, obscured the more realistic and biologically correct patterns from emerging. Before being able to establish a biodiversity assessment utilizing a particular taxon, it must first be established whether the end user of the assessment method understands the taxon well, as it enables the user to distinguish patterns occurring as well as to identify discrepancies among data points.

It is likely that less is known about *Brinckiella wilsoni* than was originally thought. Previously thought to be a winter or early spring species (Naskrecki & Bazelet 2009), perhaps the species should be reassessed, and assigned a lower KBI score, as all sampling took place during the summer months and the species may in fact be present for a period longer than originally thought. Another reason it could have been found there is that it is a single host herbivore species (Naskrecki 2008), with perhaps its host plant being present at the site. Although *B. wilsoni* is a flightless species (Naskrecki & Bazelet 2009), many katydid species are able to commute between sites in search of more favorable conditions and resources (Shelly & Bailey 1995), it is therefore likely that this individual was commuting between sites when located here. It is unlikely, owing to the degraded nature of the site and the lack of structural and plant species diversity, that the specific host plant would be present at the site.

The site was dominated by a single species of the pioneering Renosterbos which thrives in disturbed lands such as old pastures and road sides (Manning 2007). This site was severely degraded and was surrounded on three sides by invasive *Eucalyptus* species and the fourth side fronted on a large farm reservoir. It is not a site conducive to the maintenance of diverse faunal assemblages (Higgins et al. 1999; Kemper et al. 1999; Thuiller et al. 2006; Van Wilgen 2009). This site is within the severely altered transition zone and, from the habitat assessment conducted, was identified as the most degraded site, obtaining a HQI score of only 0.13. It was therefore not expected that this site would have the highest KBI score of 5 and KBI + AI score of 4.67. Only when the outlier site is removed do the differences between the scores of the different sites in the different zones become evident, with the higher scores appearing more within the buffer and core zones.

Although much is known about the katydids in South Africa, they are relatively under-sampled country-wide. Currently there are about 1 076 museum specimens from known sampled localities (Naskrecki 2008). It is therefore necessary for wide-scale improvement of sampling effort in South Africa. Furthermore, it is unknown what impacts anthropogenic land use change and climate change will have on the South African katydids. Therefore, this study fills a gap in assessments and helps determine which changing aspects of a landscape may be playing a role in katydid distribution patterns.

Through the use of acoustic sampling it is possible to quickly and accurately determine the total number of species at a site (Nityananda & Balakrishnan 2006; Grant & Samways 2011). Using both passive sampling techniques, as well as active searching, enables the local assemblage to be determined with relative ease. Acoustic sampling is perhaps even more favorable in the CFR, when the target taxa produce recognizable calls, as often the vegetation is dense and sclerophyllous, forming almost impenetrable stands of vegetation in which standard sampling techniques could not be used (Pryke & Samways 2008). This therefore enables the complete assemblage in an area to be sampled in a quick and timely manner. For this reason it is a relatively straightforward task to determine which species are found in the area, enabling accurate habitat assessments.

Conclusions

The KBI scoring system is an applicable method with which to assess the individual katydid species and the inclusion of abundances into the calculations improves the sensitivity of the scoring system. Although the low katydid species diversity in the KBR was unexpected, it is likely that the species here are considerable generalists with one or two sensitive species that occur at such low abundances so as to not have an impact on the overall trends observed. The applicability of the method would have to be tested out in a habitat more conducive to katydids, such as remnant forest patches. Perhaps the KBI scoring system cannot be so widely applied as was originally thought.

Figure 4.1: Rank abundance plot of the eight species identified in the Kogelberg Biosphere Reserve

