Structural and Functional Attributes of Heuweltjies in the Fynbos and Succulent Karoo Biomes: The Interaction of Termites, Vegetation and Geochemistry

by

Nicola Vermonti

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Supervisor: Prof S. Jacobs (Department of Conservation Ecology and Entomology)

Co- Supervisors:

Prof J. Miller (Department of Earth Sciences)

Dr M. Francis (Department of Soil Science)

Academic Advisors:

Dr C. Clarke (Department of Soil Science)

Dr C. Crous (Department of Conservation Ecology and Entomology)

Department of Conservation Ecology and Entomology

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Declaration page

By submitting this thesis electronically, I declare that the entirety of the work contained therein is my own, original work, that I am the sole author thereof (save to the extent explicitly otherwise stated), that reproduction and publication thereof by Stellenbosch University will not infringe any third-party rights and that I have not previously in its entirety or in part submitted it for obtaining any qualification.

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Abstract

The structural and functional attributes of heuweltjies in the Fynbos and Succulent Karoo biomes were studied with the aim of investigating the interaction amongst vegetation, termites, and geochemistry on heuweltjies found across the West Coast of South Africa. This study takes a multi-disciplinary approach to determine possible sources of salinity in the biotic community, that can be considered inputs into heuweltjie mound soils. Heuweltjie vegetation communities were found to host different plant species that were structured differently and consisted of different plant functional types. These differences occurred between heuweltjies found in the Fynbos and Succulent Karoo, and among the centre, slope, and off areas of the heuweltjie mound. Anthropogenic disturbance such as grazing was also found to bring about changes in the structure of the vegetation communities. A subsample of the most common plant species growing in these communities were found to host variable concentrations of plant tissue ions, with sodium, magnesium, and potassium as the major ions found in higher concentrations, in the species growing on the mounds. These plants are suspected to have an important role in maintaining, but not causing the higher ion concentrations which are seen in central heuweltjie soils, as these plants could take up ions through their roots into their tissues and return them when abscission or senescence occurs. This creates a feedback loop with the soil environment. Where soils are saline, halophytic plants would be expected to form part of this cycle. Two external sources of salt were identified as viable contributors to the accumulation of salts that are associated with heuweltjie mounds. Aerosols deposited on the outer surfaces of the plants were identified to have a marine source of sodium, magnesium, and sulphate. Calcium deposits were enriched compared with seawater ion ratios. The foraging activities of *Microhodotermes viator* and foraging rodents were the second external source investigated to assess their impact on heuweltjie soils, using stable $\delta^{13}C$ and $\delta^{15}N$ isotope analysis. An evaluation was made of the plants that most likely were targeted by termites and rodents, using observations and $\delta^{13}C$. It appeared that termites collect live plant material nearby to their foraging holes, with the exception of any succulent plant species in the Fynbos, or stem and leaf succulents in the Succulent Karoo. This study shows that calcium oxalate crystals (whewellite $CaC_2O_4{\cdot}H_2O$) are present in plant tissues on centre mound soils as well as in the plant material collected by termites and rodents while weddellite ($Ca(C_2O_4){\cdot}2H_2O$) was present but only in the centre mound plants of the Fynbos. This suggests that the heuweltjies may be suitable environments for the oxalate-carbonate pathway to take place, in which decomposition of calcium oxalate in plant material leads to the precipitation of calcite in the surrounding soil and provides a possible explanation for the calcite enrichment common in heuweltjie centres. Plant material was also found to be a source of pentahydrite ($MgSO_4.5H_20$), halite (NaCl), sylvite (KCl) and sodium oxalate ($Na_2C_2O_4$). The impact that *M. viator* has on salt dynamics of heuweltjie centre mound soils was assessed in more detail. Ion input estimations due to *M. viator* foraging, were made using plant material input rates of 0.08 g per minute per foraging hole, on one heuweltjie. If foraging occurred for an hour every day, the ions with the highest estimated inputs per year were found to be calcium with 18 g (Fynbos) and 42 g (S. Karoo), and sodium 4 g (Fynbos) and 45 g (S. Karoo). Considering the estimated age of heuweltjies (4000 to 30 000 years old), total calcium inputs could range from 74 kg to 522 kg in the heuweltjies at the Fynbos site, and the Succulent Karoo estimates could be between 166 kg to 1 ton, for just one foraging hole. These estimates could be at five times more per heuweltjie when considering the number of possible

foraging holes, the termites use. Concentrations of calcium in the plant washes were found in levels four to twenty times higher than what Midgely et at. (2012) had previously been suggested would be needed to form the calcite layer, exclusively from sources of precipitation. Considering the contributions from the plant material along with the external deposits a calcrete layer of 0.25m wide could viably be formed in less than 3600 years. This study found, quantified, and presented evidence, that the foraging action of termites and deposition of marine aerosols, are both considerable inputs into the salt dynamics of heuweltjie centre mound soils.

Figure 1.1: Photo of Heuweltjieveld, with distinct heuweltjie in foreground with clear difference in vegetation and numerous heuweltjies in the background in the Succulent Karoo, Photo taken by Jannick Nieuwoudt on 15 March 2021 in The Kommagas Municipality, Northern Cape.

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Chapter 1: General Introduction

1.1 Introduction

Heuweltjies are distinct circular soil mounds that can be found occurring in a broad band across the Mediterranean and arid biomes of the western and southern parts of South Africa. The mounds are approximately 10-30 meters wide, 1-2 meters high and cover 14-25 % of the land surface (Cox et al., 1987; Lovegrove and Siegfried, 1989, 1986; Moore and Picker, 1991). They have been described as paleo-features with estimated ages ranging from 4000 yrs. (Moore and Picker, 1991), to as far back as 20 000-30 000 yrs. (Midgley et al., 2002; Potts et al., 2009). Individual heuweltjies are likely to have been actively forming over a considerable period of time. As well as occurring in south Africa, similar ecosystem features can be found across other continents, including North and South America (Cox, 1984; Cramer and Barger, 2014; Horwath Burnham and Johnson, 2012a, 2012b; Silva et al., 2010) and Australia (Noble, 1993). In southern Africa, these mounds can be found in a range of different land types, from pristine protected ecosystems to agricultural farms across both the Fynbos and Succulent Karoo biomes (Lovegrove and Siegfried, 1986). Even after decades of ripping and ploughing of the soils, in cultivated landscapes heuweltjies can still be observed, and still influence the productivity of crops grown on such surfaces (Bekker et al., 2016; Lovegrove and Siegfried, 1986). This suggests that the influence of heuweltjie soils will impact ecosystem functioning for years to come and have done for years past.

While there is no common consensus about how heuweltjies were formed, the harvester termite species *Microhodotermes viator* appears to play a key role in their current functioning (Kunz et al., 2012; Lovegrove and Siegfried, 1986, 1989; McAuliffe et al., 2019a; Milton and Dean, 1990; Moore and Picker, 1991). *Microhodotermes viator* are known to forage in groups on the surface for plant material, which then gets brought into their nests within the soil to be used as food. Once eaten, it is digested and deposited as frass, or simply left to decompose in the soil. This process is thought to be a key source of additional nutrients added to heuweltjie soils (McAuliffe et al., 2019a). This pathway may also bring non-plant salt into the heuweltjie, as wind-blown marine salts or fog are deposited on leaf surfaces of plants in these ecosystems. Similar processes may be facilitated by rodents that also live in the mound soils and have similar plant-based diets and foraging behaviour (Cox et al., 1987; Knight et al., 1989; Lovegrove and Siegfried, 1986; Turner, 2003). The actions of termites as well as rodents could contribute to creating and maintaining the distinctly different soil chemistry that can be found on the mound in comparison to off the mound, through bringing in additional salts to the heuweltjie mound soils. This can have a direct bearing on the types of vegetation communities that develop on the mounds in comparison to off the mounds and may further impact the way in which salts may be transferred into the heuweltjie.

A unique feature of centre mound heuweltjie soils In the more arid regions is the presence of a calcite layer, which can be found at the bottom of most heuweltjies, when no calcium exists in the underlying parent material of the area (Francis et al., 2007; Petersen, 2008). Formation of this calcite layer has broad implications that heuweltjies soils have the potential to be important long-term stores of carbon. Heuweltjie calcrete is likely to be a net sink of carbon which has sequestered and accumulated between 3 and 15 tonnes of atmospheric CO_2 per hectare of land (Midgley et al., 2012). Previous studies on the presence of calcite in heuweltjies have identified surrounding plant material as calcium-oxalate enriched and suggest that a process called the oxalate-carbonate pathway could exists in heuweltjies (Francis et al., 2013; Francis and Poch, 2019; McAuliffe et al., 2019a). How this soil process is understood to work is: oxalate is oxidised by a combination of oxalotrophic bacteria and fungi (Uren 2018). These studies assume that the termites are bringing this calcium-rich plant material into the heuweltjie through their foraging activities (Francis et al., 2013; Francis and Poch, 2019; McAuliffe et al., 2019b), but it has not been evaluated previously. It can be assumed that not only calcium oxalate is being brought into mounds through the actions of termites, but other ions and salts are possibly being concentrated into mound soils through this action too (Cornell, 2014).

Although termite activity is not the only influence on heuweltjie ecosystem dynamics, it does play a significant role. Studying the entire heuweltjie ecosystem at once, is too complex for this study. Therefore, this project will focus specifically on the interaction amongst vegetation communities, termites, and rodents and what impact this could have on heuweltjie soil salinity. Other factors, including soil nutrients and structure, disturbance by animals, rainfall, climate, fire, and biological competition, can also have an impact on heuweltjie vegetation communities, and will be kept in mind when reporting the findings, as many of these processes influence plant and water relations on heuweltjies. Different plant communities have variable capacities to hold salt in their tissues depending on the plant form, part, or just the specific species that make up the communities. Different species also have variable capacity to contribute salt into the centre mound soils depending on where they grow on the mound, or if they are preferred food for termites and rodent colonies, living in the mounds or not.

The broader implications of this study is to understand how the inter-connectedness of heuweltjie ecosystem structure and saline soils, might affect groundwater conditions. Groundwater is not addressed in this thesis however, it has been suggested that heuweltjies, could be a source of natural salinisation for groundwater across the west coast of South Africa (van Gend, 2018; van Gend et al., 2021; Vermooten, 2019), due to the spatial overlap of heuweltjies, and in areas affected by variably saline soils and groundwater. This is supported by evidence for accumulation and downward movement of salts in the mounds (Clarke et al., 2022). The reason for elevated nutrients, and salts in heuweltjies soils, is unknown. Enrichment of salts in the soils has been suggested to be fostered by biological activity in the form of vegetation, foraging insects, and animals that are associated with heuweltjies. The study areas of this thesis within the West Coast District and Namaqualand of South Africa were chosen because they are areas particularly affected by saline soils and groundwater.

Understanding how and why soils and groundwater become saline is a critical issue, since human communities in these arid areas often rely solely on groundwater resources as their primary water source (Adams et al., 2004; Mukheibir and Sparks, 2005). The salt levels in groundwater, across some areas of the west coast, have been found to be too high even for agricultural use (Mukheibir and Sparks, 2005) and this significantly restricts economic development. Ground water availability and quality consequently have a direct impact on livelihoods in these regions (Mukheibir and Sparks, 2005). While large parts of the west coast are affected by anthropogenic disturbance of natural vegetation, there are considerable tracts of pristine land set aside in national parks, because of the recognised biodiversity value of the natural vegetation. In these areas, groundwater salinisation is still prevalent, suggesting that it is linked to natural biological processes. Outside of these areas, anthropogenic processes affecting the surface biota may further exacerbate salinisation of the groundwater, which can lead to irreversible soil degradation problems (Gorji et al., 2017). In order to address economic and societal challenges in these areas, understanding the causal mechanisms of soil salinisation is a necessary first step to develop appropriate adaptation and management strategies to these saline conditions.

This study takes a multi-disciplinary approach to understanding the impact of termites on heuweltjie ecosystem functioning before evaluating the inter-connectedness of heuweltjie ecosystem structure and saline soils. The first step in this evaluation will be to identify vegetation trends across heuweltjies in the two regions of the study (Fynbos and Succulent Karoo) in order to create a baseline understanding of how plant communities differ on and off heuweltjies. Although differences in plant communities are expected to correlate with differences in soil composition on and off heuweltjie mounds (Booi, 2011; Kunz et al., 2012; Midgley and Musil, 1990; Schmiedel et al., 2016; van Gend et al., 2021), the degree of surface disturbance may also be an important factor. Since disturbance can be both natural (burrowing and foraging animals) or anthropogenic (grazing by livestock), disturbance on and off heuweltjie will be considered when evaluating plant community structure and function. Thereafter, analysis will be done of which plant species are being brought into the heuweltjie soils by the termites and what chemical constituents provides the foundation for discussing the role of termites in the development of soil salinity on heuweltjies.

1.2 Aims and objectives

The central aim of this study is to investigate the interaction amongst vegetation, termites, and geochemistry on the heuweltjies of the west coast of South Africa, at a local scale (on and off the heuweltjie), and a regional scale (Fynbos and Succulent Karoo biome) to determine possible sources of salinity in the biotic community. In order to achieve this, four objectives have been developed which will be investigated in four separate Chapters.

Objective 1: To identify and describe patterns in vegetation community composition diversity and function across heuweltjies 1) between the Fynbos and Succulent Karoo biomes; 2) along a gradient from the centre to off the mound; and 3) between levels of livestock disturbance.

Key questions:

1. What are the patterns in plant community composition and diversity amongst the biomes, heuweltjie zones, and disturbance levels?
2. What are the patterns in plant functional diversity amongst the heuweltjie zones in each biome?

Objective 2: To identify salt concentrations in and on the plant species growing across the heuweltjie mound.

Key questions

1. How do concentrations of calcium oxalate and salinity (cations and anions) in plant species differ between plants growing on and off the mound?
2. Do the ions deposited on the external surfaces of plants significantly contribute to the salinity distribution of the ecosystem and can the origin of these salts be identified?

Hypothesis

1. Ion concentrations in plant tissues will differ in plants growing on and off the mound
2. Marine aerosols deposit marine salt on plant surfaces which then become a source of salt into the ecosystem.

Objective 3: To assess foraging interactions amongst termites, rodents and plants on heuweltjies.

Key questions:

1. What plant species do *Microhodotermes viator* forage and do they forage for specific parts of these plants?
2. Can plant species targeted by termites and rodents be identified using carbon isotopes?

Hypotheses

1. Termite's forage for specific plant species and for specific plant parts or litter types.
2. Carbon isotopes can be used to identify plant species foraged by termites and rodents.

Objective 4:To investigate whether termites, rodents and plants play a role in the salinity distribution in the heuweltjie ecosystem.

Key questions

1. How do concentrations of calcium oxalate and salinity (cations and anions) in plant species differ between plant matter targeted by termites and rodents, or those not targeted?
2. Could foraging activities of termites and rodents increase salinity in heuweltjie centre mound soils ?

Hypotheses

1. Salts and oxalates are being brought into heuweltjies through the foraging activities of termites and rodents.
2. Foraging activities of termites and rodents increase salinity in heuweltjie centre mound soils.

1.3 Thesis structure

The thesis consists of six chapters. Chapter 1 is a general introduction to the entire thesis, the following four Chapters (Chapter 2- 5) each correspond to an objective above and are structured each with their own introduction, methods, results, and discussion sections. Chapter 5 is a discussion chapter using information from the previous three chapters to answer the objectives. Chapter 6 is the final summary chapter for the entire thesis. See Figure 1.2 for a visualisation of how the objectives fit together.

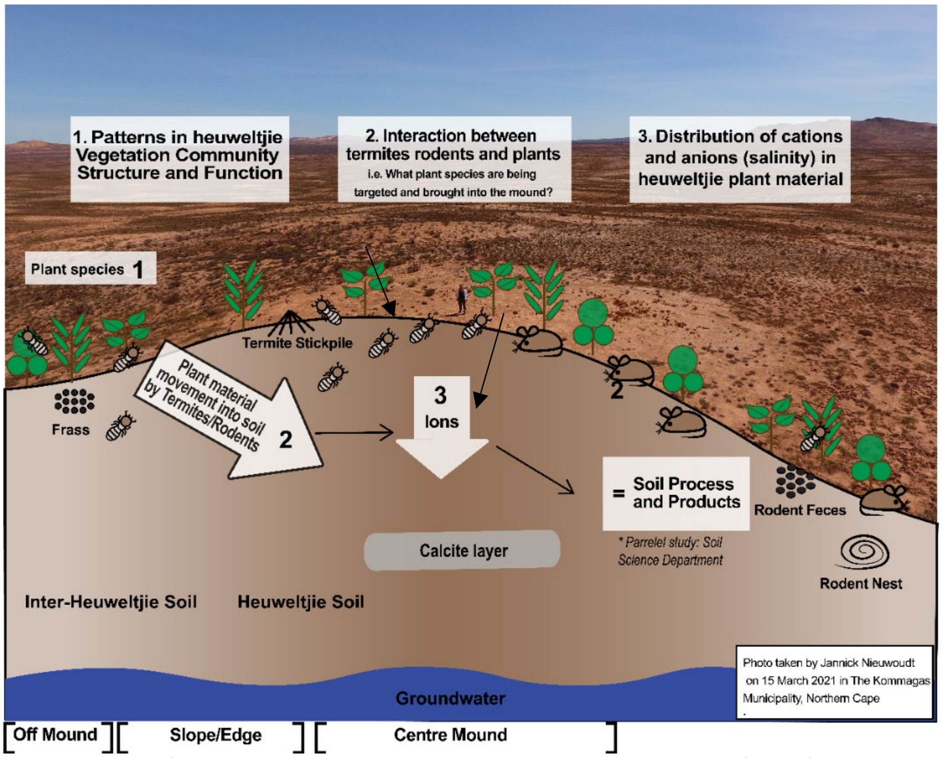


Figure 1.2: Conceptual model of the masters project showing the possible pathways of ions from plant material, moving into mound soils.In context of the mound structure as well as the parallel projects of the research group.

1.4 Study Areas

The distribution of heuweltjies occurs across the south and west coast of South Africa, but this project specifically focused on the heuweltjies found on the western coast, and the two sites occurred at the edges of the heuweltjies coastal range (Map: Figure 1.3). The sites were chosen to be representative of the heuweltjieveld that occurs in both these regions. In each biome two sub-sites were chosen: One which was slightly more disturbed, with evidence of livestock utilisation, and another that was more pristine/unaffected by anthropogenic activities, but because grazing by natural animals does still occur here, more appropriately classified as less disturbed. It unfortunately was not possible to find completely pristine sites untouched by any anthropogenic activities. Photos showing the landscape at each of the sites can be seen in Figure 2.3.

The Succulent Karoo study area is near the town of Springbok, which can be found at the northern end of the west coast. This biome is a semidesert region with an even mild climate and strong maritime influence. The more disturbed site is located near the town of Buffelsrivier (GPS: 29° 45’ 47.1’’ S; 17° 38’ 21.0’’ E) with clear evidence of overgrazing by the goats, cows and donkeys that roam freely around the town. This area is dominated by the Namaqualand Blomveld (Skn3) vegetation type (Mucina and Rutherford, 2006) and is approximately 55kms from the coast, while the less disturbed site is located on municipal land near the town of Kommagas, around 5 km (estimated) from the protected Namaqualand National Park (GPS: 29° 54’ 43.4’’ S; 17° 27’ 22.6’’ E). It was chosen as the pristine site as the vegetation was comparable with the vegetation in the protected park and it was located 20 km from the town of Kommagas and approximately 33kms from the coast. However, the vegetation is most likely still lightly grazed by some natural grazers and the odd sheep, goat, and donkey. The vegetation type of this site is Namaqualand Heuweltjieveld (SKn4).

The Fynbos study area is near the town of Piketberg on the southern end of the west coast heuweltjie range, both sites are located close to an area known as Het Kruis, on a farm (Duikerfontein), that produces vine cuttings, oats, rooibos, and cattle. It is approximately 43 kms from the coast. The more disturbed site is located next to a rooibos field with evidence of cattle presence (GPS: -32° 34’ 55.72” S; 18° 46’ 25.61”E) while the less disturbed site is on top of a nearby mountain/hill where the vegetation appears much more intact and less disturbed (GPS: 32° 34’ 25.12”S; 18° 47’ 3.38”E), cattle had access to this area but were never seen roaming, this is likely due to the location being more difficult for them to reach and thus was expected to mostly be grazed by native herbivores. The vegetation types of these sites are Graafwater Sandstone Fynbos (FFs2).