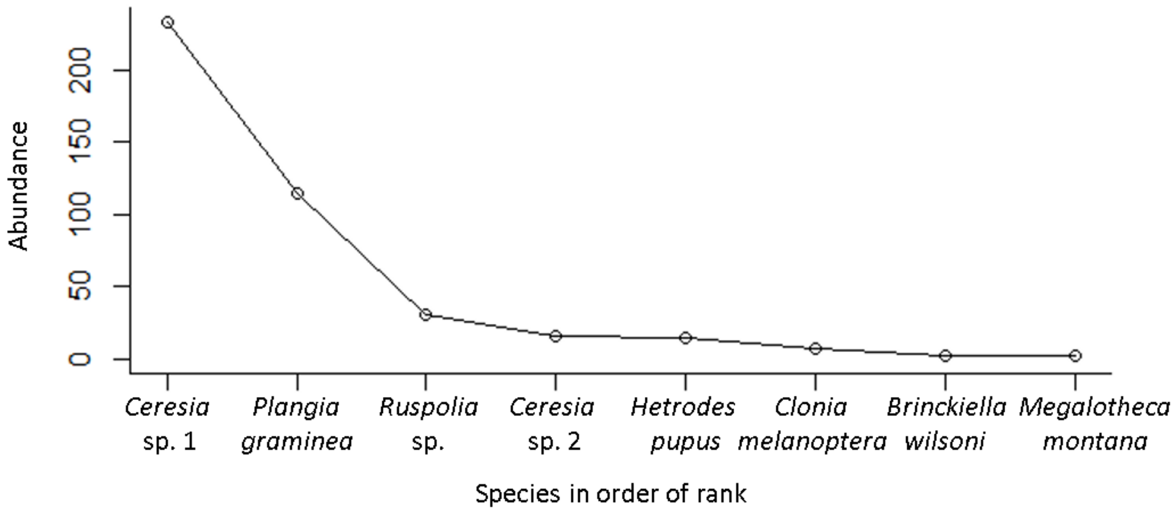


Figure 4.2: Comparison of the median Katydid Biotic Index (KBI) scores and mean Katydid Biotic Index plus Acoustic Activity (KBI + AI) scores and the median number of observations across the zones of the KBR and the HQI classes