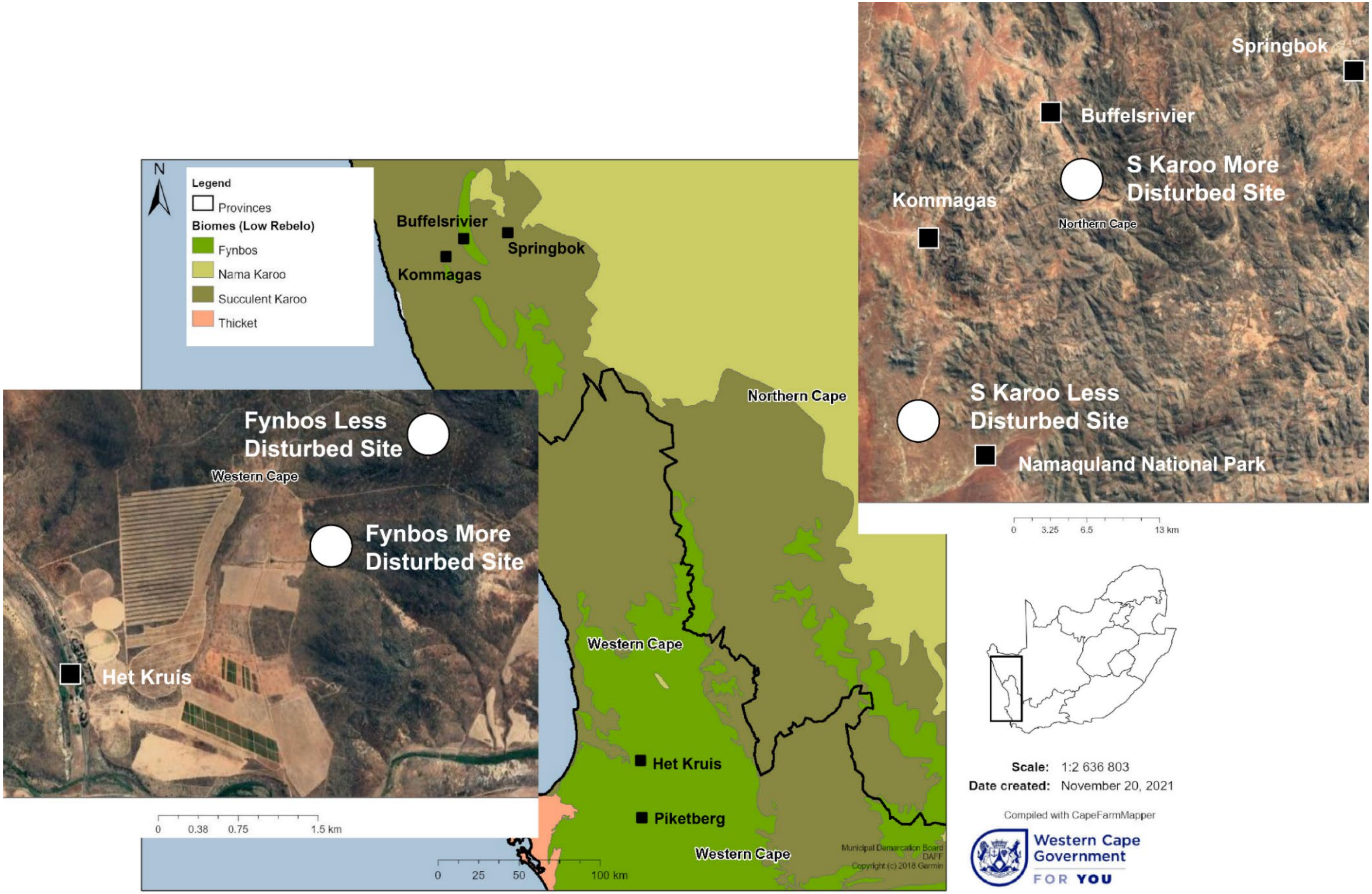


Figure 1.3: Map showing the locations of the four study sites along the western coast of South Africa, with reference to nearby towns and the Low and Rebello Biome classification depicted. White circles represent the locations of each study site and black squares represent nearby towns or locations close to the study sites.

1.5 Literature review

1.5.1 Heuweltjies contribution to landscape diversity

South Africa is home to high plant diversity, with over 20 000 species dispersed across 11 biomes (Mucina and Rutherford, 2006). Three biomes of note for this study are the Succulent Karoo, Fynbos, and Nama Karoo biomes, which occur in semi-arid to arid areas located on the west coast of South Africa. They host an unusually high plant biodiversity than what would be expected for the local climatic conditions (Cowling et al., 1999; Mucina and Rutherford, 2006; Petersen, 2008). Typically, arid ecosystems do not support very high biodiversity because of limiting resources, such as water, that characterise these environments. However, heuweltjies are a feature known to enhance biodiversity in arid ecosystems, (Desmet, 2007; Francis et al., 2013, 2007; McAuliffe et al., 2014; Midgley and Musil, 1990; Moore and Picker, 1991; Picker et al., 2007). Heuweltjie is a local Afrikaans term for these mounds, which translates to "little hill". They are sometimes referred to as Mima-like mounds and can be considered unique azonal ecosystem features that can be found dispersed across the land surface. The role they hold as a source of spatial heterogeneity in the landscape can be directly observed because the vegetation growing on heuweltjies can be visually different (Francis and Poch, 2019; Kunz et al., 2012; Midgley and Musil, 1990; Schmiedel et al., 2015). However, there are still many gaps in the knowledge relating to their basic functioning and structure.

Heuweltjies have been found to support higher plant diversity, host higher disturbance levels, and have different soil structure that supports higher nutrients and improved water content than the surrounding areas. Studies have been done to describe their density and distribution (Cramer and Midgley, 2015; Lovegrove and Siegfried, 1989, 1986), vegetation composition and cover (Booi, 2011; Knight et al., 1989; Kunz et al., 2012; Luther-Mosebach et al., 2012; Midgley and Musil, 1990; Schmiedel et al., 2016), soil composition (Booi, 2011; Cox et al., 1987; Francis et al., 2013, 2007; Kunz et al., 2012; Midgley and Musil, 1990; Vermooten, 2019), as well as to investigate the role of termites (Cornell, 2014; Dean, 1993; Francis and Poch, 2019; McAuliffe et al., 2019a, 2019b). The relationship amongst vegetation-soil-water and termites is a complicated one that needs further investigation. Understanding the ecology and the mechanisms that underlie the vegetation is important in order to have an overall understanding of the structure and function of heuweltjies and the role they play in the landscape, including with the groundwater.

1.5.2 Landscape salinisation influences

The two most commonly cited natural mechanisms of salinisation in the region where heuweltjies are found, are deposition of wind-blown marine salts and evaporative concentration of precipitation, both of which leave salts deposited on the land surface or concentrated in the shallow soil layers (Adams et al., 2004; Gorji et al., 2017). Dust is another source of salt, on the western coast of south Africa. Infrequent berg winds are a winter phenomenon that brings dust from the arid interior of the country to the coast. Dust has been found to be a source of nutrients for the nutrient poor fynbos biome where inputs of K, Ca and Zn were found to be associated with dust (Sonderberg and Compton 2007). Other studies from Namibia attribute aluminium, iron, manganese, calcium and silicon to the continental dust associated with berg winds (Annegarn et al., 1983; Chester, 1990; Swap et al., 1996; Tlhalerwa et al., 2005).

However, due to variable salinity of groundwater in these regions, more recently, it has been suggested that heuweltjies are a source of natural salinisation (van Gend, 2018; van Gend et al., 2021; Vermooten, 2019). Although numerous studies have examined elevated nutrient and salt levels in heuweltjies (Kunz et al., 2012; McAuliffe et al., 2019b, 2019a; Midgley and Musil, 1990; Midgley et al., 2012), while other studies have investigated groundwater salinity (Adams et al., 2004; Benito et al., 2008; Salama et al., 1999), it is only recently that studies in this region have looked at a possible link between the two (van Gend et al., 2021; Vermooten, 2019). These studies suggested that the link between groundwater salinity, elevated nutrients, and salts in heuweltjies, may be fostered by biological activity in the form of vegetation, foraging insects, and animals that are associated with heuweltjies. While not the focus of this study, heuweltjies are thought to facilitate the movement of salts into the ground water through preferential flow pathways associated with animal activity. Over millennia, these salts are transferred to the groundwater system via percolation of precipitation through the unsaturated zone (Adams et al., 2004; Allison, 1988; Rushton and Ward, 1979).

1.5.3 Heuweltjie vegetation

Vegetation patterns on the surface of heuweltjies mounds have been found to differ depending on the climatic conditions or disturbance regime of the ecosystem (Turner, 2003). Vegetation either dominates on the mound or off the mound. The Succulent Karoo biome has relatively nutrient-rich soils but is water limited. Rainfall is sporadic, vegetation is sparse and dominated by dwarf succulent shrubs, with many of them from the families Aizoaceae and Crassulaceae (Luther-Mosebach et al., 2012; Mucina and Rutherford, 2006). The Succulent Karoo is also known to host the seasonal mass flowering of annuals during the winter rains (Luther-Mosebach et al., 2012). The Fynbos biome, on the other hand, has nutrient-deficient soils, and a strong seasonal winter rainfall pattern. Vegetation is dominated by highly diverse fine leafed shrubs that are adapted to fire such as Proteaceae, and Ericaceae. Both biomes are part of a global hotspot known as the Greater Cape Floristic Region (GCFR), and host a high number of endemic and endangered plant species (Cowling et al., 2015).

Multiple projects have been undertaken to understand patterns in heuweltjie vegetation throughout the Succulent Karoo. Many of these projects found that heuweltjie centres are dominated by annual plants (Desmet, 2007; Luther-Mosebach et al., 2012; Schmiedel et al., 2016) and opportunistic perennials (Desmet, 2007; Luther-Mosebach et al., 2012), while total vegetation cover, species richness and perennial plants were found to be higher off mound (Schmiedel et al., 2016). Luther-Mosebach et al (2012) explains these patterns through heuweltjie plants being adapted to disturbed environments. Annual and opportunistic perennials are fast-growing plant species that make use of the increased nutrient environment. Evergreen and deciduous plants occur in equal cover on the mound while evergreen plants dominate off mound (Midgley and Musil, 1990). Deciduous plants are successful in these soils due to increased nitrogen in the soils and improved soil water accessibility (Midgley and Musil, 1990).

While most research of heuweltjie vegetation patterns has been conducted in the Succulent Karoo biome, heuweltjies also occur over areas of the Fynbos biome. Knight et al (1989) studied the heuweltjies at a Fynbos

– Karoo border in the Clanwilliam district and found that Asteraceae, succulent, and annual floras dominated on mounds. Booi (2011) was one of the few projects which studied heuweltjies in the Fynbos and Succulent Karoo biomes. The study however did not find significant differences amongst plant growth forms growing on compared to off the mounds but differences between different sites abiotic environments were observed (explained in next section). Importantly, the project did not study life-history aspects and structural attributes of vegetation such as deciduousness, plant height, and the spatial arrangement of individuals on heuweltjies.But it did find that on mound areas

had a higher cover of plant litter, mosses and termite frass, and a lower cover of lichens. Higher rainfall areas had higher non-succulent shrub cover (Booi, 2011).

Vegetation in any ecosystem establishes as a result of the surrounding environment. Soil structure, soil chemistry, water regimes, and amount and types of disturbance play a role in what type of vegetation establishes in an environment. Succulence is a water-conserving life strategy that plants have adapted to survive in dryer conditions by storing water in fleshy plant tissue (Males, 2017; Ogburn and Edwards, 2010) or in some cases it can be an adaption that halophytic plants use to survive in saline soils (Luttge, 2004; Males, 2017; Ogburn and Edwards, 2010). Opportunistic plant species like annuals, deciduous plants, or short-lived perennials have adapted to survive in highly disturbed environments (Whitford and Kay, 1999). Annual and ephemeral plant species typically have short lifespans but produce lots of seeds and grow only when conditions are optimal. Deciduousness is another plant functional type found to be important to heuweltjie vegetation (Midgley and Musil, 1990). These plants lose their leaves for part of the year as an adaptation for survival, as opposed to evergreen plants that maintain green leaves the entire year.

1.5.4 Heuweltjie soil

1.5.4.1 Structure and salt content of heuweltjie soils

Underlying these vegetation patterns is the unique soil environment of heuweltjies. Although the vegetation is mostly affected by the composition of the surface soil layer, soil changes in heuweltjie can also be found at depth. Midgley and Musil (1990) suggest that the distribution of plant life forms is because of the soil nutrients in heuweltjies soils, however other abiotic factors such as soil type, texture, and water holding capacity may also play a role in influencing vegetation patterns. Francis et al (2007) suggested that biodiversity across heuweltjie-veld is due to more than soil fertility or rooting depth since the availability of water is also important. In arid environments, soil plays an important role in connecting vegetation communities to water availability where the two most important factors in soil that influence this function is the presence of salts and clay (Francis et al., 2007). This is supported by Booi (2011) who highlighted the importance of water availability in heuweltjie soils for their structure and functioning. Heuweltjie on-mound soils have been found to have a finer texture, be more alkaline, have a higher clay content and hold more water (Francis et al., 2007; Kunz et al., 2012; Midgley and Musil, 1990). The heuweltjies also hold higher amounts of salts and nutrients, total nitrogen, total organic carbon content, phosphorous, manganese and potassium, and calcite (Booi, 2011; Kunz et al., 2012; Midgley and Musil, 1990; Moore and Picker, 1991). Kunz (2012) found the nitrogen and phosphorus levels decreased from the centre to the border of heuweltjie mounds. However, the carbon concentration did not differ across this local gradient (Kunz et al., 2012). Schmiedel et al. (2016) found elevated calcium carbonate levels in the centre of the mound, and that organic carbon was the lowest in the fringe (edge) area of the mound. Nutrients such as nitrogen and calcium are important for plant growth, which could influence floral structure and function across heuweltjies (Schmiedel et al., 2016).

1.5.4.2 Calcium carbonate and calcium oxalate minerals

The oxalate- carbonate pathway is a soil process which cycles calcium and carbon from the atmosphere through an ecosystem into the soil. Ecosystems where this process exists have the potential to act as significant carbon sinks (Cailleau et al., 2011). This means the oxalate-carbonate pathway may be important for atmospheric CO_2 reduction (Cailleau et al., 2011), which directly contributes to slowing down the effect of climate change. Previous studies in forest ecosystems have been able to prove that the oxalate-carbonate pathway occurs (Calleau et al 2011). Forest ecosystems are already seen as massive carbon storage systems due to the high biomass of the plants which make up these ecosystems. In combination with the carbon storage in soils via this pathway, a forests' role in carbon sequestration becomes even more important (Cailleau et al., 2011). However, if we can prove this process exists in heuweltjies it will have significant conservation implications for arid ecosystems. Arid ecosystems are not seen as exceptionally productive or important for carbon storage due to the often-significant lack of aboveground plant biomass. By showing that the oxalate-carbonate pathway exists in heuweltjies we can make a further case to conserve them because of their role in offsetting the effects of climate change.

1.5.4.3 Natural and Anthropogenic Disturbance

Heuweltjies soils become hotspots of biological activity as they provide resource-rich and protected environments that attract other burrowing animals in addition to termites. Mammals such as aardvark and mole ratsuse the heuweltjies as places of refuge to build burrows and forage for food (Helme, 1990; Milton and Dean, 1990). Cox et al. (1987), conducted a study looking at stone size on heuweltjies to determine the role that termite and mole rats have in heuweltjie functioning. They hypothesised that small stones get moved laterally into the mound by mole rats and that termites bring fine soil particles to the surface. Gravel and pebbles were found to be much higher on mounds than off mounds and small stone density increased with depth i.e. finer soils were near the top (Cox et al., 1987). The presence of burrowing animals introduces a significant aspect of disturbance to this ecosystem, which can be observed on most heuweltjies (Helme, 1990; Knight et al., 1989; Milton and Dean, 1990) and indicates that they significantly affect the soil and vegetation environment. Most research on heuweltjies accepts disturbance as an integral part of heuweltjie structure. In addition to this natural disturbance, anthropogenic disturbance in terms of livestock grazing has been found to have an effect on heuweltjie centre mound communities (Kunz et al., 2012; Rahlao et al., 2008; Schmiedel et al., 2016). It was shown that vegetation on heuweltjie centres is attractive to herbivores such as sheep (Kunz et al., 2012; Schmiedel et al., 2016), further increasing the amount of disturbance in these soils.

Once disturbed, heuweltjies centres take the longest time to recover and return to a pre-disturbed state, while the fringes were the least impacted by disturbance (Schmiedel et al., 2016). Schmiedel et al. (2016) concluded that this was because the unique environmental conditions of heuweltjie centres, which were described as "hotspot of disturbance", kept the heuweltjie centre in a state of disturbance catalysed by historical and current grazing pressures which prevented fast vegetation recovery.

1.5.5 Termites and Heuweltjies

1.5.5.1 The role of a termite in an arid environment

Termite species are examples of ecosystem engineers (Kunz et al., 2012; McAuliffe et al., 2019a), as they are species that can change the environment around them. This can be seen on heuweltjies where termites play a direct and indirect role in altering both biotic and abiotic components of these ecosystems. Directly, termites enrich soils with nutrients, salts, and minerals by bringing plant material into the heuweltjies and depositing frass (Francis et al., 2012). The presence of termites are normally assumed to correspond with higher micronutrients in soils where they are found, compared to adjacent mound soils, however most of the research that has been done, has been for fungus growing termite species (Mills et al., 2009; Nampa and Ndlovu, 2019). *Microhodotermes viator* does not grow fungus. Mills et al (2009) studied the effect *Trinervitermes* a non - fungus culturing (NF) termite species has on micronutrients in soils, in comparison to a fungus-culturing (F) species (*Macrotermes*) and found that the NF mounds did not show enrichment of micro-nutrients in the soil. *Trinervitermes* termite mounds were also studied by Nampa and Ndlovu (2019) where they found opposing results - that this species mound soils, are nutrient enriched. Many of the larger animals utilise them as a food source. Termite presence also changes the soil structure and soil texture of heuweltjies, as their underground nests consist of tunnels and chambers which aerate the soil (Schmiedel et al., 2015). Soil from the depths of the heuweltjie is brought to the surface as termites maintain these structures (Cox et al., 1987). Soil is also deposited on the surface with saliva to keep it stuck together (Coaton and Sheasby, 1974; Dean, 1993), which increases the soil water content, and results in soil on mounds consisting of finer materials and with higher clay contents (Kunz et al., 2012; McAuliffe et al., 2019a; Schmiedel et al., 2015). Soil water conditions are ultimately improved by termite presence because of the increased soil carbon content and clay.

1.5.5.2 Microhodotermes viator

Microhodotermes viator (harvester termite) is the termite species associated with heuweltjies. They are endemic to South Africa and found mainly in the Eastern Cape, Northern Cape, and Western Cape provinces, along with part of the North-West (Coaton and Sheasby, 1974). Evidence for *Microhodotermes viator* affecting the structure of heuweltjie soils can be found in Cornell 2014, where nutrient profiles of heuweltjie soils were found more similar in composition to termite frass than the off mound soils (Cornell, 2014). They are known locally as houtkapper (woodcutter) or stokkiesdraer (stick carrier) termites (Coaton and Sheasby, 1974). *M viator* forage in groups across the soil surface for vegetation at unpredictable times and temperatures (Dean, 1993). Workers leave nests through foraging ports or access holes located around the mound and forage anywhere from 1 to 45 meters away — depending on the density of colonies in the area (Dean, 1993). These holes are closed once foraging is completed but can be identified by stick-piles and frass dumps (Dean, 1993). *M. viator* have been found to forage on soil surface temperatures in the range of 11.5 to 55.6° C (with a mean 31.8° C) and are active all year round but are more active in summer (Dean 1993).These termites live in subterranean nests (Coaton and Sheasby, 1974), and are most often associated with heuweltjies. However, the colonies can construct small, cemented conical mounds above ground or sometimes produce no structure at all (McAuliffe et al., 2019a). Colonies are territorial with only one colony occupying a nest (Dean, 1993). These nests can be found dispersed regularly throughout the landscape, where this type of pattern is often owed to territorial competition for resources by animals, or in this case a colony. This has been used as a possible explanation for the even distribution of heuweltjies in the landscape (McAuliffe et al., 2019a).

1.5.6 Heuweltjies origin

The debate on how heuweltjies were formed is still ongoing in the literature. Heuweltjie formation is not the focus of this study, only the fact that termites *M. viator* can be found living in them now, is relevant to this study. Although for the purpose of this study formation is not important, the many theories which have been proposed for their formation will be briefly summarised. The most accepted explanations involve the termite species *M. viator*, as these species are hypothesised to modify and displace the soil around their nests to in order create the mounds (Francis et al., 2013; Moore and Picker, 1991). Other studies suggest the same method for formation, but with the addition of burrowing animals which also move soil, where they are responsible for displacing larger stones than termites can, and add further nutrients to create the heuweltjies (Cox et al., 1987; Lovegrove and Siegfried, 1986; Milton and Dean, 1990). A third theory is that they were formed by a different and extinct species of termite which existed during the late Pleistocene (Midgley et al., 2002). The fourth theory is that termites were not involved at all but were only secondary inhabitants of heuweltjies. Instead, these mounds were formed by processes, where due to the presence of dense clumps of vegetation, soil or aeolian sediments got trapped and the collected around the vegetation to create mounds (Cramer et al., 2017, 2012). McAuliffe (2019), recently proposed a system dynamics model which combines both biotic and abiotic processes together into a sequence of transitions which build-up to the creation of heuweltjies (McAuliffe et al., 2019b, 2019a).

Chapter 2: Patterns in Heuweltjie Vegetation Community Composition and Function

2.1 Introduction

The vegetation of the southwestern tip of Africa, known as the Cape Floristic Region, is recognised worldwide for is floristic uniqueness and exceptional plant diversity. The Cape Floristic Region is the geographically smallest, of only six Floral Kingdoms worldwide (Linder et al., 2010; Myers et al., 2000). The unique Fynbos biome dominates this well-known southwestern corner of South Africa (Cowling, 1992), but it is not the only floristically renowned area of the country. The Succulent Karoo biome in the northwest along with the Fynbos biome have been recognised as Global Ecoregions and Biodiversity Hotspots (Myers et al., 2000; Olson and Dinerstein, 2002). This is due to the exceptionally high plant diversity, richness, endemism, as well as rate of habitat loss that these areas experience (Myers et al., 2000). Combined, these two biomes are known as the Greater Cape Floristic Region (GCFR), recognised, and classified as one, not only due to the high rate of plant endemism occurring in both these biomes but also as both share winter rainfall climates (Born et al., 2007; Cowling et al., 1999; Mucina and Rutherford, 2006).

With the Succulent Karoo biome in the north and the Fynbos biome adjacent in the south, the GCFR dominates the landscape along the western coast of South Africa. The Fynbos biome has a strong winter rainfall climate, receiving on average 480 mm of rainfall annually (Mucina and Rutherford, 2006). The Fynbos is an extremely species rich and diverse Mediterranean-type biome, characterised by the presence of small-leafed, evergreen shrubs, bulbous plants, and reeds (Restionaceae), most of which rely on fire to reproduce and survive. Soils of the Fynbos are typically very low in nutrients (Cowling, 1992; Mucina and Rutherford, 2006; Verboom et al., 2017). The Succulent Karoo biome is a semi-desert region with an even, mild climate, and with a strong maritime influence and winter rainfall. On average, the rainfall of the Succulent Karoo is 170 mm a year, which is much lower than in the Fynbos. Soils of the Succulent Karoo have special features that amplify and maximise water supply by modifying the water infiltration processes, such as surface crusts or gravel pavements that break the fall of rain and allow water to infiltrate and reach plant roots more effectively (Francis et al., 2007; Mucina and Rutherford, 2006). Higher rainfall in the Fynbos causes leaching and, therefore, soil nutrients are also much higher in Succulent Karoo soils compared with the Fynbos (Mucina and Rutherford, 2006).