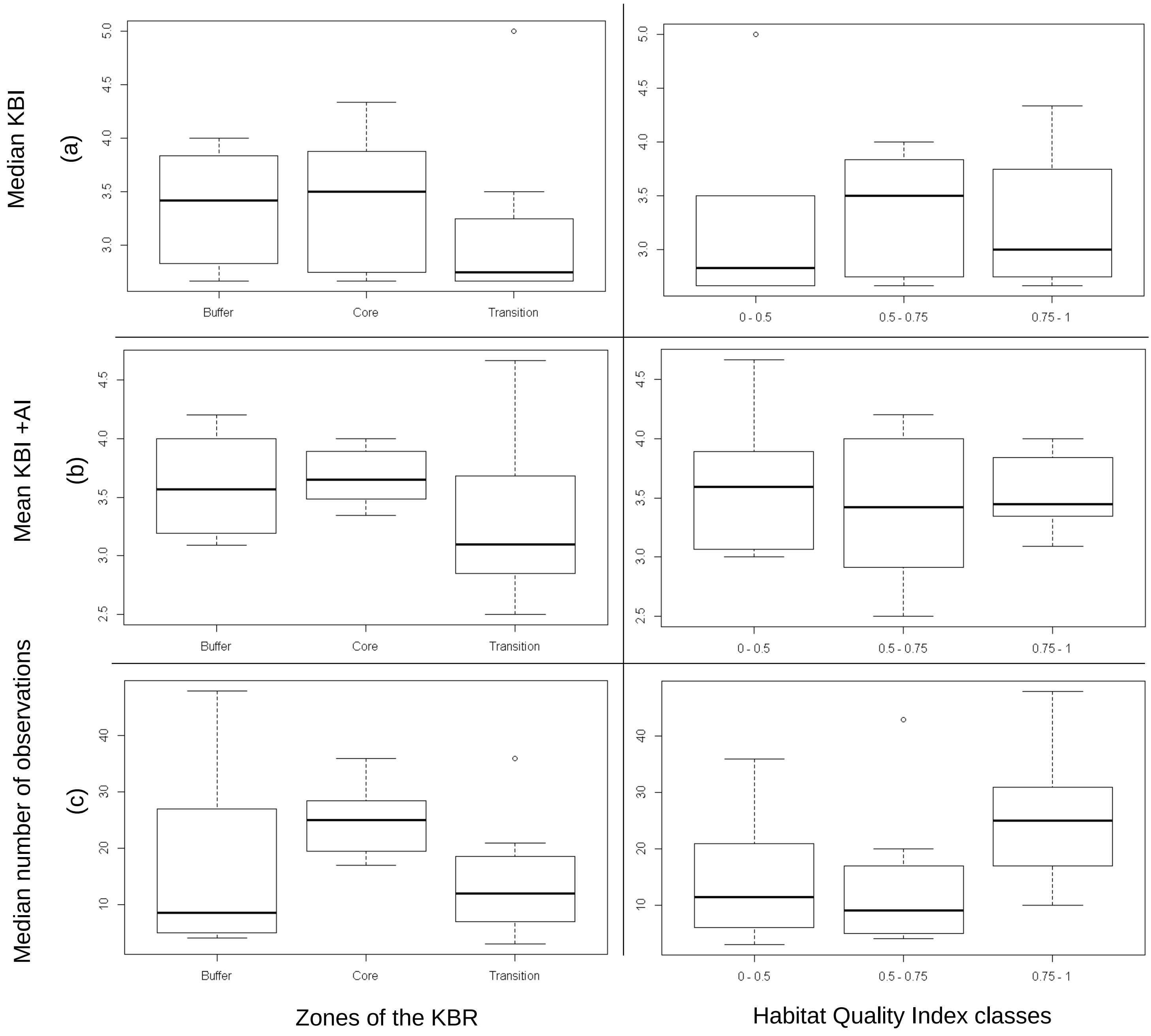


Figure 4.3: Comparison of the median KBI and the mean KBI + AI scores as well as the median number of observation across the zones of the KBR and the HQI classes following the removal of the outlier site. * p< 0.05; ** p< 0.01; ***p<0.001

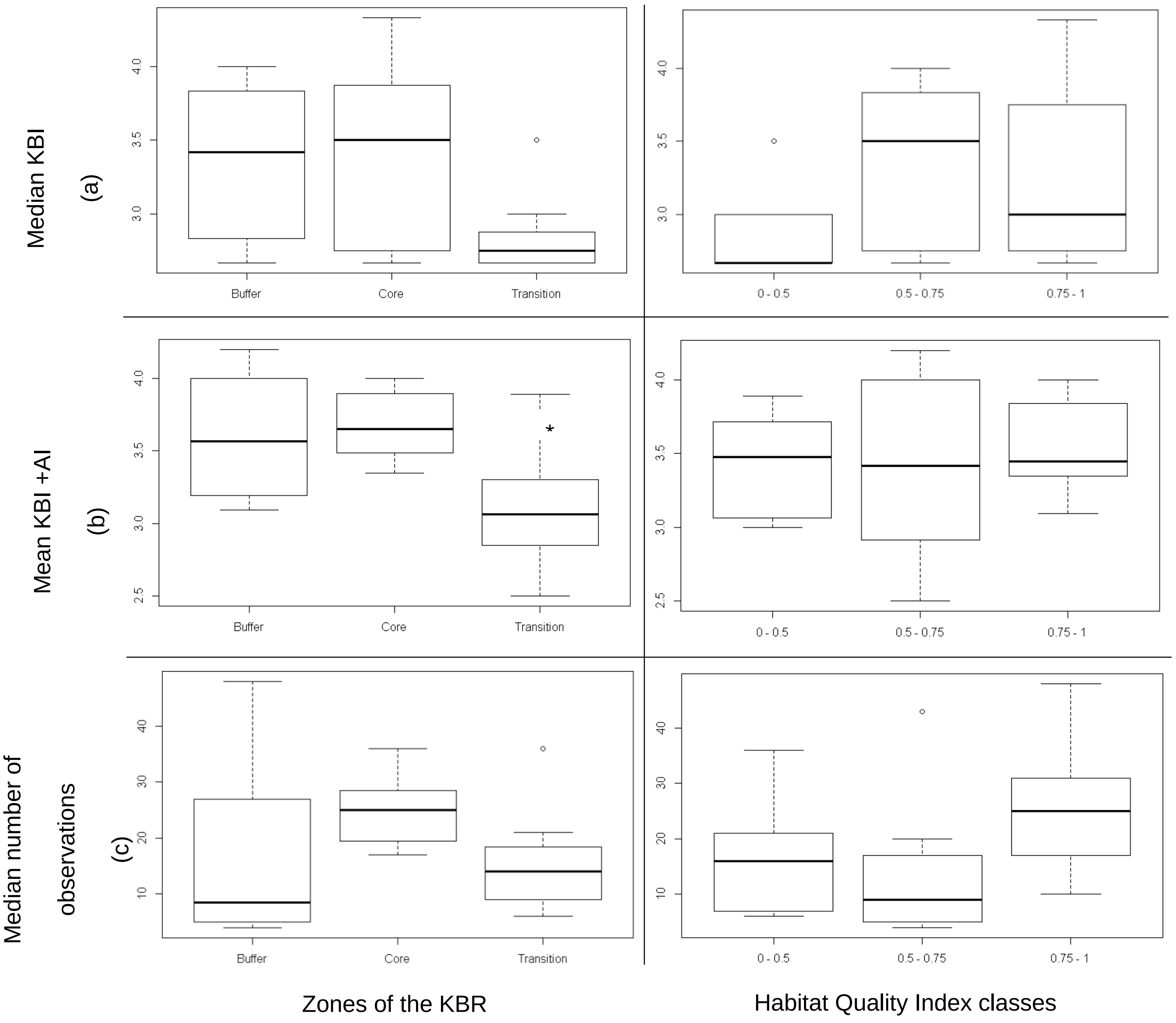
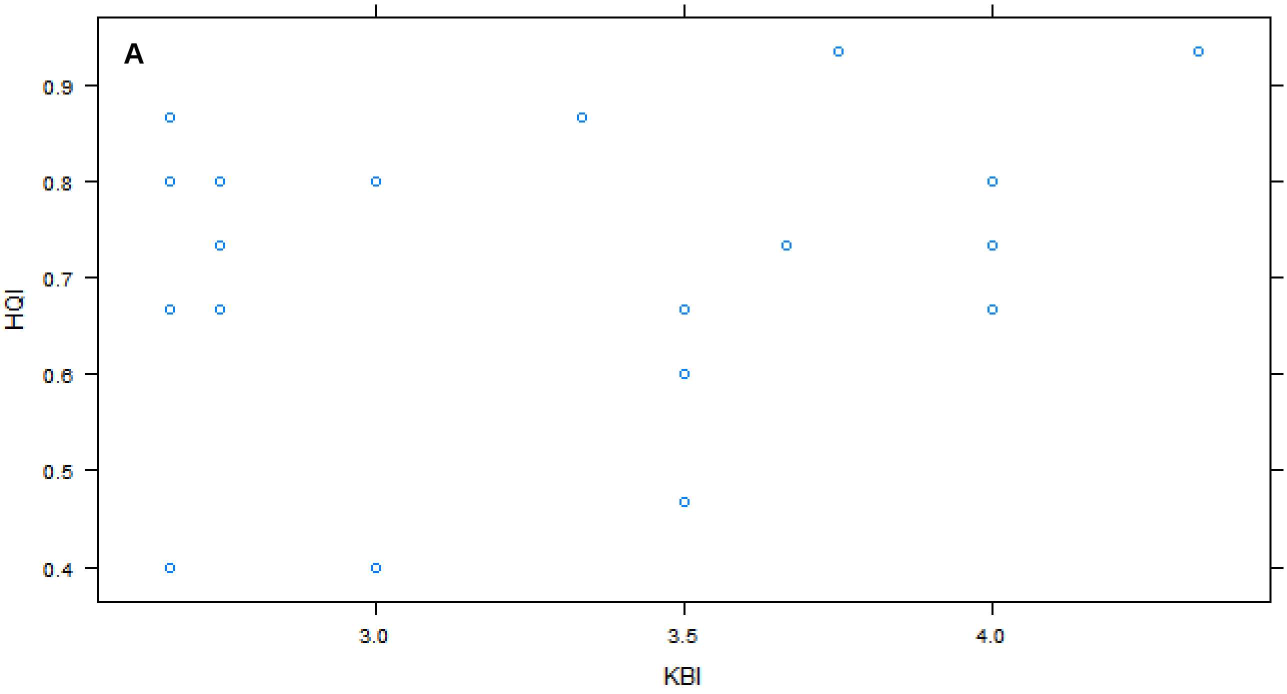


Figure 4.4: Scatter plot of a) the various KBI scores of the different sites and the sites HQI value and b) the KBI + AI scores of the various sites and the sites corresponding HQI value