Both the Fynbos and Succulent Karoo biomes host higher plant diversity than would be expected for their relatively dry climatic conditions. There are many reasons for this, including long-term climate stability and edaphic and topographic diversity (Cowling, 1992; Mucina and Rutherford, 2006). In the Fynbos biome in particular, fire-driven speciation and low rates of extinction are seen as major contributors to its high species diversity (Rundel et al., 2018). Apart from the above, the presence of heuweltjies create an interesting landscape phenomenon on the west coast in both biomes, as they have been found to impose heterogeneous vegetation patterns and soil conditions on the landscape, by hosting different suites of species on the heuweltjie mounds compared with the surrounding inter-heuweltjie matrix (Knight et al., 1989; Kunz et al., 2012; Luther-Mosebach et al., 2012; Schmiedel et al., 2016). For these reasons they are starting to be recognised as being one of the contributing factors to the high diversity of these biomes.

Figure 2.1: Heuweltjies in the Northern Cape in spring, showing mounds dominated by purple flowing Aizoaceae species, Taken by Nicola Vermonti on the 8th September 2020.

Heuweltjie vegetation assessments have been undertaken before with most studies based in the Succulent Karoo (Desmet, 2007; Kunz et al., 2012; Luther-Mosebach et al., 2012; Midgley and Musil, 1990; Schmiedel et al., 2016), or the Karoo–Fynbos interface (Knight et al., 1989). Results from each study all differed slightly. Annual plants, especially of the family Asteraceae, opportunistic perennials, succulents, or deciduous plants were found to dominate on heuweltjie mounds in the various studies (Desmet, 2007; Knight et al., 1989; Midgley and Musil, 1990; Schmiedel et al., 2016), while the total cover of vegetation, species richness, and perennial plant cover was found to be lowest on heuweltjie centres increasing to off-heuweltjie mounds (Schmiedel et al., 2016). Kunz et al. (2012) conducted a study focused on the interaction amongst heuweltjies, vegetation, and grazing and found that an increase in natural disturbance by local burrowing and foraging animals, as well as utilization through grazing, resulted in a decrease in the cover of perennial plants and an increase in annuals and geophytes. In turn, Luther-Mosebach et al. (2012) found that salinity was the most prominent environmental factor influencing species richness and composition across Namaqualand. Saline sites in the landscape are expected to include the centres of heuweltjies. As most plants are salt intolerant, we can thus expect to find salt-tolerant plants growing on the centres of heuweltjies (Milton and Dean, 1990), suggesting plant form and functional change across the mounds.

The presence of termites as well as burrowing and foraging animals, such as rodents and aardvarks, simplify the soil environments by introducing an aspect of disturbance. This interaction has been found to result in there being little difference in the species richness on or off heuweltjie mounds (Kunz et al., 2012), despite nutrient-rich centre mound soils. However, the species assemblage composition between the mound and matrix has been found to be distinctly different. The centres of heuweltjies tend to host more opportunistic plant species (annual and geophytes) that thrive in disturbed environments, while perennials dominate off mounds (Kunz et al., 2012). Apart from natural disturbance, a major human-induced disturbance to natural ecosystems is grazing pressure by livestock, which can occur on or off heuweltjies. Grazing animals have been found to prefer and remove perennial species from the landscape, which results in an increased cover of annuals species due to a release in competition for resources with the perennial species (Kunz et al., 2012). Grazing by native animals can also play a role in creating this trend in the vegetation but not to the extent that livestock farming can.

The above knowledge on heuweltjies clearly suggests environmental gradients exist from the centre of heuweltjies to the surrounding off-heuweltjie matrix. These gradients would thus affect heuweltjie vegetation communities. Therefore, the overarching aim of this chapter is to identify the vegetation community composition and diversity on and off heuweltjies across the two biomes, as well as potential differences in function, to not only add to the existing literature on heuweltjie landscapes in the GCFR but also to ensure a more systematic sampling strategy for subsequent data collection for Chapters 2 and 3; for example, to know which are the most common and abundant flora to sample per site and how species composition and life form change could impact the results. Lastly, most research on heuweltjie vegetation is confined to single sites mostly within one biome. Very few studies have attempted to identify common patterns and traits in the vegetation on heuweltjies between two biomes (Booi, 2011), nor the effect of landscape disturbance in both.

2.2 Methods and Materials

2.2.1 Sampling design

Across both biomes there were four sites in total (see Chapter 1 for an in-depth description of the sampling sites), with each biome consisting of two subsites that were classified as 1) more disturbed (MD) and 2) less disturbed (LD) (the former indicating high anthropogenic disturbance from livestock grazing, and the latter that grazing mostly by native animals occurred). At each of these subsites, four heuweltjies were chosen and sampled. Google Earth and, where possible, preliminary field visits were used to assess and choose the heuweltjies. A stratified sampling method was used to identify the right set of conditions to choose the heuweltjies that were studied. They needed to be distinctive in the landscape, be approximately 15–30 m wide, have the typical round shape, show active termite activity (have frass present), and occur on a west-facing slope. Heuweltjies that met these conditions and were the most accessible were subjectively chosen (see Figure 2.3).

Each heuweltjie mound was then split into three zones: the centre of the mound, the slope (or intermediate area of the mound), and off the mound. Zones were determined by visually assessing the slope or gradient changes of the ground as well as measuring the distances from the centre point of the heuweltjie to the edge of the mound to best estimate the sampling zone boundaries. Photo evidence using a drone (see Appendix 6) was used to confirm that the sampling locations were in fact within each of the zones.

Both study areas experience winter rainfall (Mucina and Rutherford, 2006). The fieldwork, therefore, occurred during spring to early summer, as this is the flowering season for plants. Because annual or geophyte species only temporarily appear, plant diversity is at its highest during this time; additionally, the plants were flowering, which was needed to identify the plants species (Richard et al., 1999).

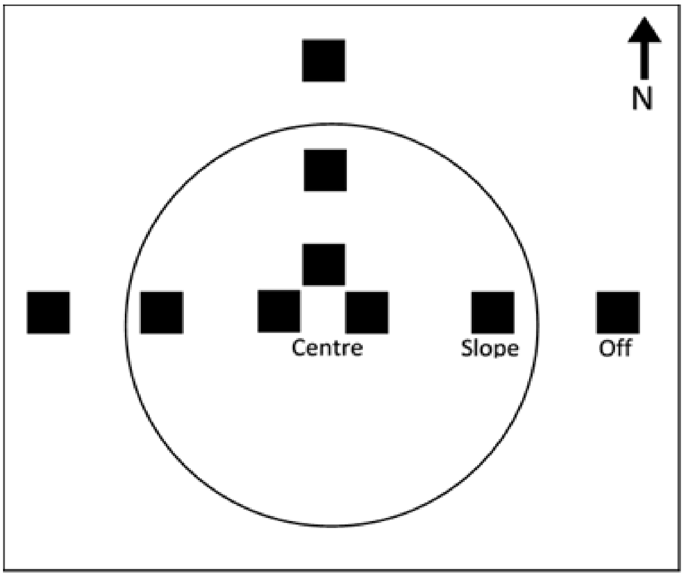


Figure 2.2: Diagram of the vegetation sampling design of the study. The circle represents a heuweltjie, and each zone is labelled (Centre, Slope, and Off). The black squares represent the quadrats where the vegetation samples were taken.

2.2.2 Field Work

Fieldwork was completed from September to December 2020. Quadrats (2 x 2 m) were used to collect the vegetation and surface characteristics' data. Quadrats were placed at points along an imaginary line, starting at the centre to off the mound and moving off, in the direction of a cardinal point, with one in each zone. Three of these transects were taken, resulting in three quadrats sampled in each of the heuweltjie zones, as depicted in Figure 2.2. One orientation (north, south, east, and west) was randomly left out on each heuweltjie. This resulted in nine samples being recorded per heuweltjie; thus, thirty-six (4 x 9) quadrats were sampled at each of the sites, the only exception being the Succulent Karoo less disturbed subsite, where seven heuweltjies were sampled (due to extra time), resulting in sixty-three quadrats being recorded.

Plants species were each given a unique code and were identified where possible to species level (see species list in Appendix 7). An estimate of the percentage cover of each species, the number of individuals, and height of the tallest individual of each species in each quadrat was made (to get a very general description of vegetation height). Bias and estimation errors were reduced by using two observers to assess the percentage cover and ensuring one person (myself) was always a constant. Species identifications were done by myself, using iNaturalist and various field guides (Manning, 2018; Manning and Goldblatt, 2012; Smith et al., 2017; Snijman, 2013). Species field codes are reported alongside species names throughout the thesis to acknowledge that possible errors could exist in the identifications. The heuweltjie surface characteristics per quadrat were also recorded, as a percentage cover or presence/absence in each quadrat. The surface characteristics include plant litter cover, frass cover, cryptogram cover (consisting of lichen, crust, and moss that covered the ground), the presence of stick-piles and earthworm casts, as well as holes (ranging from aardvark burrows to scorpion nests and termite openings) and surface disturbances. Holes per quadrat consisted of extra small (~<0.5 cm), small (~1 cm-5 cm), medium (~5-15 cm), and large (~>15 cm) sized holes, and surface disturbances consisted of any "diggings" or pits (medium and large), mole hills, and raised mounds found on the soil surface. At each quadrat, the GPS coordinates were recorded, photos were taken, the slope was estimated (ground angle), and the zone (location of the sample) recorded.

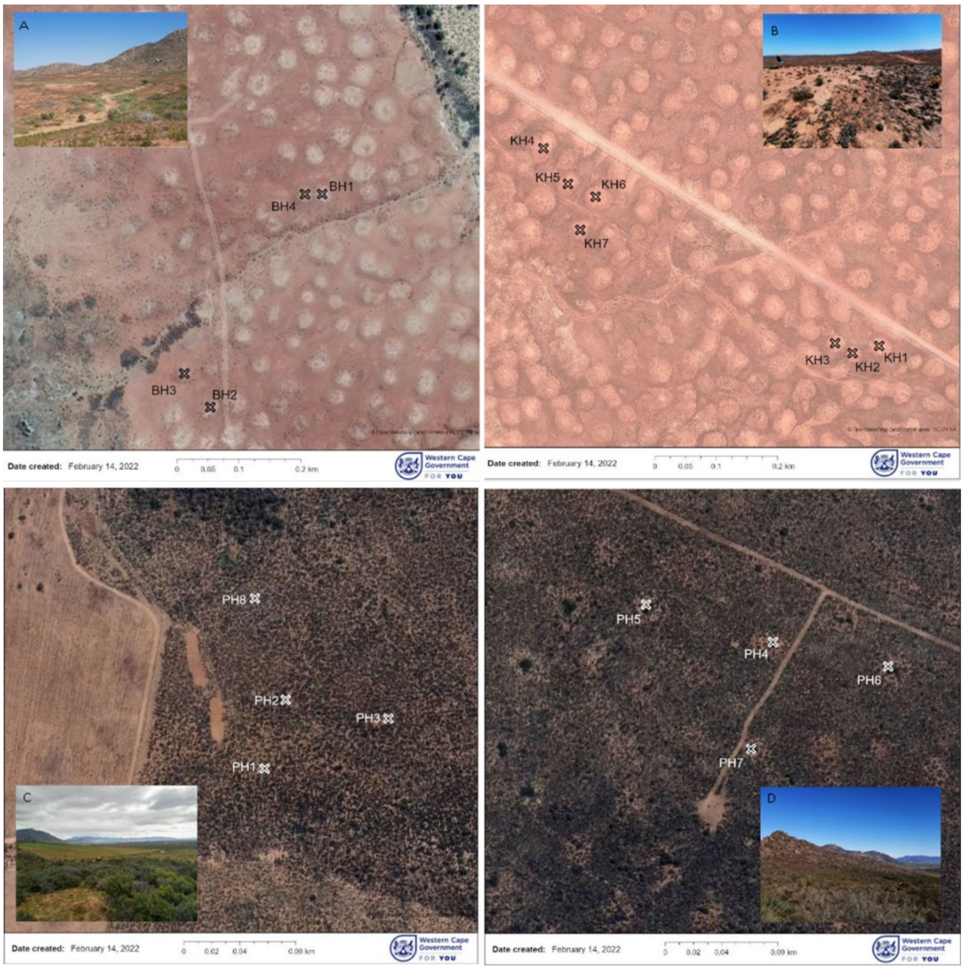


Figure 2.3 Maps and photos showing the landscapes and location of chosen heuweltjies at each of the study sites. A: Succulent Karoo more disturbed site; B:Succulent Karoo less disturbed site; C:Fynbos more-disturbed site; D:Fynbos less disturbed site.

2.2.3 Data Analysis

Using the plant species richness, abundance, and cover data per quadrat, vegetation community composition, diversity, and functional diversity were investigated by calculating the diversity indices and life-form changes across the heuweltjie zones and sites. Specifically, the calculated biodiversity indices were Shannon (H) diversity, Simpson (D) diversity, species richness, and Pielou's evenness. Species richness is the number of unique species found in an area. Abundance is the number of individuals of each plant species. Species diversity (Shannon and Simpson) are indices that can be calculated to give an indication of the number of species in a community as well as their relative abundances (evenness), where a Pielou's evenness index (J) of 0 means no evenness (dissimilar abundances among species) and 1 complete evenness (similar abundances among species).

Plants were then classified into vegetation life-form compositions (according to Table 2.1 in Mucina and Rutherford, 2006). The above indices were then calculated for each life-form group, for life-form diversity, life-form richness (number of species per life form), life-form evenness, and the percentage cover of each life form, for each heuweltjie zone.

Formulas

1) Species Richness:

The number of species

2) Shannon (H) index:

$$H = -\sum pi * ln(pi) \text{ (Equation 1)}$$

where pi = proportion of individuals in the i^{th} species

3) Simpson's index:

$$D = \sum pi^2 \text{ (Equation 2)}$$

4) Pielou's evenness index (J):

$$J = H/\ln(S) \text{ (Equation 3)}$$

where H = Shannon (H) index.

2.2.4 Statistical Analysis

The following statistical analyses were run using R, PRIMER 6, or CANOCO 5 software, as indicated:

Biodiversity statistics (species accumulation curves, vegetation cover (%), average species height (cm), abundance (per m^2), species richness (per m^2), Shannon (H) diversity, Simpson (D) diversity, and Pielou's evenness) were calculated using the vegan package in R. Plant species were then grouped according to functional type (life form) and the above statistics were rerun. The above indicators were then compared across biome, zone, and disturbance level using 1) t-tests: The data was not normally distributed and not all the variables' variances were equal; therefore, Welch Two Sample t-tests were run to compare the data between the two biomes and between the two disturbance subsites within the biomes. 2) Kruskal–Wallis tests: The data was not normally distributed, so Kruskal–Wallis tests were used to compare and find significant differences in the vegetation structure as well as the surface characteristics amongst the three heuweltjie zones (centre, slope, and off); this was followed by pairwise Wilcoxon tests.

Importantly, because we based the above tests on the quadrat data, and thus not pooled per heuweltjie, issues around pseudo replication are a concern. This would limit the statistical inferences made comparing these variables across; however, we decided to do so to capture the variety of data across and within the heuweltjies, as for this descriptive part of the thesis there were time constraints to sample a large enough number of heuweltjies. Nonetheless, with the community composition analysis, one would get an initial picture of what patterns to expect given the tested variables, and which might provide information to aid formally testing hypotheses in this regard.

PRIMER 6 was used to investigate the differences in plant communities across sites: Firstly, non-metric multidimensional scaling (NMDS) was performed on the plant species quadrat data as an indirect, distance-based technique to show any differences in the plant communities, using Bray–Curtis distances. The variables biome, zone, and disturbance level were then manually overlaid to visualise any dissimilarities among the groups.

Secondly, multivariate analyses were run on the environmental diversity indicators and heuweltjie surface characteristics) and species data for each site: PERMANOVAs and ANOSIM were run to see if there were differences amongst the plant communities and heuweltjie diversity characteristics across biome, zone, and disturbance level, where all the variables/species could be considered at once. This was followed by using SIMPER to investigate which specific species dominated the plant communities.

2.3 Results

2.3.1 Heuweltjie Vegetation Community Composition

Species accumulation curves (Figure 2.4) were run first to test if enough sampling had been done. Species accumulation curves at all sites are flattening out towards an asymptote, missing only some rare or less abundant species. The study occurred in the Fynbos and Succulent Karoo biomes, which both hosts extremely high local plant biodiversity, meaning asymptotes would be very difficult to reach. Nonetheless, the analysis shows that there were enough samples recorded to confidently characterise the main trends in richness and diversity of the flora of each site. In the Fynbos, 100 and 91 species were observed from the 72 quadrats for the LD and MD subsites, respectively. In turn, 68 and 23 species were observed from the 99 quadrats for the LD and MD subsites from the Succulent Karoo, respectively.

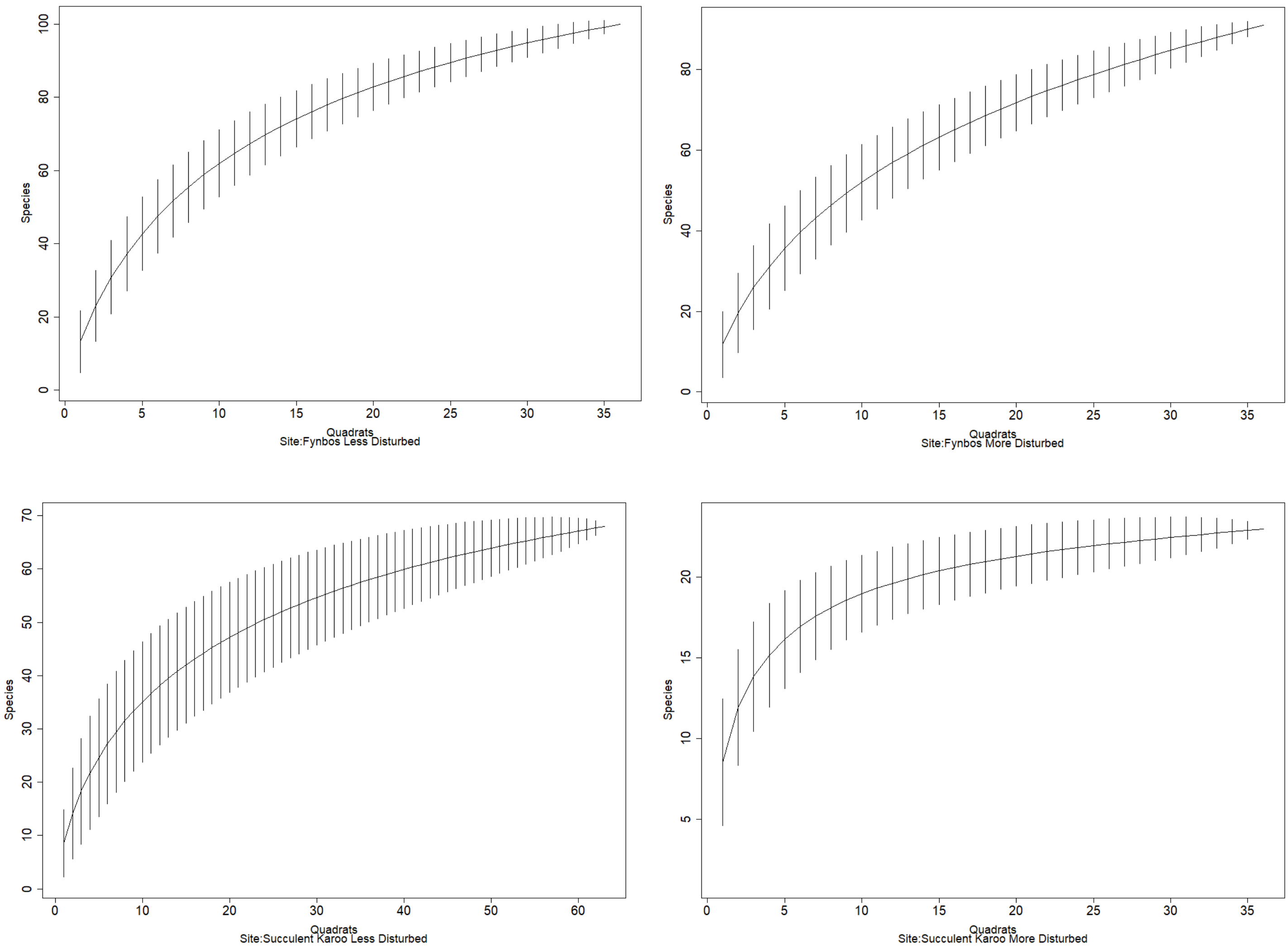


Figure 2.4: Species accumulation curves for the Fynbos and Succulent Karoo heuweltjie sampling sites, also differentiating between the more or less disturbed subsites within each biome. Showing the accumulative species richness on the y-axis and accumutive number of quadrats on the x-axis.

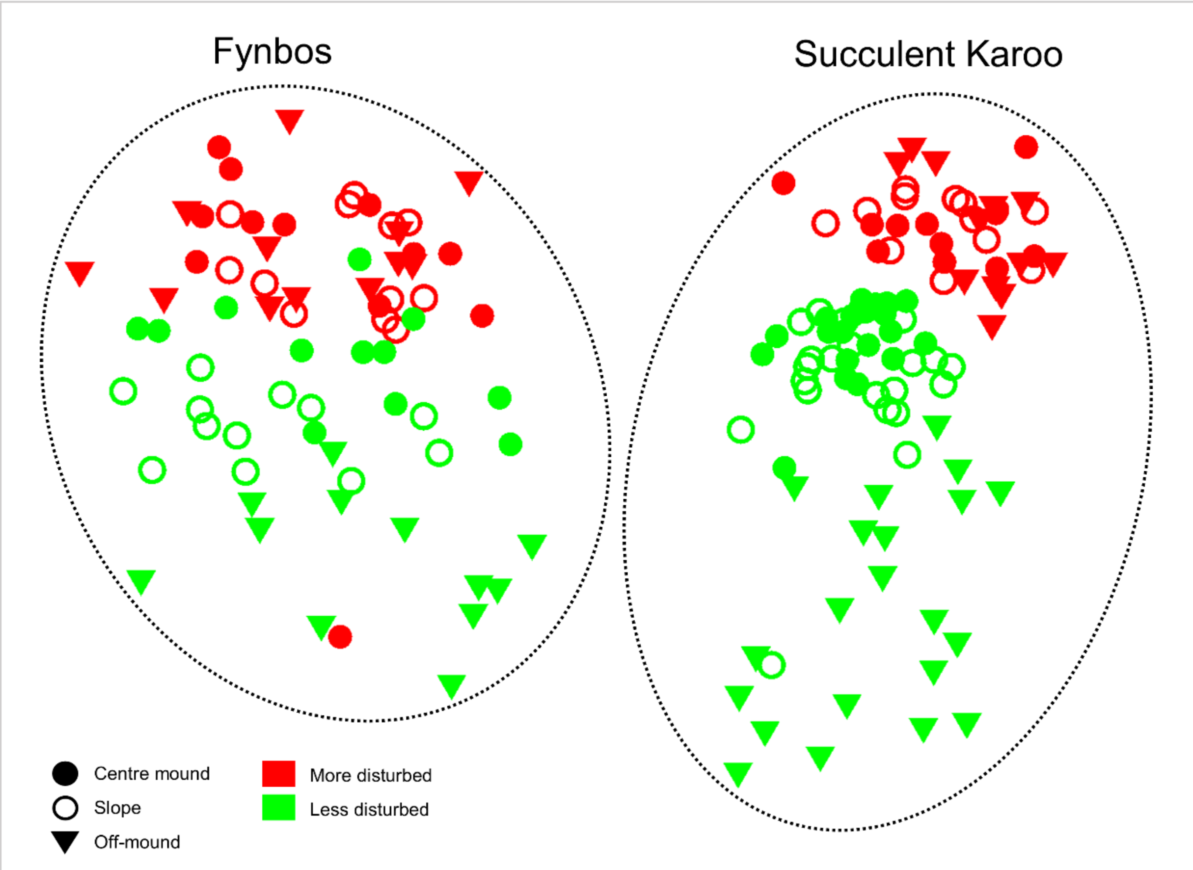


Figure 2.5: Non-metric multidimensional scaling (NMDS) plot of the heuweltjie sample–species resemblance matrix based on Bray–Curtis distances, with biome, sampling zone, and disturbance level as factors. Data were square-root transformed. x-axis = NMDS 1; y-axis = NMDS 2. 2D stress = 0.13.

Figure 2.5 shows there are distinct dissimilarities in the vegetation communities across biomes, zone, and level of disturbance. As expected, biomes separate the most obviously, with disturbance clearly influencing the plant community composition in each biome. Interestingly, even the centres of the mounds in the less disturbed areas were more similar to the disturbed sites, highlighting the effect of disturbance on heuweltjie vegetation in general. In turn, off-mound areas, or the heuweltjie matrix, are the most dissimilar among the zones in the less disturbed area, for both biomes. This data shows that most of the variation is thus driven by biome, then disturbance, and lastly the off-mound zone.

<u>PERMANOVAS analyses were run</u>. For the Succulent Karoo, the analyses indicated significant differences between the plant communities in the disturbance subsites (*Pseudo-F= 47.39; p=0.001*), and amongst the heuweltjie zones (*Pseudo-F= 12.39; p=0.001*), with the interaction amongst the zones and subsites being significant (*Pseudo-F= 11.57; p=0.001*), meaning that the disturbance level and zone depend on each other and thus should not be interpreted separately (see Table 2.1). Differences were also found amongst the environmental data for both groups of predictor variables at the Succulent Karoo: the disturbance subsites

(*Pseudo-F= 14.85; p=0.001);* the heuweltjie zones, (*Pseudo-F= 6.44; p=0.001*); and the interaction of subsites and zones (*Pseudo-F= 2.41; p=0.002*) (Table 2.1).

ANOSIM pairwise tests (Table 2.1) were then run as a post-hoc test to assess the specific the levels of similarity amongst the factor levels within each predictor variable (ANOSIM values closer to 1 indicate complete dissimilarity). Pairwise tests (Table 2.1) for and all of the zone comparisons in the Succulent Karoo were found to be significantly dissimilar from each other ($p<0.05$), with the least difference being between the centre and the slope ($R=0.05; p=0.002$); the next highest dissimilarity was between the slope and off ($R=0.30; p=0.001$), with the species composition off the mound and at the centre ($R=0.44; p=0.001$) being the most different.

<u>Succulent Karoo</u>

Table 2.1: Primer results showing PERMANOVA and ANOSIM results comparing plant communities in the Succulent Karoo amongst disturbance subsites and zones

PERMANOVA			ANOSIM: Pairwise tests by Zones		
Factor	Pseudo-F	P(perm)	Zones	R-statistic	Significance level %
<u>Species</u>					
Subsite	47.39	0.001	Centre, Slope	0.11	0.2
Zone	12.39	0.001	Centre, Off	0.44	0.1
SixZo	11.57	0.001	Slope, Off	0.30	0.1
<u>Environmental</u>					
Subsite	14.85	0.001	Centre, Slope	0.11	0.1
Zone	6.44	0.001	Centre, Off	0.20	0.1
SixZo	2.41	0.002	Slope, Off	0.06	0.9

<u>Fynbos</u>

Table 2.2: Primer results showing PERMANOVA and ANOSIM results comparing plant communities in the Fynbos amongst disturbance subsites and zones

PERMANOVA			ANOSIM: Pairwise tests by Zones		
Factor	Pseudo-F	P(perm)	Zones	R-statistic	Significance level (%)
<u>Species</u>					
Subsite	11.52	0.001	Centre, Slope	0.10	0.8
Zone	3.37	0.001	Centre, Off	0.24	0.1
SixZo	2.64	0.001	Slope, Off	0.09	2.7
<u>Environmental</u>					
Subsite	1.85	0.073	Centre, Slope	0.012	24.6
Zone	1.72	0.052	Centre, Off	0.027	10.4
SixZo	2.62	0.002	Slope, Off	-0.01	62.3

PERMANOVAS were also run to compare the Fynbos plant communities and environmental variables (Table 2.2). There were no significant differences ($p>0.05$) found for the environmental data between disturbance sites (*Pseudo-F= 1.85; p=0.073*) and zones (*Pseudo-F= 1.72; p=0.052*); however, there was still significance seen in the interaction between the environmental variables of the sites and zones (*Pseudo-F= 2.62; p=0.002*). The plant species communities, however, were found to be significantly different between subsites (*Pseudo-F= 11.52; p=0.001*) and zones (*Pseudo-F= 3.37; p=0.001*), with the interaction also being significant (*Pseudo-F=2.64; p=0.001*). Therefore, despite no differences occurring in the study-tested environmental characteristics of the Fynbos heuweltjies across zones, it does appear that the plant community composition is different in each zone.

ANOSIM pairwise similarity tests were again run to determine similarity/dissimilarity between the zones in the Fynbos (Table 2.2). The species composition in each of the heuweltjie zones was found to be dissimilar from each other ($p < 0.05$), with the centre and slope (*R=0.096; p=0.008*; Table 2.2) and slope and off mound (*R=0.10; p=0.027*; Table 2.2) having very similar R values, respectively, which were close to 0 (more similar); in turn, the species composition between off the mound and the centre (*R=0.24; p=0.001*; Table 2.2) was the most dissimilar.

2.3.2 Vegetation Structure, and Diversity

Plant diversity variables were each compared across the zones among the heuweltjies at each disturbance subsite (MD and LD) within each biome (see Table 2.3 for the results of the zones split by subsite and Appendix 3 for boxplots and Kruskal–Wallis tests by biome and Appendix 17 for pooled averages of each variable across the site). The plant community at the Fynbos site appears uniform with almost no significant differences found for any of the vegetation variables amongst the zones of the heuweltjies, while there were significant differences found across the vegetation indices at the Succulent Karoo site.

There was no significant difference found in the vegetation cover amongst the different zones on any of the heuweltjies in the Fynbos biome (Appendix 3 and Table 2.3; *p-value* < 0.05), while in the Succulent Karoo, the average vegetation cover across each of the zones at both subsites are significantly different from each other (Table 2.3; SLD: *p-value* < 0.001; SMD: *p-value* < 0.01). Pairwise comparisons show the significant differences amongst all the zones at the LD site, with the lowest vegetation cover occurring on the centre of the heuweltjie, increasing to the highest cover off the mounds and significant differences only between the centre and off mound at the MD site, where again cover is lower on the mound centres. Average vegetation cover between the Fynbos sites were found to be significantly different to each other *(p-values* < 0.01*)*, with the LD site having greater plant cover on average compared to the MD site.

The average height of the vegetation was found to be significantly different amongst zones of the heuweltjies across the Succulent Karoo site (Appendix 3; *p-value* < 0.001*)*, with the height on the centre of the mounds being significantly lower than on the slope and off the mound. This pattern is significant and reoccurs when looking at the data when split into the two disturbance subsites (Table 2.3; SMD: *p-value* < 0.05*;* SLD: *p-value* < 0.05). This pattern was also found at the Fynbos MD site (Table 2.3: *p-value* < 0.05), where the centre mound vegetation is significantly shorter than the slope and off-mound plant communities. It appears vegetation on heuweltjies grows shorter on the centre of the mound and increases as it moves off. The only exception to this pattern in this study is the Fynbos LD site (Table 2.3*: p-value* $= 0.084$). However, the Fynbos LD site was unique in that there were small trees (*Searsia undulata*) that grew distinctively on the centre of each heuweltjie (field observations), which would logically increase the average height of the vegetation (which was in general a low-shrub-dominated landscape (see Section 2.3.3 below). Average height of the vegetation between the Fynbos sites were also found to be significantly different to each other *(p-values < 0.05),* with the LD site having higher plants on average.

Table 2.3: Means and SD of the vegetation data grouped by heuweltjie zone across the Fynbos and Succulent Karoo biomes for two disturbance levels, to ascertain any significant changes in plant diversity and vegetation characteristics across zones. "n"= number of quadrats. Significance between the zones compared using Kruskal–Wallis chi-squared tests and followed up with pairwise comparisons using Wilcoxon rank sum tests. (Kruskal-Wallis: p < 0.05; p < 0.01**; p < 0.001***; Wilcoxon test: different letters show groups that are significantly different from each other (p < 0.05)). The last colomn shows any significant differences between the sub-sites when the total dataset for each site is compared using Welch Two Sample t-tests, for pooled averages see Appendix 17.*

Subsite	More disturbed (MD)				Less disturbed (LD)				Total site
Zone	Centre	Slope	Off	p-value	Centre	Slope	Off	p-value	p-value
Fynbos (F)	(n=12)	(n=12)	(n=12)		(n=12)	(n=12)	(n=12)		
Vegetation cover (%)	68.2±19.7a	71.1±10.6a	63.8±15.9a		85.8±16.2a	71.8±16.8a	75.9±14.5a		**
Average height (cm)	29.0±10.1b	40.8±12.1a	44.7±22.5a	*	61.7±29.6a	41.1±20.1a	48.9±14.2a		*
Abundance (m^2)	24.3±14.8a	17.7±14.8a	29.9±25.3a		19.6±13.6a	27.5±14.3a	19.0±18.0a		
Species richness (m^2)	2.73±1.0a	2.9±1.1a	3.3±0.9a		3.1±1.4a	3.8±1.0a	2.9±0.7a		
Shannon (H) diversity	1.9±0.3a	1.9±0.4a	1.7±0.4a		1.9±0.5a	1.9±0.3a	1.9±0.3a		
Simpson (D) diversity)	0.8±0.1a	0.8±0.1a	0.7±0.2a		0.8±0.1a	0.8±0.1a	0.8±0.1a		
Evenness (J)	0.8±0.1a	0.8±0.1a	0.7±0.2a		0.8±0.1a	0.7±0.1a	0.8±0.1a		
Succulent Karoo (S)	(n=12)	(n=12)	(n=12)		(n=21)	(n=21)	(n=21)		
Vegetation cover (%)	30.9±8.9a	44.1±16.5ab	49.0±15.5b	**	20.7±8.1a	31.7±10.9b	53.1±13.6c	***	
Average height (cm)	7.3±1.9a	8.9±3.2ab	9.8±2.4b	*	8.7±5.1b	10.3±2.9a	9.8±2.3a	*	
Abundance (m^2)	46.9±20.7a	37.2±21.4a	42.9±11.1a		17.6±9.4a	11.4±5.8b	21.3±12.9a	**	***
Species richness (m^2)	2.1±0.5a	2.1±0.4a	2.2±0.6a		1.6±0.5a	2.1±0.6b	2.8±0.8c	***	
Shannon (H) diversity	0.9±0.4a	1.2±0.2a	1.4±0.4a	p = 0.07	1.3±0.3b	1.7±0.4a	1.8±0.4a	***	***
Simpson (D) diversity	0.5±0.2a	0.6±0.1a	0.6±0.1a	p= 0.067	0.6±0.1b	0.8±0.1a	0.7±0.2a	***	***
Evenness (J)	0.4±0.2b	0.6±0.1a	0.6±0.1a	*	0.7±0.1a	0.8±0.1b	0.7±0.2c	***	***

Box plots for the biomes and subsites, as well as the Kruskal–Wallis tests and pairwise comparisons using Wilcoxon rank sum tests, can be found in the Appendix 3

When looking at the abundance of plants across the Succulent Karoo LD subsite, there were significant differences found in plant abundances across the local gradient of the heuweltjie mound (*p-value < 0.01*), with the slope having a significantly lower abundance compared with the centre or off the heuweltjie. However, the MD site in the Succulent Karoo as well as both Fynbos subsites showed no differences in plant species abundance across the zones *(p-value > 0.05)*. Differences were found between the Succulent Karoo subsites where abundance was significantly higher at the MD site (*p-value < 0.01*).

Species richness was not significantly different across zones in both the Fynbos subsites and the Succulent Karoo MD site (Table 2.3; *p-value > 0.05*), but again species richness was significantly different across all zones in the Succulent Karoo LD site (Table 2.3: *p-value < 0.001*), with lower richness (less species) in the centre, significantly increasing as one moves off the mound.

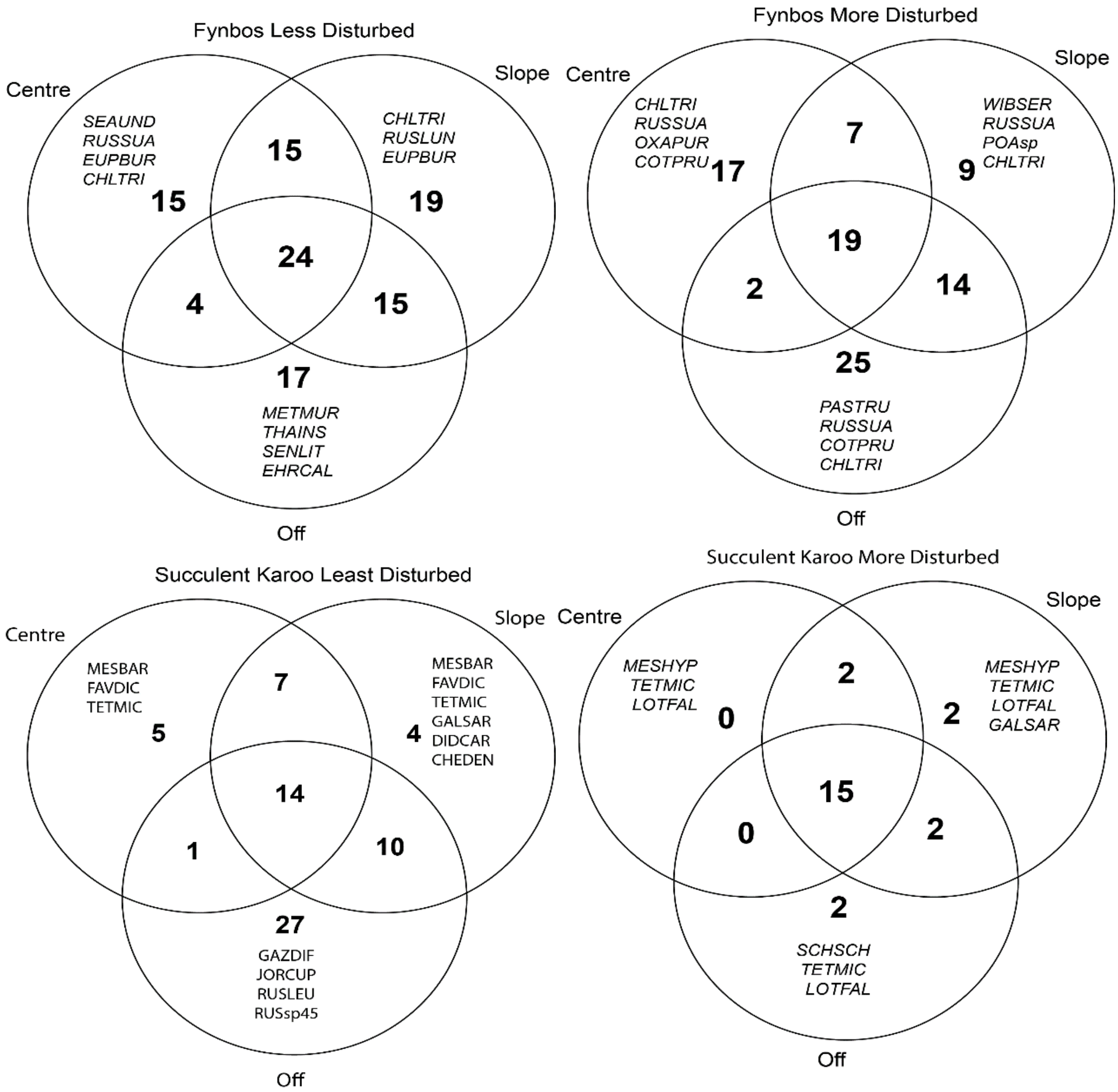
Both diversity indices were found to be significantly different across the zones on the mounds in the Succulent Karoo LD site (Shannon: *p-value* < 0.001; Simpson: *p-value* < 0.001), with diversity again having the pattern of being significantly lower on the centre of the mounds and higher off the mounds. In turn, the Succulent Karoo MD site and both Fynbos sites were not significantly different across mound zones (*p-value* > 0.05) (Table 2.3). The Succulent Karoo MD site was found to have significantly higher Shannon (H) diversity (*p-value* > 0.001) and Simpson (D) diversity (*p-value* > 0.001) , than the LD site.

Lastly, there were no differences found in evenness of the plant communities across the Fynbos heuweltjie zones (Appendix 3 and Table 2.2; *p-value* > 0.05), while the evenness was significantly different amongst the zones of the heuweltjies in the Succulent Karoo (SLD: *p-value* < 0.001; SMD: *p-value* < 0.05), with the centre of the mound hosting significantly lower evenness at both subsites. Evenness was also found to be significantly lower (*p-value* > 0.001) on average at the Succulent Karoo MD site than the LD site.

2.3.3 Plant species unique to the different heuweltjie zone and sites

Each disturbance site and zone within each biome appears to be dominated by a different set of species. SIMPER was run in PRIMER to find which species contributed the most to each disturbance site and zone on average by abundance and cover. Sp25 (*Lotononis falcata*), Sp99 (*Mesembryanthemum hypertrophicum*), and Sp10 (*Tetragonia microptera*) each contribute more than 10% in abundance and cover and Sp42 (*Galenia sarophylla*) contributes more than 10% in cover at the Succulent Karoo MD site, while the LD site is dominated (abundance > 10% and cover > 10% contribution) by Sp5 (*Mesembryanthemum barklyi*), Sp1 (*Faveolina dichotoma*), and Sp10 (*Tetragonia microptera*). At Fynbos, the MD site is dominated (abundance and cover > 10%) by FB (*Chlorophytum triflorum*) and FE (*Ruschia suaveolens*) while the LD site is dominated by FB (*Chlorophytum triflorum*) (abundance > 10%), CH (*Searsia undulata*) and CS (*Metalasia ssp.*) (cover > 10%).

The communities across zones were depicted using Venn Diagrams to show how many plant species are unique to each zone of each site and how many are shared amongst zones, as well as what species are significant in each zone (Figure 2.6), performed using SIMPER. The LD site in the Succulent Karoo was found to share 14 species across all zones, with 5 species unique to the centre, and Sp5 (*Mesembryanthemum barklyi*), Sp1 (*Faveolina dichotoma*), and Sp10 (*Tetragonia microptera*) dominating (abundance and cover > 10%); 4 species are unique to the slope, with this zone being dominated (abundance and cover >10%) by Sp1 (*Faveolina dichotoma*), Sp42 (*Galenia sarophylla*), Sp10 (*Tetragonia microptera*), Sp5 (*Mesembryanthemum barklyi*), Sp41 (*Didelta carnosa*), and Sp8 (*Cheirodopsis denticulata*), contributing >10% to cover only; in turn, 27 species are unique to the off-mound areas, with Sp44 (*Gazania difusa*) and Sp16 (*Jordaaniella cuprea*) contributing more than 10% abundance and cover each and Sp15 (*Ruschia lecosperma*) and Sp45 (*Ruschia ssp.*) contributing more than 10% cover. The remaining 18 species found were shared between a combination of two zones. At the MD site in Succulent Karoo, 15 species are shared across all three zones, with no unique



Key	
Code	***Species name (Field code)***
	S. Karoo
FAVDIC	*Faveolina dichotoma (sp1)*
MESBAR	*Mesembryanthemum barklyi (sp5)*
CHEDEN	*Cheirodopsis denticulata (sp8)*
TETMIC	*Tetragonia microptera (sp10)*
RUSLEU	*Ruschia leucosperma (sp15)*
JORCUP	*Jordaaniella cuprea (sp16)*
GALSAR	*Galenia sarophylla (sp42)*
LOTFAL	*Lotononis falcata (sp25)*
SCHSCH	*Schismus schismoides (sp33)*
RUSCEN	*Ruschia centrocapsula? (sp40)*
DIDCAR	*Didelta carnosa (sp41)*
GAZDIF	*Gazania difusa (sp44)*
RUSsp45	*Ruschia sp? (sp45)*
MESHYP	*Mesembryanthemum hypertrophicum (sp99)*
Fynbos	
EUPBUR	*Euphorbia burmanni (AR)*
POAsp	*Poaceae sp (B)*
SEAUND	*Searsia undulata (CH)*
RUSLUN	*Ruschia lunulata (CL)*
THAINS	*Thamnochortus insignis (CT)*
METMUR	*Metalasia muricata (CS)*
SENLIT	*Senecio littoreus (CU)*
OXAPUR	*Oxalis purpurea (FA)*
CHLTRI	*Chlorophytum triflorum (FB)*
RUSSUA	*Ruschia suaveolens (FE)*
WIBSER	*Wiborgia sericea (FH)*
PASTRU	*Passerina truncta ssp. Truncata (FI)*

Figure 2.6: Venn diagrams showing the number of plant species unique to each zone and shared amongst zones for each disturbance subsite in bold, as well as the plant species that cover more than 10% or have a relative abundance of more than 10%. All plant species IDs can be found in Appendix 7.

species occurring in the centre, and none shared between the centre and off-mound areas. The slope of the mounds, off the mound, between the centre and slope, and between the slope and off, all have 2 unique species in each zone, respectively. The centre of the mounds is dominated (abundance > 10%) by Sp99 (*Mesembryanthemum hypertrophicum*) and Sp25 (*Lotononis falcata*), which contribute 67% to the abundance of the plant community, as well as by Sp99 (*Mesembryanthemum hypertrophicum*), Sp10 (*Tetragonia microptera*), and Sp25 (*Lotononis falcata*) (cover >10%), which contribute 70% to the cumulative cover of the plant community. Sp25 (*Lotononis falcata*), Sp42 (*Galenia sarophylla*), Sp10 (*Tetragonia microptera*), and Sp99 (*Mesembryanthemum hypertrophicum*) each contribute (abundance and cover > 10%) to the slope communities, while Sp25 (*Lotononis falcata*), Sp10 (*Tetragonia microptera*), and Sp33 (*Poaceae ssp.*) dominate (abundance and cover >10%) the off-mound communities.