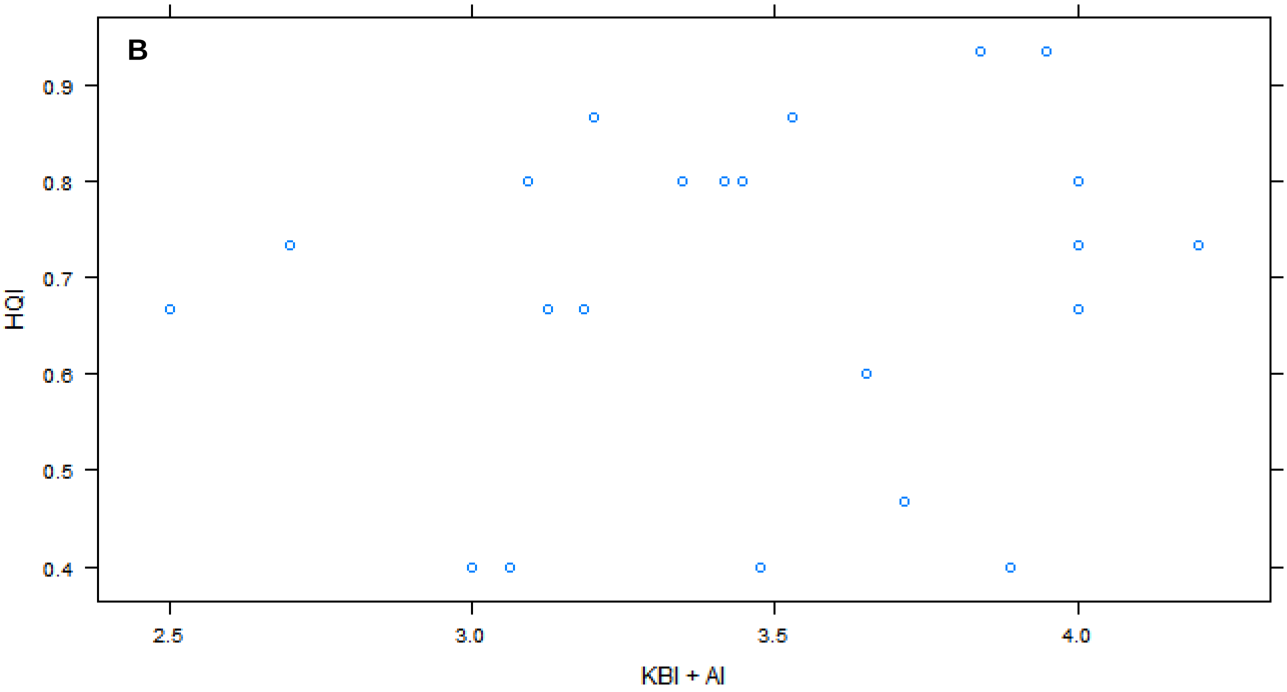
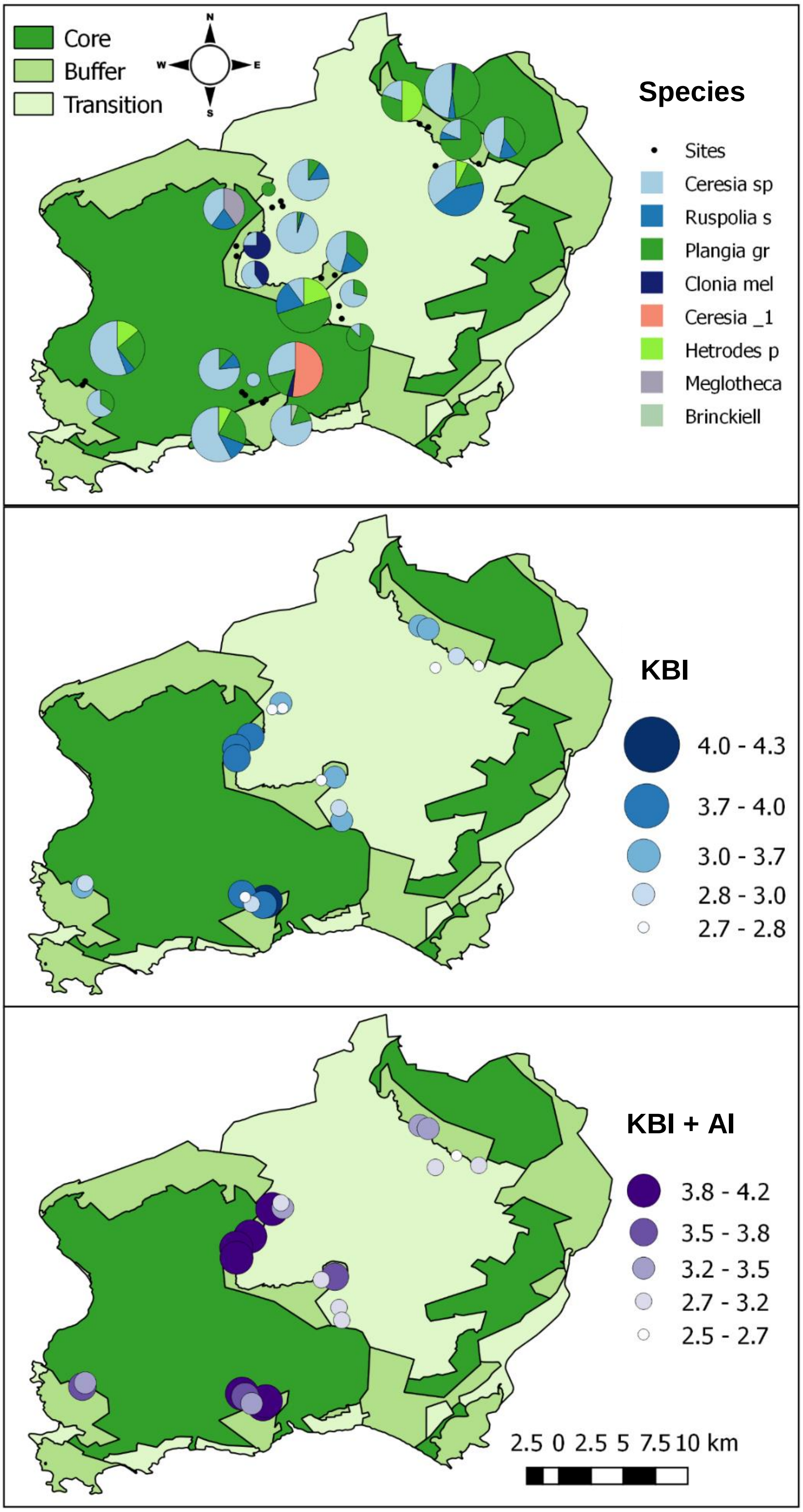