The Fynbos LD site shares 24 species across all zones with 15 species occurring uniquely on the centre of the mounds, 10 unique to the slopes, and 17 unique to the off-mound areas, while 15 are shared between the centre and slope, as well as a different 15 shared between slope and off and 4 species shared between the centre and off-mound areas. The centre of the mounds are dominated (cover and abundance >10%) by FB (*Chlorophytum triflorum*), CH (*Searsia undulata*), AR (*Euphorbia burmanni*), and FE (*Ruschia suaveolens*); slope is dominated by FB (*Chlorophytum triflorum*) when looking at cover and abundance, but also by CL (*Ruschia* *lunulata*) and AR (*Euphorbia burmanni*), when looking at the cover of each species (cover>10%); and off is dominated by CS (*Metalasia ssp.*), CU (*Asteraceae ssp.*), and DE (*Poaceae ssp.*) (abundance > 10%), while CT (*Thamnochortus insignis*) and CS (*Metalasia ssp.*) dominate the cover of the land (cover >10%). The MD site shares 19 species across all 3 zones; 17 species are unique to the centre, which is dominated (abundance >10%) by FB (*Chlorophytum triflorum*), FE (*Ruschia suaveolens*), FA (*Oxalis purpurea*) and E (*Cotula pruinosa*) as well as by FE (*Ruschia suaveolens*) and FB (*Chlorophytum triflorum*) (cover>10%); 9 species are unique to the slope of the mound, with the area being dominated (abundance >10%) by FB (*Chlorophytum triflorum*), FE (*Ruschia suaveolens*) and B (*Poaceae ssp.*), while FE (*Ruschia suaveolens*), FB (*Chlorophytum triflorum*) and FH (*Wiborgia ssp.*) contribute the most to cover (cover > 10%); and lastly, 25 plant species are unique to the off-mound areas, FB (*Chlorophytum triflorum*) and FE (*Ruschia suaveolens*) adding the most to the abundance and cover followed by E (*Cotula pruinosa*) (abundance > 10%) and FI (*Passerina truncta ssp. truncata*) (cover > 10%); the remaining 23 species were shared across two zones.

2.3.4 Plant functional type diversity across biomes and zones

2.3.4.1 Plant functional type cover

Plants were classified into functional types (life forms) according to Table 2.1 in Mucina and Rutherford (2006). Figure 2.7 shows the percentage cover of each life form for heuweltjies in each biome in total and split by zone. The heuweltjies in the Succulent Karoo are dominated (highest % cover) by herbs (19.01 %) and succulent herbs (19.30 %), with the centre of the mounds being dominated by the succulent herbs (25.95 %), the slope dominated by herbs (20.26 %), and off-mound areas dominated by succulent shrubs (31.69 %). Other notable patterns in the graphs are that the cover of succulent shrubs, herbs, graminoid, and geophytic herbs increases from the centre moving to off the mound, while Succulent herbs decreases from the highest cover on the centre to the lowest cover off the mound. Approximately half of the plants are succulent across all the zones, with succulence also apparently increasing from low on the centre to high off the mounds. Low shrubs are the most dominant life form that can be found across the heuweltjies in the Fynbos; Succulent shrubs (35.5 %) occur in the highest cover on the centre of the mounds, whereas the heuweltjie slope and off-mound areas are dominated by low shrubs (slope: 42.75 %; off: 47.92 %). Other notable patterns in life-form cover at Fynbos are small trees, which have a much higher cover on the centre (26.88 %) of the mound than off and on the slope. Succulent shrubs, herbs, and graminoid- restios decrease in cover along the gradient from the centre to off. Succulence also decreases from a high on the centre of the mounds to a low off the mounds. The cover of succulent shrubs, herbs, and succulence increase in opposite directions in these two biomes.

Figure 2.8 shows the plant functional cover, with the zones split by subsite. Succulent Karoo centre mound communities are still dominated by succulent herbs at both sites, slope is still dominated by herbs at the more disturbed site and succulent shrubs at the less disturbed site, off mound is dominated by the same plant forms as the slope. In the Fynbos, trees have the highest cover on mound centres of the less disturbed site, but herbs dominate the more disturbed site. Low shrubs still dominate the slope and off mound areas of the less disturbed site, but herbs again dominate the slope of the more disturbed site and herbs, and low shrubs can be found to dominate in equal proportion off the mound.

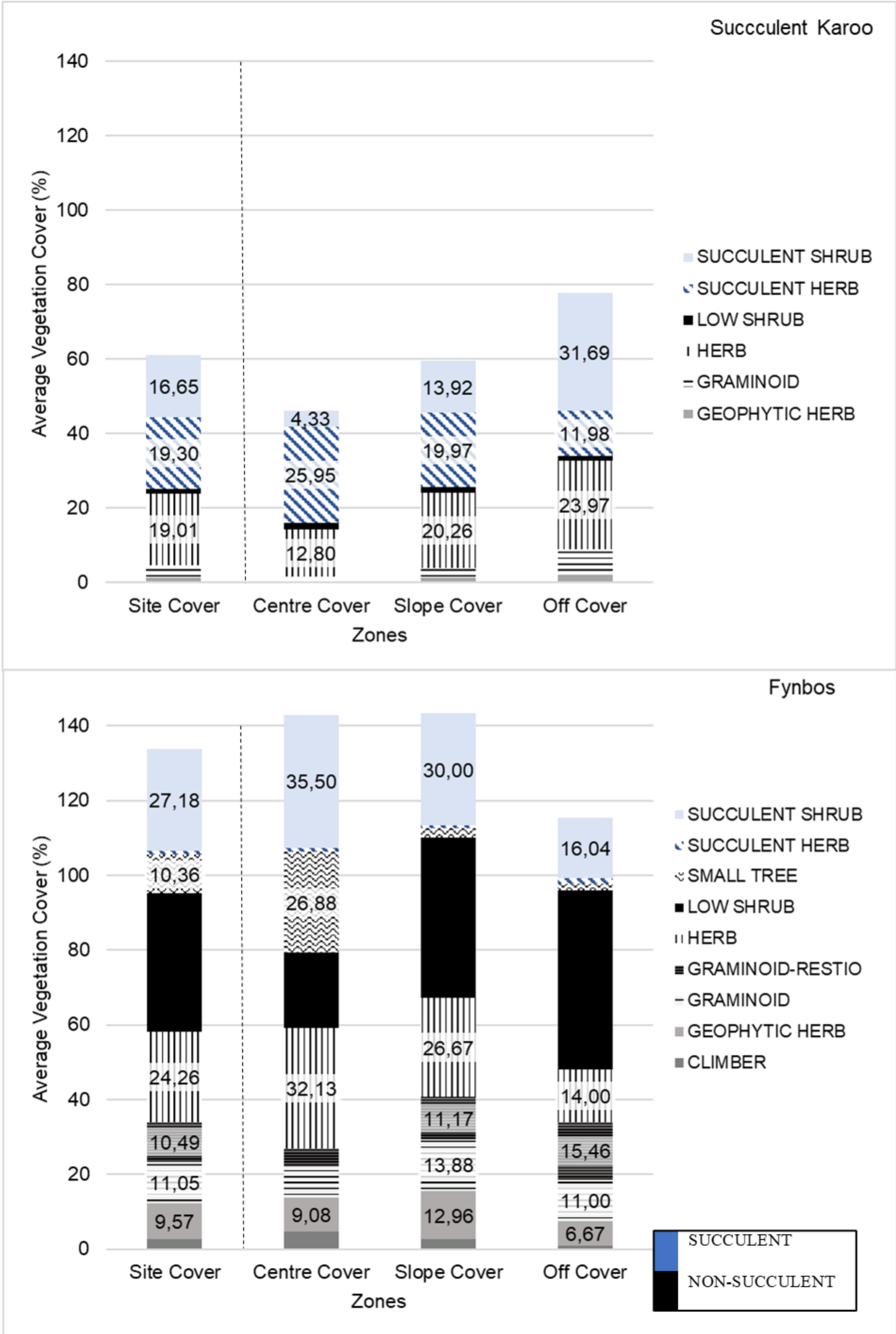


Figure 2.7: Vegetation cover of plants by plant functional type (life form) for each biome and split by zone.

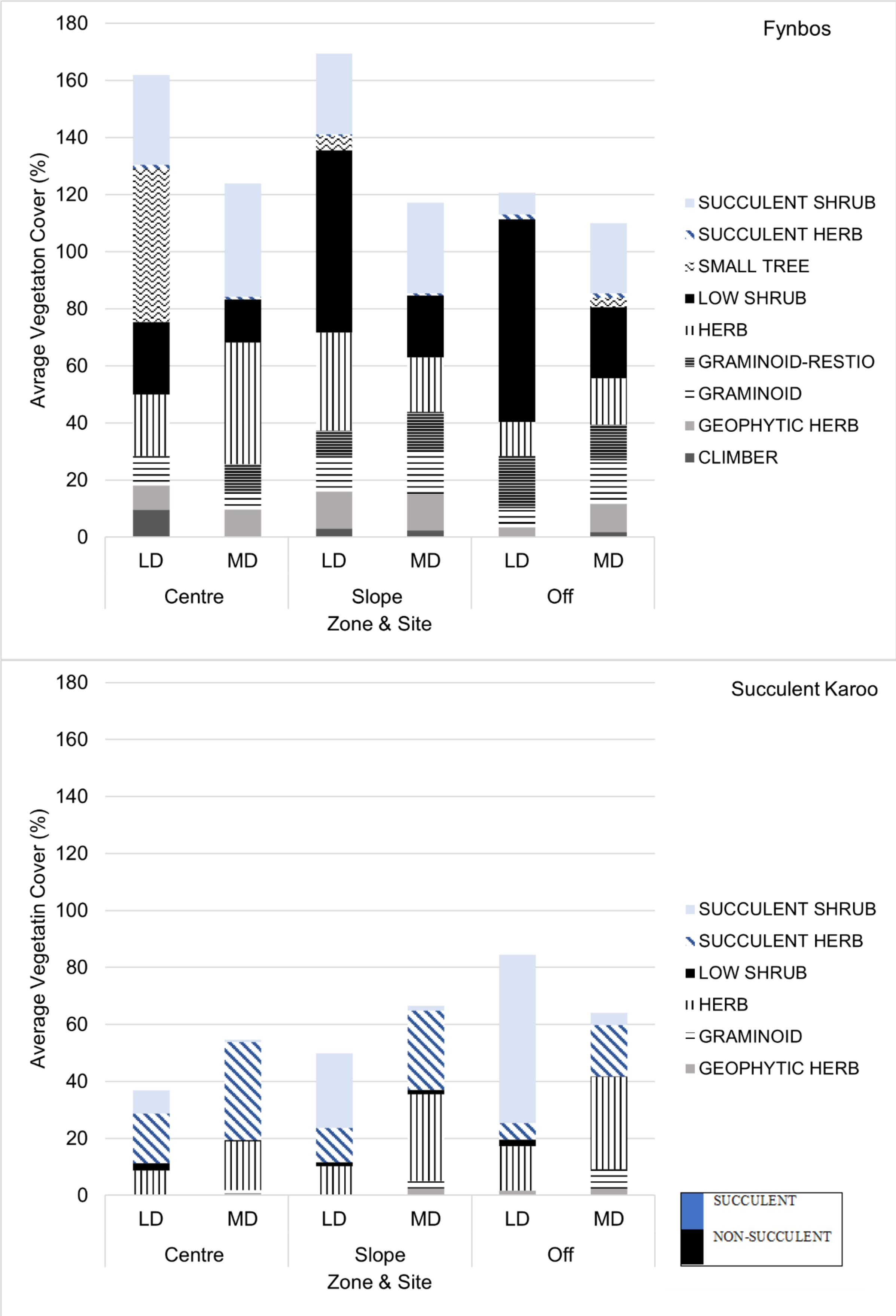


Figure 2.8: Vegetation cover of plants showing average cover per plant functional type (life form) for each biome. Data is split by zone and by sub-site (LD= Less disturbed; MD= More disturbed).

2.3.4.2 Plant Functional Type Diversity

By Zone

The life-form indices were split according to the zones they were sampled in and compared across this heuweltjie gradient, presented in Table 2.4 below. No differences were found in the life-form composition amongst the zones of the Fynbos heuweltjies. At the Succulent Karoo, there also was no difference in the evenness of the plant life forms across the heuweltjie gradients. However, diversity (Shannon (*p-value* < 0.001) and Simpson (*p-value* < 0.001)) is significantly lower on the centre of the heuweltjies. Life-form richness (*p-value* < 0.001) was also found to be different across the zones, with the centre of the mounds hosting a significantly lower number of life-forms to the off-mound areas.

Table 2.4: Diversity indices were worked out per life-form group, life-form diversity (Shannon and Simpson), life-form richness (number of species per life form), and life-form evenness for each zone at each biome. P < 0.05; p < 0.01**; p < 0.001***. Wilcoxon test: significant differences amongst groups shown using lowercase letters where different letters indicate differences.*

Variables	Centre	Slope	Off	Kruskal-Wallis chi-squared
Fynbos				
Life-form richness	5.4± 1.3	5.6± 0.8	5.3± 0.9	
Life-form Shannon (H) diversity	1.0± 0.4	1.2± 0.3	1.1± 0.2	
Life-form Simpson (D) diversity	0.5± 0.2	0.6± 0.1	0.6± 0.1	
Life-form Pielou's evenness (J)	0.6± 0.2	0.7± 0.1	0.7± 0.1	
Succulent Karoo				
Life-form richness	3.2± 0.8a	3.6± 0.9ab	4.3± 0.8b	***
Life-form Shannon (H) diversity	0.7± 0.2a	0.9± 0.2b	0.9± 0.3b	***
Life-form Simpson (D) diversity	0.4± 0.2a	0.1± 0.1b	0.5± 0.2b	***
Life-form Pielou's evenness (J)	0.6± 0.2	0.7± 0.2	0.6± 0.2	

2.4 Discussion

In the GCFR, heuweltjies are unique ecosystem features known to enhance local biodiversity. To understand the broad influences that heuweltjies have on biodiversity, they need to be studied at various spatial scales. At a regional scale the Succulent Karoo and Fynbos biomes differ in the amount of rainfall they each receive and the presence of absence of fire. Moreover, at a local scale, a gradient exists across the heuweltjie mounds, where the centre, slope, and off-mound areas are treated as distinct zones. Diversity patterns are affected, which suggests a heuweltjie should not be seen as a uniform vegetation feature in the landscape. Lastly, living in the Anthropocene, disturbance, such as livestock grazing, might also affect heuweltjies, and negatively impact local diversity. Therefore, given the above, it is necessary to look at the broad influences that affect heuweltjie diversity patterns spatially, in order to understand the structure and function of heuweltjie vegetation in the GCFR. In this chapter, the vegetation structure and diversity were investigated to see if differences occurred between the biomes, on and off the heuweltjies, and between the disturbance levels, with further investigation into the functional types that made up the plant assemblages, in order to describe heuweltjie plant community structure.

Differences in heuweltjie vegetation community composition and diversity between the Fynbos and Succulent Karoo biomes, on and off the heuweltjie, and between the disturbance levels were shown. Differences between the two biomes were expected, given the exceptional local diversity both the Fynbos and Succulent Karoo biomes are known for. The community composition analysis also showed differences between the disturbance levels and heuweltjie zones, with the latter not as clear, however, compared to the former.

2.4.1 The effect of disturbance on heuweltjie plant communities

By looking at the differences amongst the subsites, anthropogenic disturbance or grazing pressure can be investigated. Disturbance was found to have an influence on the heuweltjie plant communities. The Fynbos vegetation was found to cover more of the ground and to grow taller at the less disturbed site. In turn, between the subsites in the Succulent Karoo, the plant communities were more abundant at the more disturbed site but more diverse (Shannon and Simpson) and even at the less disturbed site. Site evenness was investigated using Pielou's evenness and rank abundance curves (see Appendix 1). Looking at the evenness at each subsite revealed an interesting pattern. The subsites allowed us to assess the impact of disturbance. There was no significant difference found in evenness between the two plant communities in the Fynbos; however, the evenness between the two subsites in the Succulent Karoo was found to be significantly different, with the more disturbed site having a much lower evenness than the less disturbed site.

The results indicate that the evenness of a plant community is lower where there is greater human disturbance. Low evenness is commonly associated with disturbed sites (Blanchard and Holmes, 2008; Gaertner et al., 2012; Sagar et al., 2003). Many rare plants need highly specific conditions to survive, and disturbances create a more uniform environment where a few ecosystem generalists will thrive, but the less abundant ecosystem specialists will be lost. These results stand out, as the more disturbed site at the Succulent Karoo was located near the town of Buffelsrivier and was observed to be the site with the highest impact of human disturbance (overgrazing, litter pollution, and erosion due to roads) of all the sampling sites, while the Succulent Karoo less disturbed site was possibly in the most pristine state due to its location close to the Namaqualand National Park, 20 km from the closest town. That the two sites located on a farm in the Fynbos did not show any difference in evenness might indicate that the more disturbed Fynbos site had not yet been hugely affected by disturbance, despite there being evidence of grazing by cattle. The Fynbos sites were located on the same farm and were observed to be less impacted by human disturbance than at the Succulent Karoo more disturbed site. However, data on the stocking rates between the two biomes was not collected, which could also explain this difference in impact.

For the Succulent Karoo heuweltjies, abundance and diversity were found to have the same pattern as evenness. Abundance of plants was found to be significantly higher and diversity significantly lower at the more disturbed site. The major anthropogenic disturbance in this case is overgrazing, where plants get removed and trampled by livestock without enough time for many species to recover. Schmiedel et al. (2016) studied the effect of grazing in heuweltjieveld, finding that grazing results in plant communities consisting of species with a high turnover that thrive in disturbed environments, while long-term perennial species get removed. Rahlao et al. (2008) conducted a study to understand the effect of grazing on heuweltjies by comparing plant communities on and off heuweltjies after 67 years of rest from grazing. It was found that off heuweltjies the plant communities recover faster than the plant communities at the centre of the mound. More significantly, the plant forms change, with disturbed sites being dominated by stem-succulents and deciduous shrubs while recovered sites host evergreen shrubs and trees (Rahlao et al., 2008). Between the sites in this current study, the Fynbos least disturbed site was the only site with trees found growing on the centre on the heuweltjie mounds. However, Schmiedel et al. (2016) found no significant changes in plant forms with grazing pressure. Nonetheless, their sites had only been excluded from grazing for eight years, and they also found that once disturbed, the centre of a heuweltjie takes the longest time to recover to its pre-disturbed state (Schmiedel et al., 2016). Lastly, Kunz et al. (2012) also studied the effect of grazing on Succulent Karoo heuweltjies and found that plant communities showed a general loss in perennial species and a gain in ephemeral species with increased grazing pressure (Kunz et al., 2012).

Disturbance was found to change the dominant plant form on each zone of the heuweltjies in this study. Succulent herbs dominated both sub-sites in the Succulent Karoo centre mound communities; and although this study did not specifically distinguish the succulent plants by stem or leaf succulents, stem succulent herbs (e.g., Sp 99 (*Mesembryanthemum hypertrophicum*)) dominated the more disturbed site, while leaf succulent herbs dominated the less disturbed sites, which corroborates with the findings of Rahlao et al. (2008). Herbs dominated the slope at the more disturbed site, but succulent shrubs dominated at the less disturbed site, while the off-mound plant communities at each site were dominated by the same plant forms as the slope. In turn at the Fynbos less disturbed site, trees dominated the mound centres, but herbs dominated the more disturbed mound centres. Low shrubs dominated the slope and off-mound areas of the less disturbed site, whereas herbs again dominated the slope of the more disturbed site and lastly herbs, and low shrubs were found to dominate in equal proportion off the mound.

Ecosystem interactions are complex, and many different components have to be maintained in a delicate balance to keep them functioning. For example, heuweltjies centres are natural hotspots of disturbance but it appears that when anthropogenic disturbance is introduced the heuweltjie ecosystems might get pushed over a threshold that changes the types of plants found in the communities, which is then difficult to recover from. Added to this, arid ecosystems are known to recover very slowly from disturbance (Rahlao et al., 2008; Schmiedel et al., 2016). The results suggest that where possible, heuweltjieveld needs to be protected from overgrazing, as especially in the Succulent Karoo this veld type takes a long time to recover.

2.4.2 Plant communities across heuweltjie mounds

Plant diversity indices in the Succulent Karoo were found to be different across zones using both univariate and multivariate methods. The centre of the heuweltjies across both Succulent Karoo sites hosts plant communities with lower diversity (Shannon (H) and Simpson (D)), community evenness (J), average vegetation height, vegetation cover, life-form richness, and life-form diversity (Shannon and Simpson). However, the number of individuals (abundance) and number of species (species richness) were found to have significant patterns across the zones only at the less disturbed site, with the number of individual plants being lower on the slope than the centre or off mound areas, and the number of species following a common pattern- increasing from the centre to off the mound. The Fynbos site appears to be uniform across zones with no differences found amongst the zones for the species diversity or life-form indices.