Figure 4.5: Maps showing a) the species assemblages of each sites, b) the distribution of the KBI scores across the KBR following the removal of De Rust 4 and c) the distribution of the KBI + AI score across the KBR following the removal of De Rust 4:



Appendix 4.1: Likely species identifications used in KBI calculations as well as the KBI scores and numbers identified in each zone of the Kogelberg Biosphere Reserve

			Number of observations		
Species Name	**KBI Score**	**Threat Status**	**Core**	**Buffer**	**Transition**
Ceresia sp. 1	4	LC	110	11	35
Ruspolia sp. 1	1	LC	7	11	13
Plangia compressa	3	LC	32	48	35
Clonia melanoptera	4	LC	1	6	
Ceresia sp.2	4	LC	16		
Hetrodes pupus	3	LC	7	5	3
Megalotheca montana	6	LC		2	
Brinckiella wilsoni	6	LC	1		1

Chapter 5

General conclusions

One of very few studies focusing on the acoustic monitoring of katydids in South Africa, and in particular within the Cape Floristic Region (CFR), in this study I aimed to determine whether or not monitoring katydids in the landscape would enable their use as biological indicators. Acoustic monitoring of the katydids within the fynbos vegetation is feasible as it allows non-invasive observations. As well as circumvents some challenges faced when attempting to sample invertebrates using traditional methods, such as sweep netting, in the dense and impenetrable fynbos vegetation.

The Katydid Biotic Index (KBI) shows promise at a coarse-scale, as it identifies ecosystems in need of conservation attention even when using a subset of museum records, and even though the CFR is under-represented by these records. Only intended to be a case study to highlight the areas within the CFR that required conservation action, this study also enabled the identification of problems within the KBI assessment method that required further attention. As a result of this, intensive sampling over a four month period was carried out in the KBR as it allowed for the katydid assemblage response to be monitored across a gradient of changing habitat quality.

Unlike the dragonflies of the KBR, it was found that the katydids of the KBR are neither complementary across the zones nor are they highly diverse across the entire reserve. Only eight species were acoustically identified in this unique and biodiverse region of the CFR. This suggests that the katydid species recorded here within the KBR are not a successful group in terms of species richness to the prevailing environmental conditions, such as the fire regime of the CFR. The katydids were also found to respond towards measured habitat quality but only when abundance was taken into account.

The abundances of the various katydid species differed across the zones of the KBR, yet not to the degree that was expected. The three most abundant species, *Ceresia* sp 1, *Plangia* graminea and *Ruspolia* sp., scored very low KBI values, and as they are spread across all three zones and habitat qualities, the presence of the less common and higher scoring species becomes important. Therefore, the inclusion of abundance information into the KBI calculations for each site enables the assessment method to take into account not only the common low scoring species but also the less common higher scoring species. It is these higher scoring species that are expected to be more sensitive towards the differences between the natural and the disturbed habitats, and therefore their presence and absence should be noted.

Due to the katydid assemblages differing across the summer months, it is essential to monitor the katydid assemblages across the entire period to gain a full understanding of the changes in species distribution. This will ensure that the adult males are sampled, as the species will not reflect on the recordings if they are only present as nymphs or as females only. If sampling were to only have covered a smaller time window, the entire assemblage would not have been sampled and any conclusions drawn would have been an inaccurate description of the processes at play in the KBR.

When considering the KBI assessment method and applying it to the katydid species sampled within the KBR, it becomes evident that the inclusion of abundance information into the calculations is necessary to improve the sensitivity of the method for application in the fynbos. Even with the inclusion of abundance information, it appears as if the katydid assemblages present were only sensitive towards coarse-scale habitat changes that occur between the transition zone and the core and buffer combined. There was no response when fine scale measurements, stemming from the habitat quality assessments, were taken into account. The low species diversity in the area is likely responsible for the low sensitivity of the katydid assemblage. If there were more species present within the KBR, the KBI assessment method would likely have been more sensitive to changing habitat types. There would be a chance for a greater variety of high scoring species being present across all habitat types, thereby allowing the KBI assessment method to infer the quality of a site with greater accuracy at finer scale.

Katydids in the CFR are perhaps more generalist than was originally thought, and likely to be poorly adjusted to the frequent fire regimes that dominate the fynbos biome. Other taxa, such as dragonflies and flighted grasshoppers are either able to escape fires by moving beyond their reach or are able to find fire refugia within the landscape. The *Ceresia* sp. 1 was a flightless species, as well as the most dominant species within the KBR. Therefore, in the advent of a fire, it is likely that individuals of this species would be unable to escape. As a result of this, this species cannot be expected to be present in a post fire environment, as well as taking a long time to recover post fire. Due to their being flightless, recovery of *Ceresia* sp. 1 in the environment will depend on the slow colonisation by species slowly dispersing back into the post fire environment.

Not as applicable in the fynbos as was originally hoped, the KBI method should not be written off entirely. The calls of the various species were learned with relative ease as the assemblage was relatively small when compared to katydid assemblages from other regions in the world, for example the tropical forests of India. The katydid species encountered in the Kogelberg Biosphere Reserve (KBR) were all morphologically distinct species. The calls belonging to the different species were also distinguishable based on their call structure when viewed as a spectrogram, as well as when listened to in the field or when played back in the recordings. These factors combined to ensure that the identification of the resident species was both a quick and a relatively straightforward process. By combining passive recordings, spot counts and active searches across the entire summer period, it was ensured that the entire assemblage was identified and assessed.

Currently, there are few rapid biodiversity assessment methods in South Africa that can be used to accurately determine the quality of terrestrial environments even though South Africa is home to three biodiversity hotspots. For this reason alone, there is a great need for a method that can be used to accurately assess these varied environments. Future work should focus on applying the KBI assessment method to more traditional katydid habitats such as forests. Even though katydids are likely not a suitable taxon for country-wide habitat assessments, they were still shown here to be relatively sensitive to habitat quality. However, the right environment in which to test the KBI assessment method is yet to be found.

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