Values of the Simpson's diversity index range from 0 to 1, where 1 means complete diversity while 0 means no diversity. The Shannon diversity index is based on abundance, where values close to 0 represent communities that are dominated by only one species and the greater the value, the greater the diversity. The Fynbos Shannon diversity ranges across the zones from a low of 1.73 ± 0.40 (Fynbos MD) off mound, to a high of 1.99 ± 0.31 (Fynbos LD) off mound. Simpson diversity indices of the Fynbos also had its lowest diversity off mound at the more disturbed site and highest off mound at the less disturbed site, with values of 0.71 ± 0.15 and 0.81 ± 0.07, respectively. However, there were no significant statistical differences amongst the diversity indices of the Fynbos mounds. In the Succulent Karoo, Shannon indices were lowest on the centre of the more disturbed site 0.97 ± 0.40 and highest on the off-mound areas of the less disturbed 1.75 ± 0.44. This is comparable to values found by Schmiedel et al. (2016) from Succulent Karoo heuweltjies, where centre mounds have Shannon indexes of 1.29 ± 0.089 while the off mound/matrix areas have a diversity of 1.7 ± 0.099. The Simpson diversity in this study on Succulent Karoo heuweltjies was found to range from 0.45 ± 0.21 at the more disturbed sites centre to 0.76 ± 0.10 on the less disturbed sites slope. Schmiedel et al. (2016) calculated Simpsons index values of 0.581 ± 0.033 for centre heuweltjie areas and 0.686 ± 0.037 for the matrix/off-mound areas.

Results indicate that plant communities in heuweltjie centres in the Succulent Karoo are unique, with possibly stunted plant growth and lower diversity. The stunted growth could also be seen in the Fynbos more disturbed centre mound communities. Distinct heuweltjie plant communities on the centres of mounds have been found in previous studies (Knight et al., 1989; Kunz et al., 2012; Luther-Mosebach et al., 2012; Schmiedel et al., 2016). There are multiple possible explanations; overgrazing of the sites might have impacted the centre plant communities and they are slow to recover (Rahlao et al., 2008; Schmiedel et al., 2016), or it could be due to the heuweltjie soils. It is known that the soil environment changes across the heuweltjie mound (Booi, 2011; Kunz et al., 2012; Midgley and Musil, 1990; Schmiedel et al., 2016; Vermooten, 2019). High salinity found in heuweltjie soils would only allow salt-adapted plants to grow, as saline conditions and salt stress have been found to restrict plant growth (Zhu, 2007). Another possibility is because of the high ions in the soils, many animals (wild and livestock) could use the centres as a salt lick, which would increase traffic on heuweltjie centres and cause many plants to be trampled/grazed on more frequently. In fact, Kunz et al. (2012) found that sheep graze preferentially on heuweltjies. Lastly, heuweltjie centres naturally sustain more disturbance, with the Succulent Karoo heuweltjies especially showing significantly higher amounts of surface disturbances ("diggings" or medium to large pits on the soil surface were classified as surface disturbances, (see Appendix 4) than the off and slope heuweltjie surfaces, which further adds to the theory of animals visiting the centres.

Interestingly, the distance-based similarity matrix (Figure 2.5) showed that the plant communities from the heuweltjie centres in less disturbed areas are generally more similar to the plant communities of the more disturbed sites, highlighting the role of disturbance in changing the plant communities of heuweltjies in general, arguably to plant assemblages more adapted to disturbance. Natural disturbances could be a reason for the lower diversity on the mound centres, as this natural disturbance simplifies the soil environment and creates a space for opportunistic species such as annuals to dominate. This has been suggested before by Luther-Mosebach et al. (2012) and Knight et al. (1989) who found that disturbance was the reason for heuweltjie plant communities on the centres being different, as disturbances on and off mounds created a non-uniform environment, and the mound plants were adapted to disturbed environments. Natural and anthropogenic disturbance can thus drastically change heuweltjie plant diversity in the landscape, especially in the semi-arid Succulent Karoo biome.

Another interesting pattern that emerged in the plant traits was the trends of succulence. In the Fynbos, the percentage cover of succulent plants was found to be highest in the off-mound plant communities while Succulent Karoo plant communities have higher succulence on the centre of the mounds. Succulence is a plant trait that evolved as a response by plants to survive in dry environments with little access to water, but also as an adaption to highly saline conditions (Males, 2017; Ogburn and Edwards, 2010). Drought-avoiding succulence and halophytic succulence most commonly did not evolve together (Ogburn and Edwards, 2010). Drought-tolerant succulent plants have traits that limit the loss of water, such as a thick cuticle, low stomatal opening, and CAM photosynthesis (Borland et al., 2009; Luttge, 2004; Ogburn and Edwards, 2012), while succulence in halophytic plants is thought to have evolved as a strategy that dilutes salt accumulation in the plant cells, as a method to maintain ionic balances (Luttge, 2004; Ogburn and Edwards, 2010). Bladder cells are another strategy used by halophytic plants which presents as succulence as excess salts get placed in glistening epidermal cells. The Aizoaceae family, which many of the plant species in this study belong to, are however considered to be an exception. They were found to have mostly evolved succulence as a method to conserve water, although they have adapted to survive high amounts of salt stress too (Ogburn and Edwards, 2010). Therefore, when examining what role, the succulent plants growing on heuweltjies have, adaptions to salinity and low water availability must considered.

2.4.3 Limitations

The limitations of a study like this are in the number of replicates and the number of sites that were sampled. The patterns found in the vegetation communities on heuweltjies should be considered to be very site specific and only for that time of the year. More heuweltjies and more sites would need to be sampled to obtain more representative data, which could then more reliably be applied to heuweltjies across the entirety of each biome. With an increase in the number of samples, the issues of pseudo-replication in this descriptive study, due to not pooling samples from the same heuweltjie, can be addressed, underlining further that the results above must be interpreted with this issue in mind. Future studies should rather sample one transect of quadrats (centre, slope and off) per heuweltjie and increase the number of heuweltjie sampled. Despite these limitations, the value in a study like this is in the creation of baseline vegetation data, showing the broad influences of disturbance on the plant vegetation composition of heuweltjies and the unique plant flora across a heuweltjie gradient stretching from the centre to off the mound.

2.5 Conclusion

The results show that heuweltjie vegetation composition, diversity, and function are not homogenous at the specific localities studied within the Fynbos and Succulent Karoo biomes. As expected, biome differences were the most important factor explaining plant community patterns among the heuweltjies. Importantly, the next strongest variable was disturbance in the landscape, such as by livestock as the plant communities associated with the different levels of disturbance in the same biome were found to be different, with different dominant plant functional types. Vegetation community differences were lastly explained by the zone on each mound from where samples were taken. These are thus important factors to take into consideration when studying heuweltjie vegetation communities, especially the anthropogenic disturbance since over short distances, and depending on the stocking rates, the diversity patterns might drastically change. Plant functional type differences across the heuweltjie zones and biomes were also evident. Lastly, this study raised questions about the spatial and historical impact of succulence, water availability, soil salinity, and the animals associated with these conditions, on heuweltjie vegetation communities, diversity, and function. The following chapters use the most common plant species by cover, identified in this chapter, as the basis to investigate the ion contributions in the heuweltjie environments that specifically come from plant material.

Chapter 3: Ion distribution in heuweltjie plants

3.1 Introduction

The soil environment is an important part of any ecosystem, where it plays a role in being the intermediatory region between the biotic environment on the surface and the groundwater system below. Heuweltjie soils have been shown in previous studies as being important accumulators of salts for these arid ecosystem soils (van Gend et al., 2021). The process of how these higher salt concentrations accumulate, specifically in the mound soils is an interesting phenomenon still being understood. Evaporation of surface precipitation, deposition of marine blown salts, or fog events which commonly occur on this coastline, are some of the most common explanations for soil salinisation in arid areas (Adams et al., 2004; Gorji et al., 2017; van Gend, 2018; van Gend et al., 2021; Vermooten, 2019). An array of ions dissolved in solution presumably accompany the water that these precipitation events bring. These deposits would be expected to coat the external surface of the soils and all plants evenly, other wind-blown aerosols would do the same. If these common explanations for salinisation, were the only processes occurring in heuweltjies, soil would be expected to be uniformly saline both on and off mounds. Because this is not the case, the question arises as to whether there are additional processes occurring that cause centre mound heuweltjie soils to accumulate ions in the levels which have been found.

The compositions of plant communities have been found to distinctly change with changes in heuweltjie mound zones (Chapter 2). This corroborates with past findings of heuweltjies found at other locations (Desmet, 2007; Knight et al., 1989; Luther-Mosebach et al., 2012; Midgley and Musil, 1990; Schmiedel et al., 2016; Turner, 2003). Distances and sizes of mound zones exist over a relatively small local gradient, so to see such a significant change in plant communities amongst zones, underlying processes must be taking place. The vegetation found growing on heuweltjies consists of an array of different plant forms that should hold an array of different ions in their tissues (Midgley and Musil, 1990). This plant material and salt will eventually get returned to the soil.

The aim of this chapter is to understand what chemical contribution plants are making to heuweltjie mound soils - firstly by identifying and quantifying the surface and tissue salt content of plants on and off the heuweltjies, and second to determine the original source of the salts found on the external surface of plants. The salinity concentrations of internal plant material growing on heuweltjies have been investigated before, however not for these particular sites or plant species and with no separation of internal and external plant salt deposits (Midgley and Musil, 1990). It is also known that many plants produce calcium oxalate crystals in their tissues, so plant species were also screened for the presence of calcium oxalate and included as one of the minerals investigated when looking at how these chemical species are distributed across the mound.

3.2 Methods:

3.2.1 Field Work

3.2.1.1 Sample collection

Sampling was conducted during two sampling seasons: the first, October to December 2020, in late spring (Fynbos)/ early summer (Succulent Karoo) and the second (February-March 2021) in late summer (for both biomes. Sections of stems and leaves were collected from a range of plants per zone, ensuring the source plants remained intact. The samples were collected from the two distinct subsites sites within each biome. The six plant species were the two plants species with the highest cover per zone of the heuweltjie, from each subsite (the chosen species were determined during preliminary analysis for Chapter 2). These samples will be referred to as the collected plant species (See Table 4.3; and Appendix 8). Other non-plant biological samples, such as frass, termites, stick-piles (termites), rodent nests and rodent faeces were also collected.

3.2.1.2 Surface salts wash

Each of the collected plant species were washed in the field to determine what salts occurred on the surface of the plant material. Three 5 cm (visually estimated) long sections of fresh plant material from three different plants of the same species growing in each zone, were collected in the field. Plant material was then immediately washed in 50 ml of milli q water, for 30 seconds in field. Water was collected in test tubes and taken back to the lab for filtering and plant material was collected in brown paper bags. The water was stored in a fridge once brought back from the field and between filtering. The surface washes in 2020 were only conducted at the Fynbos site during the beginning of December. In 2021 the Fynbos washes were completed at the end of February and the Succulent Karoo washes at the beginning of March 2021.

3.2.2 Lab Analysis

In the lab the plant tissues and biological samples that were collected and had not yet been washed were rinsed once with distilled water for 10 seconds and dried in an oven at 60 degrees. The dried material was then milled into a powder and used for the analysis's in Chapter 3 and 4 that follow.

3.2.2.1 Analysis of cations in plant material

The plant tissue samples used for cation analysis were the sample plant samples used in the plant washes (section 3.2.1.2) as well as the other non-biological samples. Milled dried plant material was subsampled and sent for analysis of cations to the Soil, Water and Plant Laboratory based at Elsenberg, Department of Agriculture Western Cape. Analysis followed the Method no. 6.1.1. For Feeds and Plants, set out in Handbook on Feeds and Plant Analysis (ALASA) (Palic et al., 2007). Samples were ashed overnight, then taken up in HCl solution and heated for 30 minutes. Samples were then filtered to make a filtrate and made up to volume. Ion concentrations were then read on the ICP. Cations tested were sodium (%), calcium (%), magnesium (%), phosphorus (%), potassium (%), aluminium (mg/kg), boron (mg/kg), copper (mg/kg), iron (mg/kg), manganese (mg/kg) and zinc (mg/kg)

3.2.2.2 Analysis of Cation and Anions from the external surface of plant material

The water samples that were collected in the field were filtered using nylon (2020 samples) or cellulose acetate (2021 samples) 0.45um filters, within 2 days of collection. Water was stored in the fridge between collection and filtering. It was tested for EC (µS/cm) and submitted for analysis of cations and anions to BIOGRIP Node for Soil and Water Analysis at Stellenbosch University, which is managed at part of Central Analytical Facility (CAF). Analysis was done using a Metrohm 930 Compact IC (Ion Chromatography) Flex.

Blank samples (consisting of just milliq water) and a control (water brought to the field and treated the same as the plant washes but just without the plant material) were included for each sampling batch. These samples were analysed first to evaluate potential contamination. Before injection into the machine wash samples with EC (µS/cm) above 1000 µS/cm, were auto-diluted, and then all samples were filtered again by the machine using a 0.22 µS cellulose inline auto-filtration cell. Cations and anions analysed in this thesis were Ca^{2+}, Cl^{-}, Mg^{2+}, Na^{+} and SO_4^{2-}.

Photos were taken of the plant material for each of the samples and the surface area of each plant sample was determined using Image J computer software.

3.2.2.3 XRD Analysis

Mineral species in the milled plant samples were determined by XRD (X-ray Diffraction) analysis. This was done for only one set of samples per site (i.e., no repeats). This analysis was conducted by at iThemba LABS. X-ray diffractometry enables direct identification of any crystalline material based on its unique crystal structure. Measurements were performed using a multipurpose X-ray diffractometer D8-Advance from Bruker operated in a continuous ϑ-ϑ scan in locked coupled mode with Cu-K_{α} radiation with a step size of 0.034° 2ϑ at a 0.5 sec/step. The samples were then grouped according to classifications (Biome, Zone, Targeted and Plant form) and * were assigned if samples within each category had the mineral present.

3.2.3 Data analysis

Ion concentrations of the ashed plant tissue and the concentrations of external ions from the plant washes were categorised according to the locations or zones where the plants were found growing. Decisions were made to represent the data from each biome separately, but to group the samples collected in the different 1) years and 2) sites because ions would naturally accumulate in and on the plant throughout the year. It was thought to be more important to show salt accumulation from plants across each biome, rather than at specific sites. Kruskal-Wallis tests, followed by post hoc Wilcoxon rank sum test, were run in R to investigate differences between ions found in and on the plant material amongst the zones, growing on the mound and amongst the different plant species. This was done for the ashed plant material as well as the ions washed off the external surface. Boxplots were made to show the differences amongst the ashed plant material (see Appendix 9).

The concentration of the major ions (Na^{+}, Mg^{2+}, Ca^{2+}, SO_4^{2-}) in the surface plant washes concentrations were plotted against Cl^{-}, because of the low reactivity of this ion in solution. Cl^{-} behaves conservatively during brine evolution until halite precipitation, is redissolved quantitatively, and anion exchange is minor (Eugster and Jones, 1979). The ion ratios were compared with literature values for local seawater, fog, rainwater, and aerosols, to investigate what source the external plant salts could originate from. Three plant species (*Mesembryanthemum barklyi* (Sp5), *Tetragonia microptera* (Sp10), and *Mesembryanthemum hypertrophicum* (Sp99)) were removed from this analysis due to the exceptionally high EC (µS/cm) of these washes which gave outlier results. This was suspected to be because they were the only plant species with bladder cells on their surfaces which could have burst and increased the ion concentrations in these washes (see results section below for further support, Figure 3.1). The exceptionally high EC (µS/cm) of the solutions were suspected to interfere with ion concentrations which may have precipitated out of solution and got removed during the filtering process.

Data on concentration of ions in seawater was obtained from Germs (2004) for seawater from Strandfontein, Western Cape, South Africa, Scott (1999) for seawater samples collected from Saldanha, Western Cape, South Africa and from Nordstrom et al. (1979) which is commonly cited for seawater composition. Seawater data in all three literature sources reported Ca^{2+}, Mg^{2+}, Na^{+}, SO_4^{2-} and Cl^- values. When the values from each source were compared, the ratio of the major ions to Cl^- were all found to be similar. Since most of these sources are from unpublished theses, in this particular thesis data from all the sources were averaged to provide a more accurate and reliable value for local seawater concentration. This mean value is plotted on the graphs (Figure 3.2) as "Seawater".

Information on ion concentrations in fog was collected from two different literature sources (Eckardt and Schemenauer, 1998; Olivier, 2002). Olivier (2002) was a study on fog conducted across the western coast of South Africa, they published ion concentrations of two fog water samples, collected from a fog collection screen set up at Cape Columbine, while Eckardt and Schemenauer (1998), reported the fog water chemistry from samples collected in Namibia, obtained with the AES/ASRC fog collector in the Central Namib Desert. These three points are plotted on the graphs (Figure 3.2) separately and called "Fog". Eckardt and Schemenauer (1998) also published ion concentrations of dry depositions that collected on the fog collector and were washed off with deionized water, this data is recoded on the graphs (Figure 3.2) as "Aerosols". Rainwater ion concentrations were found in a study done by Sonderberg and Compton (2007), where multiple rain samples were collected in Olifants River Valley near the town of Citrusdal and from Cape Town. The mean value for rainwater reported and used to plot on the graphs (Figure 3.2) as "Rainwater".

3.3 Results

3.3.1 Ions brought into mounds in the plant material

In both the Fynbos and Succulent Karoo, sodium was significantly higher ($p<0.05$) in plants growing on the centre of heuweltjie mounds than both the plants growing on the slopes and off the mounds (Table 3.1). Calcium in Fynbos plant samples-was significantly-different ($p<0.05$)-when looking at the zones where the plants grow, with the off-mound plants having the highest amount of calcium , centre mound plants having the second highest amount and the lowest amounts in the plants growing on the slope. In the Succulent Karoo, the amount of calcium in the plants growing on the slope was significantly higher ($p<0.05$) than the plants growing off and, on the centre (Table 3.1).

The plants growing on the centre of the mounds in both biomes had significantly different magnesium from the other zones, however there were opposite trends found between the biomes. The Fynbos centre mound plants had much higher amount of magnesium than the other zones, while in the Succulent Karoo the centre mound plants had a much lower amount of magnesium than the plants growing on the slope and off the mounds (Table 3.1). No differences due to location on the mound ($p>0.05$) were found in phosphorous, aluminium, iron, manganese, or zinc concentrations for either biome (Table 3.1). In both biomes, the amount of potassium in the plants growing on the centre of the mounds was significantly higher ($p<0.05$) than in the plants growing off the mound. In the Fynbos, the amount of potassium found between the centre and slope plants was also significantly different ($p<0.05$) (Table 3.1). Slope growing plants in the Fynbos had significantly lower amounts of boron than the plants growing in other zones and in the Succulent Karoo the plants growing off the mound had significantly lower boron than the plants growing on the centre and slopes (Table 3.1). In the Fynbos, copper was found to be significantly higher in plants growing on the centre of mounds compared with plants growing off mound, while in the Succulent Karoo copper was not found to be significant in the plants across the mounds (Table 3.1).

Ion concentrations in the stick-pile and rodent nest (see Table 3.1) can be used to give a first indication of what ions termites and rodents are bringing into mound soils and which animals might be contributing more to each ion input. In the Fynbos, sodium was highest in the stick-pile samples, but highest in the rodent samples for the Succulent Karoo. Calcium and magnesium were also higher in the stick-pile samples for the Fynbos, but the Succulent Karoo samples were very similar. Phosphorous concentrations were similar amongst the groups in both biomes. Potassium, boron, and manganese had a similar pattern to sodium, with the Fynbos stick-piles having the higher concentration while in the S. Karoo, rodent nests samples were higher for those ions. Aluminium was highest in the rodent nest plant samples for both biomes, while copper, iron and zinc were highest in the termite stick-piles for both biomes.

Table 3.1 Means and SD of cation concentrations found in plant species, termite stickpile and rodent nest plant material. Plant species data was grouped and averaged according to the heuweltjie zone where the plant species was found growing. Major ions are in % while trace ions are in mg/kg. Lowercase letters indicate significant differences ($p<0.05$) between the plant species growing in different zones using Kruskal-Wallis tests, followed by post hoc Wilcoxon rank sum tests. $p < 0.05$; $p < 0.01$**; $p < 0.001$***.*

Ions	Biome	Centre (Fynbos: n=32 S Karoo n=31)	Slope (Fynbos: n=32 S Karoo n=32)	Off (Fynbos: n=32 S Karoo n=29)	Significance level (p-value)	Termite stick-pile (Fynbos: n=26 S Karoo n=22)	Rodent nest (Fynbos: n=2 S Karoo n=4)
Sodium (%)	Fynbos	1.11 ± 0.93b	0.73 ± 0.73a	0.62 ± 0.73a	*	0.15 ± 0.19	0.06 ± 0.01
	S. Karoo	16.59 ± 10.85b	4.34 ± 2.98a	3.38 ± 2.43a	***	0.66 ± 0.49	2.23 ± 0.84
Calcium (%)	Fynbos	2.06 ± 0.51a	1.48 ± 1.05b	2.30 ± 0.60c	***	1.20 ± 1.06	0.58 ± 0.13
	S. Karoo	1.58 ± 1.13a	2.78 ± 1.59b	1.88 ± 1.90a	**	3.02 ± 1.21	2.59 ± 0.58
Magnesium (%)	Fynbos	0.89 ± 0.75b	0.49 ± 0.54a	0.38 ± 0.46a	***	0.31 ± 0.33	0.07 ± 0.01
	S. Karoo	0.53 ± 0.17b	1.42 ± 1.49a	0.73 ± 0.25a	***	0.48 ± 0.19	0.52 ± 0.07
Phosphorus (%)	Fynbos	0.06 ± 0.03a	0.05 ± 0.02a	0.05 ± 0.02a		0.03 ± 0.01	0.04 ± 0.01
	S. Karoo	0.09 ± 0.03a	0.12 ± 0.07a	0.11 ± 0.03a		0.09 ± 0.09	0.06 ± 0.01
Potassium (%)	Fynbos	1.34 ± 0.62b	0.92 ± 0.69a	0.89 ± 0.43a	**	0.26 ± 0.18	0.12 ± 0.04
	S. Karoo	1.59 ± 0.58a	1.34 ± 0.47ab	1.21 ± 0.35b	*	0.32 ± 0.16	0.58 ± 0.04
Aluminium (mg/kg)	Fynbos	374.03 ± 773.28a	437.34 ± 524.57a	272.88 ± 234.04a		2285.77 ± 1292.90	2900.00 ± 2969.85
	S. Karoo	1528.97 ± 1827.15a	1066.88 ± 794.58a	953.27 ± 731.01a		3263.64 ± 626.06	4175.00 ± 1244.66
Boron (mg/kg)	Fynbos	32.79 ± 5.73a	23.39 ± 7.73b	139.93 ± 13.39a	***	21.49 ± 11.54	10.64 ± 3.89
	S. Karoo	42.70 ± 23.51a	39.65 ± 19.83a	27.90 ± 10.34b	**	23.67 ± 6.61	27.88 ± 3.40
Copper (mg/kg)	Fynbos	7.04 ± 4.08a	4.94 ± 1.98ab	3.92 ± 1.74b	**	4.54 ± 1.74	3.33 ± 1.25
	S. Karoo	3.76 ± 2.28a	4.10 ± 1.57a	4.75 ± 1.06a		6.13 ± 1.54	5.26 ± 1.03
Iron (mg/kg)	Fynbos	485.9 ± 1212.58a	479.10 ± 692.54a	251.02 ± 201.63a		2444.48 ± 3158.52	1931.25 ± 2018.44
	S. Karoo	1244.62 ± 1389.76a	1060.86 ± 957.85a	873.75 ± 512.56a		3398.27 ± 1671.49	2989.25 ± 680.02
Manganese (mg/kg)	Fynbos	1414.92 ± 1230.95a	1055.57 ± 1090.79a	674.16 ± 608.49a		1024.61 ± 1154.56	223.60 ± 157.26
	S. Karoo	122.48 ± 36.38a	256.27 ± 228.75a	171.91 ± 136.22a		257.52 ± 113.09	324.90 ± 28.68
Zinc (mg/kg)	Fynbos	17.17 ± 8.97a	18.30 ± 9.15a	22.68 ± 6.60a		16.84 ± 7.99	10.00 ± 0.23
	S. Karoo	11.39 ± 3.79a	13.20 ± 5.88a	14.06 ± 4.80a		14.28 ± 4.27	12.82 ± 3.50

Table 3.2 shows that all of the different plant groups and almost all of the plant species within those groups were a source of calcium oxalate in the form of whewellite ($CaC_2O_4{\cdot}H_2O$), except for graminoids in the Fynbos and that it was present in only one out of three of the low shrub plant species. While there was no calcium oxalate in weddellite ($Ca(C_2O_4){\cdot}2H_2O$) form found in the Succulent Karoo plant samples, it did occur in some of the Fynbos plant samples, specifically in the succulent shrubs in the Fynbos, found growing on the centre and slope of the mound. No sodium oxalate ($Na_2C_2O_4$) was found in the Fynbos plant samples but all of the Succulent Karoo samples had sodium oxalate ($Na_2C_2O_4$) present. Pentahydrite ($MgSO_4.5H_2O$), a source of sulphate and magnesium, halite($NaCl$), a source of sodium and chloride and sylvite (KCl) a source of potassium and chloride were found in the plant samples across both biomes in plants growing on all areas of the mounds. Pentahydrite, sylvite and halite were only present in the succulent shrubs in both biomes, and in the succulent herbs from the Succulent Karoo. While halite and sylvite were also found in the geophytic herb sample from the Fynbos.

*Table 3.2: The presence of various minerals in plant samples identified using XRD analysis. * represents if a mineral was found present in the plant species that make up the sample group. Minerals are first grouped by where the plant species was found growing on the mound and secondl by the different types of plant forms with the specific species that form part of the group under. 'n' represents the number of samples that were assessed for mineral presence.*

Biome	Grouping	No. samples	Weddellite $Ca(C_2O_4)\cdot 2H_2O$	Whewellite ($CaC_2O_4\cdot H_2O$)	Sodium oxalate ($Na_2C_2O_4$)	Pentahydrite ($MgSO_4.5H_2O$)	Halite ($NaCl$)	Sylvite (KCl)
By Zone								
Fynbos	Centre	n=4	*	*		*	*	*
	Slope	n=4	*	*		*	*	*
	Off	n=4		*		*	*	*
Succulent Karoo	Centre	n=4		*	*	*	*	*
	Slope	n=4		*	*	*	*	*
	Off	n=4		*	*	*	*	*
By Plant Form	Plant species							
Fynbos								
Geophytic Herb	*Chlorophytum triflorum* (FB)	n=2		*			*	*
Graminoid-Restio	*Thamnochortus insignis* (CT)	n=1						
Low Shrub	*Metalasia muricata* (CS)	n=1						
	Wiborgia sericea (FH)	n=1						
	Passerina truncta ssp. truncata (FI)	n=1		*				
Small Tree	*Searsia undulata* (CH)	n=1		*				
Succulent Shrub	*Ruschia lunulata* (CL)	n=1	*	*		*	*	*
	Ruschia suaveolens (FE)	n=4	*	*		*	*	*
Succulent Karoo								
Herb	*Lotononis falcata* (Sp25)	n=2		*	*			
Succulent Herb	*Tetragonia microptera* (Sp10)	n=4		*	*	*	*	*
	Mesembryanthemum barklyi (Sp5)	n=1		*	*		*	*
	Mesembryanthemum hypertrophicum (Sp99)	n=1		*	*		*	*
Succulent Shrub	*Ruschia leucosperma* (Sp15)	n=1		*	*		*	*
	Jordaaniella cuprea (Sp16)	n=1		*	*	*	*	*
	Ruschia centrocapsula? (Sp40)	n=1		*	*			*
	Cheirodopsis denticulata (Sp8)	n=1		*	*		*	

3.3.2 Ions being brought into mounds on the external surface of the plant material

Plant wash data was investigated for patterns and outliers before it was interpreted. The surface areas of the plant material that created the wash samples, were found to be not correlated to the EC (µS/cm) of the corresponding solutions (r-=0.03; $p>0.05$; Appendix 12). EC (µS/cm) of the washes allocated according to different plant morphological categories (plant shape, leaf shape and surface texture), showed no noteworthy patterns, the only exception being the EC (µS/cm) of the plants, when the plants were grouped into the leaf surface textures (hairy, smooth, waxy or bladder cells). The plants with bladder cells had exceptionally high EC (µS/cm) compared with the rest of the surface types (Figure 3.1). Plants with bladder cells happened to be *Mesembryanthemum barklyi* (Sp5), *Tetragonia microptera* (Sp10), and *Mesembryanthemum hypertrophicum* (Sp99), and the EC (µS/cm) of the washes for just these three plant species were a magnitude higher than the rest of the plant species washed in this project for both biomes. See appendix 18 for Figure 3.1 without the plants with bladder cells. No other patterns existed between the other three groups.

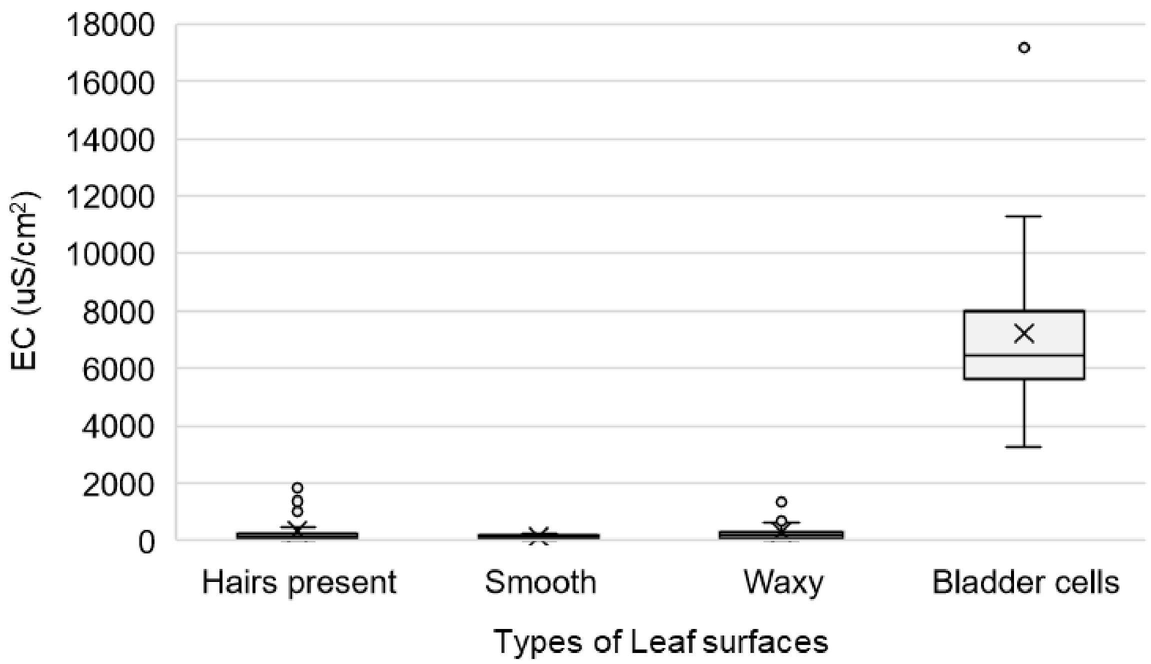


Figure 3.1: EC (µS/cm) of the solutions washed off of plant surfaces, catergorised by the different plant surface types / epidermis.

The plant surface aerosol ratios of Na^+, Mg^{2+}, and SO_4^{2-} with Cl^- showed a strong marine influence (Figure 3.2). Ca^{2+} on the plant surfaces were more enriched compared with the concentrations in seawater. Ca^{2+} from the wash samples followed the trends of fog and is consistent with the enrichment seen in the fog and aerosols from the same region. The Fynbos washes were collected 43 kms from the coast, while the Succulent Karoo washes were 33kms from the coast at the least disturbed site and 55 km from the coast at the more disturbed site.

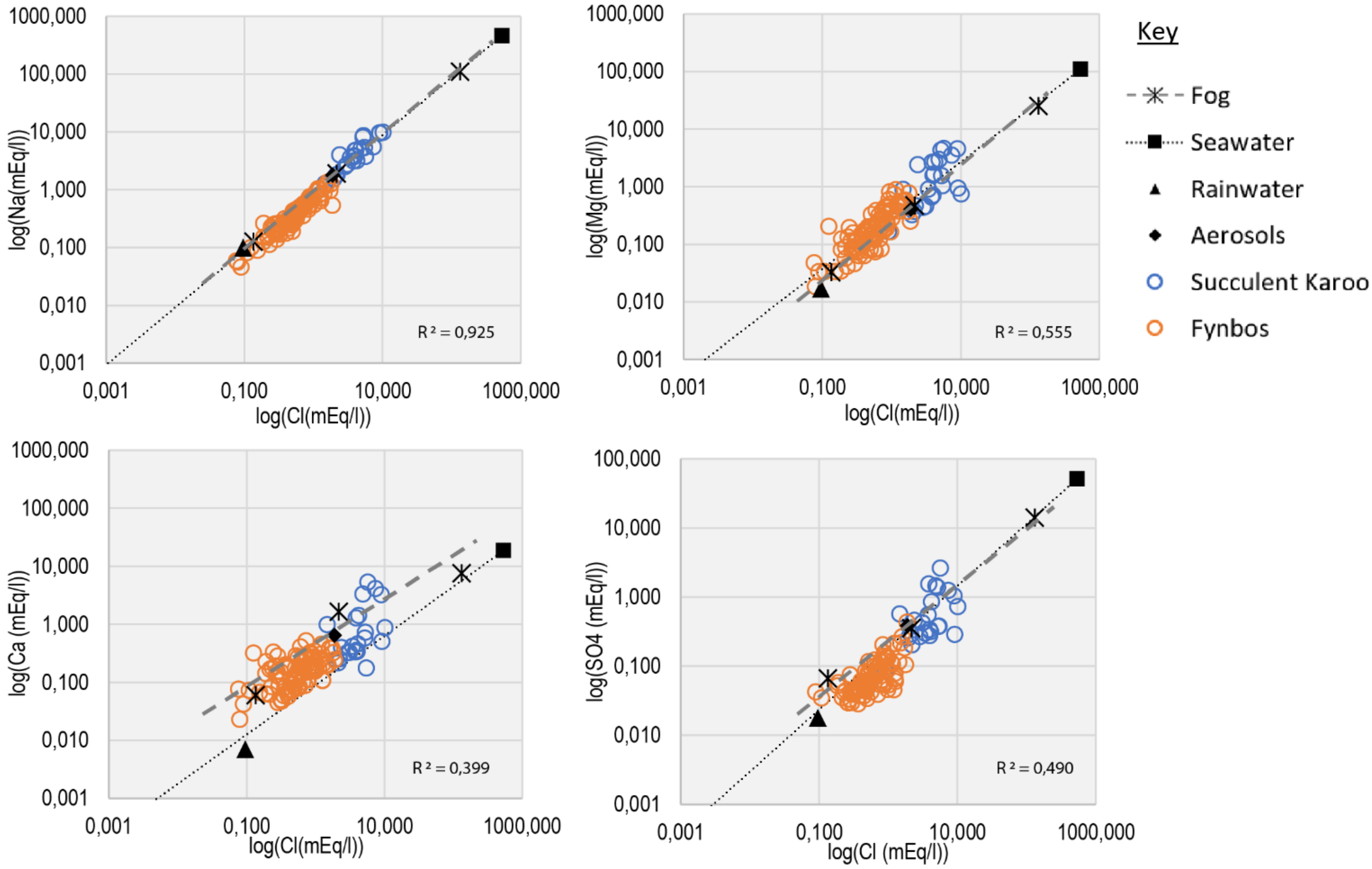


Figure 3.2: Na^{+}, Mg^{2+}, Ca^{2+} and SO_4^{2-} plotted vs Cl^{-} measured from solutions washed off the external surface of various plant species. Values for the concentrations of these ions in Seawater (Germs, 2004; Nordstrom et al., 1979; Scott, 1999), Fog (Eckardt and Schemenauer, 1998; Olivier, 2002), Aerosols (Eckardt and Schemenauer, 1998) and Rainwater (Soderberg and Compton, 2007) were obtained from the literature.

3.4 Discussion

Heuweltjie vegetation communities have been found to differ across heuweltjie mound zones. When the plants die or drop their leaves, they will contribute any plant material to the soil where they grew. This plant material is accompanied by different ions that make up the plant tissues and ions that accumulate on the plant surfaces.

3.4.1 Plant tissues ions

Sodium, magnesium, potassium in both biomes, boron in the Succulent Karoo and copper only in the Fynbos, were significantly higher in the plant tissues of the centre growing plant species. No differences were found in the plant ion concentrations amongst the zones for phosphorous, aluminium, iron, manganese, zinc, in either biome or copper in the Succulent Karoo. Boron in the Fynbos was significantly higher in the plants growing on the slope. In the Fynbos, calcium had significantly different concentrations among the plants growing on all three areas of the mound with off mound plants having the highest average concentrations of calcium while in the Succulent Karoo the slope growing plants had significantly higher amounts of calcium than the plants growing on the other areas of the mound. These results differ from the foliar composition results of Midgely and Musil (1990), where, for all the ions that were tested, plant communities on the mound held higher elemental foliar concentrations than off the mound.

The ion concentrations in the termite stick-piles and rodent nest can give a first indication of the contribution that these two types of foraging mammals are making to the heuweltjie ion pool. In the fynbos termite stick-piles have higher sodium, calcium, magnesium, potassium, boron, copper, iron, manganese, and zinc, while the rodent nests contain relatively higher phosphorous and aluminium than the stick-piles. In the Succulent Karoo the stick-piles contain relatively higher amounts of calcium, phosphorous, copper, iron, and zinc than the rodents' nests which have higher sodium, magnesium, potassium, aluminium, boron, and manganese. The ion concentrations of all of the sample groups (stick-piles, rodent nests and plants grouped by zone), have large variation in their average ion concentrations. This is probably because each group is made of a cumulative number of different plant species. Appendix 9 shows the concentration of ions in each sample. These ion concentrations are only made of a sub-sample of the most common plant species found growing on the mound, so in the actual ecosystems one would probably expect even more variation in the complete plant community ion concentrations.

XRD analysis was used to identify if various minerals were present in this studies' sample of plant species found growing on heuweltjies. This analysis shows that plants are sources of calcium oxalate (weddellite $Ca(C_2O_4)\cdot 2H_2O$) and whewellite ($CaC_2O_4\cdot H_2O$)), sodium oxalate ($Na_2C_2O_4$), pentahydrite ($MgSO_4.5H_20$), sylvite (KCl) and halite (NaCl). Even though this method of analysis does not give quantifiable amounts of each mineral, it does show that the plants do not just hold ions in the plant material but are sources of discrete minerals. Upon abscission of plant leaves, death of the plant or herbivory, these minerals will be directly introduced into the soils through the plant material. The solubility of the mineral then would depend on the soil chemistry of the location the plant material is introduced. Plant species in both biomes were found to be a source of calcium oxalate, with whewellite occurring in plants growing on the mound centres in both biomes Whewellite was also found in all of the different types of plant species except for *Thamnochortus insignis* (CT) a restio, which was the only graminoid sample and two of the three low shrub species, *Metalasia muricata* (CS) and *Wiborgia sericea* (FH), neither of which were central mound plant species. Weddellite was found only in the two succulent shrub species of the Fynbos, which were collected from the centre and slope of the mounds. Sodium oxalate was only found in Succulent Karoo plants, where it occurred in the centre mound growing plants, as well as all of the different plant species. Therefore, it can be concluded that plants species found growing on heuweltjie mound centres are a source of oxalate.

The plant samples that were assessed for the presence of oxalate are made of a small subsample of the plant species from the heuweltjie plant communities. These were the species that had the highest cover and take up the most space, on average, in each area of the mound. They would therefore be big contributors of plant litter to the zones where they were collected from, however no assumptions should be made that these are common patterns across the entire mound plant community. The areas where this study took place were areas of exceptionally high diversity and species richness. A broader range of different plant species would need to be sampled to get a more complete picture of which plant species have the different minerals and in what quantity.

Quantifying the amount of each mineral in the plant species would be especially important for the oxalate minerals, in order to understand the viability of the oxalate-carbonate pathway. Factors to be considered are whether there are only trace amounts of these minerals, or whether there is enough oxalate in the plant material for these plants to make a viable contribution to the pathway and ultimately calcrete deposition.

3.4.2 Surface washes

The ions which accumulate on the external surface of the plants are another possible input of ions in these systems. Ions that accumulate on the external surface of the plants were sampled and analysed. First, an analysis was done to understand why there might be differences in the wash data amongst the plant species. This was done by looking at the different shapes, sizes, and surfaces areas of the plant species, that might influence the results of the ion concentrations. Surface area of the entire plant sample that was washed was compared to the EC (µS/cm) of the corresponding sample and was found to not be correlated with each other (r-=0.03; $p>0.05$; Appendix 12). This was opposite to what was expected, as logically the more surface area a plant has the more salts it would be expected to collect. However, this was not the case, further investigation showed that surface texture of the plant species, particularly those with bladder cells, influences deposition of ions on the plant surface more than surface area. As a result, the washes of *Mesembryanthemum barklyi* (Sp5), *Tetragonia microptera* (Sp10), and *Mesembryanthemum hypertrophicum* (Sp99), were removed from the following analysis (Figure 3.1).

Figure 3.3: Photos of incoming fog over the landscape of the Fynbos study site, taken by Nicola Vermonti September 2020.

The most common external source of the ions on the plant surfaces, was shown to be from marine aerosols (Figure 3.1). The results showed that the plant washes, fog, rainwater, and aerosol values of Na^+, Mg^{2+} and SO_4^{2-} all maintain the same ratio of original seawater with Cl^- (Figure 3.2). This is consistent with work from the wider coastal region (Francis et al., 2020; Smith and Compton, 2004; Soderberg and Compton, 2007) and shows that marine aerosols are an important driver of salt dynamics in the western coastal ecosystems.

Midgley et al. (2012) studied the formation of the calcrete layer found in heuweltjies in the Fynbos and proposed that most likely source of Ca^{2+} in the calcrete are marine aerosols, other possible sources included long distance aeolian dust, local calcareous formations, or weathering of parent rocks. In this study Ca^{2+} was found enriched on the plant material compared with seawater and followed the same trend as fog. Eckardt and Schemenauer (1998), also found that in their samples Ca^{2+} was enriched relative to seawater, as well as in their fog samples. This was attributed to continental inputs (dust) (Eckardt and Schemenauer, 1998).

3.4.3 Heuweltjie soil chemistry

Centre mound soils of heuweltjies in the Succulent Karoo More Disturbed Site (Buffelsrivier) were found to be dominated by anions "Cl>SO4>>HCO3" and cations "Na>Ca>Mg>K." and can be considered saline (Vermooten, 2019). A study that characterised the soils of the heuweltjies at the Fynbos More Disturbed Site (near Piketberg) found centre mound soils to be enriched in Ca, Na, Cl, Mg and SO_4 (Hattingh, 2021) (submitted for examination). Only the centre mound soils of the Succulent Karoo can be considered saline, but in these soils, it can only be assumed that halophytic plants will be found growing. Halophytic plants are a plant group which have adapted mechanisms to help them survive in environments with high salt stresses. There are a range of different adaptions that these plants have evolved to become tolerant of salt stress. Many plant species manage salts by excluding ion uptake at the root. However, this method fails at high concentrations and other adaptions have to be used. Halophytic plant species use different strategies to survive. These include root turnover and the use of bladder cells - where ions get accumulated and compartmentalised in vacuoles. Succulence is another an adaption used to dilute the higher concentrations of salt taken up in the leaves, as is leaf senescence and there is also the secretion of salts out of the leaf material (Humphries et al., 2011; Males, 2017; Ogburn and Edwards, 2010; Volkmar et al., 1998). The dominance of deciduous plants on heuweltjie mounds centres found by Midgley and Musil (1990), could be explained as an adaption by the plants to the more saline soils of heuweltjie centres, where plants drop their leaves as an adaption to salt stress. This idea is supported by suggestions that that the development of drought deciduous and halophytic plant communities might correspond with heuweltjie mound formation (Milton and Dean, 1990). It is difficult to distinguish if the plants' presence caused the soils to become salty or if it is because of pre-existing conditions of the soil environment that halophytic plants are found in centre mound soils. But even without understanding which came first , it is clear that these plants and soil environments create a feedback loop where salts are taken up into the plant material and returned when the plant dies. These factors would help maintain heuweltjie centres as hotspots of high salinity.

3.4.4 Limitations

One limitation found in this study was how plant surfaces interact with salts. Bladder cells are external epidermis cells which function as stores for salt. It is one of the adaptions that some halophytic plant species use to survive in highly saline environments. There is a possibility that these bladder cells burst during the washing and released their salt stores into the wash, resulting in the exceptionally high EC (µS/cm) of *Mesembryanthemum barklyi* (Sp5), *Tetragonia microptera* (Sp10), and *Mesembryanthemum hypertrophicum* (Sp99) samples, as seen in the Figure 3.1. In fact, these were also the only species sampled from the centre of mounds in the Succulent Karoo, which provided evidence for the assumption made above of only halophytic plants growing in these soils. Other adaptions that some plants use is to exude salts from their surfaces, which would contribute additional, non-external salts to the plant washes. Not enough information exists on all of the differ plant species that were washed to check if any of them had this particular adaption, but since no salt crystals were observed on the plant surfaces during field work and as most of the plants had a thick waxy cuticle as their external surface coating, it could be assumed that the plant species do just act as a surface onto which salts to accumulate.

Plant samples are made of a small sub-sample of the plant species that can be found in heuweltjie communities, however the sample species were chosen because they are the species that take up the most space on average, in each area and would therefore be big contributors of plant litter and consequently ions, to the areas where they are found. However, we would not recommend assuming that any that these results are common patterns across all plant communities. The areas where this particular study took place were areas of exceptionally high diversity and species richness. A broader range of different plant species would need to be sampled to get a more complete picture of what mineral and salt contribution the plant communities are making. For future projects it would also be interesting to split the plant species into different plant parts (leaves, stems etc) and see if different areas of the plant have different amounts of mineral and ions. Lastly, quantifying the amount of each mineral in the plant material would also be important, especially for the oxalate minerals, in order to understand whether there are only trace amounts of oxalate or whether there is enough oxalate to make a viable contribution to the calcrete- deposition through the oxalate carbonate pathway.

3.5 Conclusion

The possibility that the addition of the external plant salts would be enough to increase salinity of mound soils will be investigated and discussed later. At this point however another component that must be taken into consideration are herbivorous foragers. Many plants and animals associated with heuweltjies can be considered ecosystem engineers, due to their ability to change the environment around them. Rodents and termites living in heuweltjie mound soils forage in the surrounding inter-heuweltjie matrix and bring the plant material back into mound. Chapter 4 will investigate the above sub-sample of common plant species and identify if they were foraged by rodents and termites or not. The higher salts in centre mound soils could enter heuweltjie soils through plant material which is brought into the mound by these termites and rodents, together with the calcium oxalate and any ions deposited on the external surface of the plants.

In the centre mound growing plants sodium, magnesium andpotassium was found to be higher in the plants of both biomes, boron was found to be higher only in the plants of the Succulent Karoo and copper only in the Fynbos plants. Boron was found to be highest in the plants growing on the slopes of the Fynbos/Calcium was highest in the plants from slopes in the Succulent Karoo and lowest in the plants from the slopes of the fynbos. No differences were found in the plant ion concentrations amongst the zones for phosphorous, aluminium, iron, manganese, zinc, in both biomes and copper in the Succulent Karoo. Plants were found to be sources of calcium oxalate (weddellite $Ca(C_2O_4)\cdot 2H_2O$) and whewellite ($CaC_2O_4\cdot H_2O$)), sodium oxalate ($Na_2C_2O_4$), pentahydrite ($MgSO_4.5H_20$), sylvite (KCl) and halite (NaCl). They were also found to collect marine aerosols on their surfaces. Another finding was that centre mound plants possibly form feedback loops for sodium, magnesium, and potassium with the soils, cycling these ions through their leaves and depositing them back into soils when they die, which would help maintain heuweltjie centres as hotspots of higher salinity.

This chapter provides a basis for understanding the potential ion contribution by plants into heuweltjie mounds. After further investigation into rodent and termite foraging has been analysed (Chapter 4), it will be possible to identify and quantify the surface and tissue salt of plant species, targeted and not targeted by termites and rodents (Chapter 5). It is hypothesised that the plants growing on the mound are the primary biological source of salts, but the activities of termites and rodent species living in the mound are a viable means of increasing salinity in heuweltjie centre mound soils. The results of this chapter contribute to the growing understanding of salinity dynamics on heuweltjies and help complete the picture of the structure of heuweltjie ecosystems in each biome.

Chapter 4: Termite and rodent foraging activities on Heuweltjies

4.1 Introduction

The definition of an ecosystem is a biological community of interacting organisms and their physical environment. In the case of heuweltjies, a simplified ecosystem is made up of the large soil mound, the plants growing on the surface, as well as the animals and insects residing in or passing through the area, which creates the trophic structures of the biological community. Added to that is the abiotic and climatic elements that the mound is subjected to. The presence and activities of termites will have an impact on ecosystems where they are found, specifically the soil and vegetation components (Kunz et al., 2012; McAuliffe et al., 2019a). Bioturbation of soils, and litter collection and decomposition, are common outcomes of most termite species activity (Cheik et al., 2019; McAuliffe et al., 2019b). These actions result in changes to the physical, chemical and hydraulic soil properties of the landscapes they live in (Cheik et al., 2019; Seymour et al., 2014), and as consequence termites are known as ecosystem engineers. Changes in soil properties often coincide with changes in vegetation structure and this appears to be one of the main outcomes of the inter-relationship between *Microhodotermes viator* and heuweltjies in semi-arid South Africa. As seen in the previous chapter, the composition of the plant communities on and off heuweltjies are not the same, which could in part be due to the influence of termites.

This chapter will further investigate the theory that termites are one of the main drivers in the concentration of salts into heuweltjie soils due to their foraging activities (Francis and Poch, 2019; McAuliffe et al., 2019b), by using isotopic analysis techniques to link plant species growing on mounds to the diets of foraging animals that live in the mound. Termites (*M. viator*) and rodent species (most likely a type of mole-rat (*Cryptomys sp*)), which forage for plant material and concentrate the plant material into their nests located in central mound soils, are the species that were considered in this study (Cox et al., 1987; Davies and Jarvis, 1986; Helme, 1990; Lovegrove and Siegfried, 1986; McAuliffe et al., 2019b). It is not known whether termites target preferred plant species as a food source, whether they gather litter that has collected on the ground, or whether they forage opportunistically for the first plant species they come across. Related questions, which will be investigated in Chapter 5, are whether they target any specific species for foraging, and whether these species have significantly elevated ion and salt levels?

4.2 Methods:

4.2.1 Field Work

4.2.1.1 Termite behavioural study

An observational study was done to assess the foraging activities of *Microhodotermes viator,* at the same time as the vegetation study in the previous chapter. Each time a termite colony was seen foraging, the termites were observed, and plant parts being carried in were collected and identified where possible. Only large groups of termites out foraging were chosen for observational studies. This was done on three different heuweltjies in the Fynbos and only one heuweltjie in the Succulent Karoo. Termite colonies are territorial to their mound (Dean, 1993; McAuliffe et al., 2019a), so by observing colonies on different mounds we ensured that there was no pseudo-replication. This study presented many challenges as the termite species forage on the surface, randomly, for a limited time per day (Dean, 1993), so it was impossible to predict when foraging would occur and consequently, fewer observations were conducted than planned.

The aim was to observe which plant species were favoured and which plant parts were favoured. Two observers spent two minutes at each foraging hole collecting all the plant parts being carried by the termites that came within 10 cm of the hole. A minimum of two entrance/foraging holes were chosen, and, where possible, four were chosen. Plant parts carried by the termites during a foraging event, that could be recognised as a particular species, were noted. Foraging Event 1, 2 and 3 were all based on different Fynbos heuweltjies, and collections were made from four different foraging holes on each heuweltjie while Foraging Event 4 was collected at a heuweltjie in the Succulent Karoo from only 2 foraging holes. The termites saw the observers as predators and were easily frightened and would disappear quickly when approached. By moving slowly and keeping collection to 2 minutes at each foraging holes per person (i.e., 4 minutes of collection in total), foraging was less disrupted and kept as normal as possible.

4.2.1.2 Plant collection

Only the six most common species for each subsite were collected for each study area. These were the same samples used for the ion analysis (Chapter 3). The number of samples submitted for each subsite were (n=4) for each of the plant species from each zone, termite samples (workers collected while foraging on the surface) (n=2 or 4), rodent nests (n=2), rodent faeces (n=1 or 2), stick-piles (4 or 7) and frass (4,5,6 or 8). Variable sample numbers are due to opportunist collection. Samples numbers for the variable samples (i.e., non-plants) can be see below in Table 4.1.

Table 4.1 Number of non plant samples collected for each sample type per site.

	Termites	Stick-pile	Frass	Rodent nest	Rodent Faeces
Fynbos - LD	4	7	4	0	0
Fynbos - MD	2	4	6	2	1
S. Karoo LD	4	7	8	2	2
S. Karoo MD	2	7	5	0	0

4.2.2 Lab analysis

4.2.2.1 Termite Litter Characterisation

Each litter piece that was collected during the observational study was identified and classified according to the type of plant part (i.e., sticks (with woody secondary thickening), stems (no secondary thickening), leaves and flowers or reproductive parts (including seeds and bracts). Each litter piece was tallied in its group and weighed to record the biomass of each plant part collected by the termites. Any remaining plant material was weighed, and an attempt was made to tally the pieces. This proved difficult as the collections mostly consisted of small pieces of broken unidentifiable plant tissues and therefore count data must be interpreted with human error included. A species list of plants utilised by termites was produced, from the observations of the plants that were seen being carried in and collected and climbed upon (See Appendix 16).

4.2.2.2 Isotope analysis

5 g of dried and milled plant material, frass, stick-piles, rodent nests, and rodent faeces (as described in section 3.2) as well as individual termites, was was sub-sampled and submitted for carbon and nitrogen isotope analysis at the Stable Light Isotope Laboratory based in UCT.

Samples were weighed into tin cups to an accuracy of 1 microgram on a Sartorius M2P or Sartorius MSE3.6P-000-DM or Mettler Toledo micro balance. The cups were then squashed to enclose the sample.

The samples were combusted in a Flash 2000 organic elemental analyser and the gases passed to a Thermo Scientific Delta V Plus isotope ratio mass spectrometer (IRMS) via a Conflo IV gas control unit. Data quality of the analysis was evaluated by analysing Acacia and Nast standards in conjunction with the plant-based samples (plant species, stick-piles, frass, rodent nest, and rodent faeces). The termite bodies were evaluated using Choc, Mew Mg and Valine standards. The δ notation was used to report the data where R represents $^{13}C/^{12}C$ or $^{15}N/^{14}N$. (Equation 4)

$$\delta(‰) = \left(\frac{R_{Sample}-R_{Standard}}{R_{Standard}}\right) \times 1000 \text{ (Equation 4)}$$

Samples were run in batches grouped according to the type of sample material. Five batches were run, three for the plant-based samples and two for the termite samples. Standards were run for each batch. Standard deviations for Acacia $\delta^{15}N$ were ± 0.63‰; ± 0.401‰; , and ± 3.729; while Acacia $\delta^{13}C$ standard deviations were ± 0.14‰; ± 0.185‰; and ± 0.21‰.The batch with the large standard deviation for $\delta^{15}N$ consisted of the rodent nest, rodent faeces, and termite frass samples. Nast standard deviations for $\delta^{15}N$ were ± 0.313‰; ± 0.161‰; and ± 0.020 and Nast $\delta^{13}C$ standard deviations are ± 0.096‰, ± 0.156‰, and ± 0.137‰. Standard deviations of the termites $\delta^{15}N$ standards were Choc: ± 0.072‰ and ± 0.12‰; New MG: ± 0.081‰ and ± 0.081‰ and Valine: ± 0.107‰ and ± 0.063‰. standard deviation of termite $\delta^{13}C$ standards were Choc: ± 0.146‰ and ± 0.048‰; New MG: ± 0.204‰ and ± 0.048‰ and Valine: ± 0.083‰ and ± 0.086‰. Standard deviations <0.2 are considered normal.

4.2.3 Statistical analysis

Pearson's correlations were run using excel data analysis package, to compare $\delta^{13}C$ and $\delta^{15}N$ values. ANOVAS were run in r to compare the $\delta^{13}C$ values among different plant samples. ANOVAS were used as the variance was the same according to the levene test for all groups. The ANOVAS were followed with post hoc Tukey HSD tests to investigate which groups were significantely different *($p<0.05$)* from each other. Comparisons were done between each type of sample (plant species, termites, frass, stick-piles, rodent nests, and rodent faeces) with the plant species split by the zone they were collected in.

The collected plant species were classified according to whether or not they were targeted by termites and rodents. This was done by assessing if the plant samples met three conditions. The first condition was whether the species were observed being carried into the mound or seen in a termite stick-pile (observational study table 4.2). The second condition was based on whether the sample had a $\delta^{13}C$ value not significantly different to the termite stick-piles or termites, and the third condition was if there were no significant differences between the rodent nest sample $\delta^{13}C$ values and the plants. If the sample met all three conditions, it was allocated too "targeted". If it only met one or two conditions, then "maybe targeted" and if it met none then "not targeted". This was done to provide greater confidence in producing a list of plants targeted by termites and rodents (i.e., plants being brought into mound soils) and reduced the chances of misclassifications.

4.3 Results

4.3.1 Observational Field study

At each foraging occurrence the termites ventured out from two to seven "foraging holes" to collect plant material. The termites with large pincers would climb onto nearby plants and cut parts off them, other termites would walk on the ground and collect plant material (see Figure 4.1). The choices appeared to be completely random as a wide range of different plant species and plant parts comprised of the plant species closest to the hole were seen being cut and carried (See Figure 4.1, Table 4.2, and Appendix 16). Plants that were too difficult to cut or carry were either abandoned or multiple termites could be seen working together to carry larger sticks. Plant material was mostly live/fresh plant parts. Plant material was carried back to the foraging hole where it was either taken straight underground or deposited on the growing pile of plant material that formed around each hole (stick-pile). It appeared that only pieces which could fit down the holes were chosen, but there were multiple observations of termites making a concerted effort to drag larger sticks into the holes and plant material was also observed being brought back

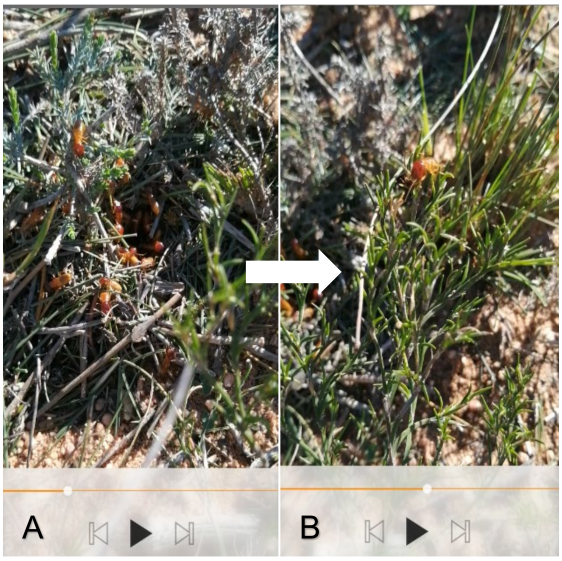


Figure 4.1: Screenshots of a panoramic video taken of a foraging group of termites. In Image A shows the foraging hole, stick-pile and active termites, with the plant from image B in the foreground. Image B shows a nearby plant with a single termite cutting plant material from the top branches. The video was taken in June 2020, At the Fynbos site by Nicola Vermonti

out. Large groups of termites were seen foraging and gathering large quantities of plant material simultaneously into the stick-piles. However smaller groups of termites were also seen foraging and carrying the plant material from the gathered food or stick-piles, a piece at a time into the underground nests, away from the main foraging event. Termites were seen to collect a range of different plant parts, instead of just sticks (Figure 4.2 and 4.3). Leaves were the most common type collected by number and weight with sticks being the second in terms of biomass/weight while the reproductive parts were the second most common by number (Figure 4.2).

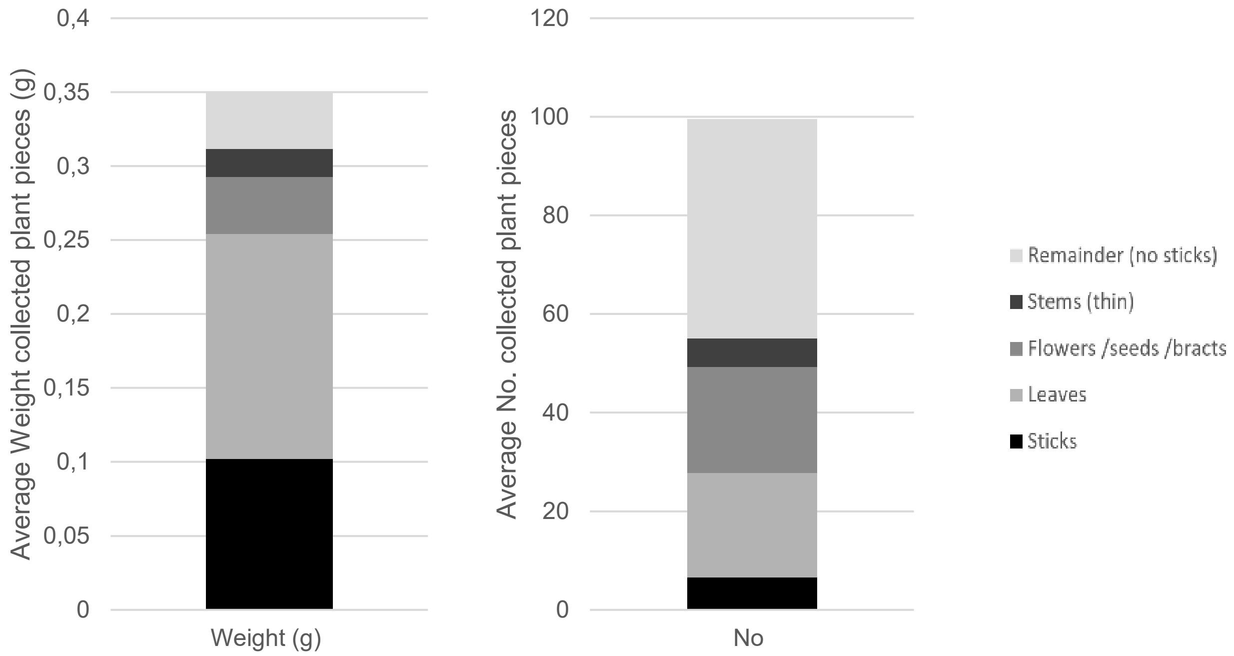


Figure 4.2: Bar graph showing the average number and weight of plants collected from foraging termites in 4 minutes, (n=14). Collected plants were mostly fresh at the time of collection and were assigned a category as to the type of plant part they were. The plants collected from foraging termites were tallied (no.) and weighed.

Figure 4.3: Photos of foraging termites collecting different types of plant parts and plant species. Taken by Nicola Vermonti in 2020 at the Fynbos Site.

Foraging event details with the plant species seen being collected can be found in Table 4.2. At foraging Event 1 the termite activity was mostly seen below an 80 % dead *Ruschia suaveolens* (FE), but no *Ruschia suaveolens* (FE) was seen being collected or cut by these termites. This foraging event was of a large foraging group with around 7 foraging holes being used. From observations the termites were foraging for any plants close to the hole and did not seem to show any preference, with 90 % of termite activity occurring withing 1 m of the hole. The termites at foraging Event 2 were collecting plant material under the shelter of the tree *Searsia undulata* (CH) growing on the heuweltjie centre and were very skittish (i.e., disappeared quickly back into the holes as people approached). Foraging Event 3 was large with 2/3 foraging groups being located under the tree *Searsia undulata* (CH) on the centre but the remaining (3/4) under various bushes growing across the slope of the mound. Foraging Event 4 was the only event recorded in the Succulent Karoo and was just a small group of termites foraging off a mound.

Stick-piles

Despite observing the termites collecting a range of different plant parts, the remaining stick-piles were found distributed on the surface on all areas of the mound (no differences were found in the presence or absence of stick-piles amongst the different zones of the mound; See Appendix 4) and were observed to be dominated in composition by sticks. The stick-piles consisted of many different plant species (see Figure 4.4 below and Appendix 15 for photographic evidence).

Figure 4.4: Photos of various stick-piles found at the Fynbos site. The first two photos were taken during a foraging event with termites collecting and bringing fresh plant material onto the stick-piles and into the mound. Photos taken by Nicola Vermonti in 2020.

Table 4.2: Details of foraging events recorded in the observational study.

Foraging Event no.	Date	Time	GPS	Location	Weather	Recognised species (in Field)	No. holes sampled (n)
1	17/09/20	@2pm-3pm	32° 34' 56.6" S 18° 46' 24.2" E	Fynbos Heuweltjie 2 (PH2) (North -Centre/Slope)	Sunny (19C) (Day before – rain)	Plants seen being carried: • *Cotula pruinosa* (E) • *Chlorophytum triflorum* (FB) • Poaceae Sp, Plant species seen cut/being climbed on • *Cotula pruinosa* (E) • *Oxalis purpurea* (FA) • *Arctotheca calendula* (AA) • Oxalis Sp	4
2	01/10/2020	@4pm	32° 34' 25.2" S 18° 47' 08.4" E	Fynbos Heuweltje 6 (PH6) (North- Slope)	Clouds/Light Rain (13C)	Recognised in stick-pile • *Metalasia muricata* (CS) • *Ruschia lunulata* (CL) • *Thamnochortus insignis* (CT) • *Macrostylis hirta* (DO) • Lanchenalia Sp Plant species seen cut/being climbed on: • *Metalasia muricata* (CS) • *Thamnochortus insignis* (CT) • *Macrostylis hirta* (DO) • *Lapeirousis fabicii* (J)	4
3	02/10/2020	@1pm	32° 34' 24.6" S 18° 47' 05.0" E	Fynbos Heuweltjie 4 (PH4) (North Centre/Slope)	Sun behind Clouds (16C)	Plants being carried • *Chlorophytum triflorum* (FB) • *Searsia undulata* (CH) • *Eriocephalus africanus* (CF) • No ID (DF) • No ID (CX) Recognised in stick-pile • Searsia undulata (CH) Plant species seen cut/being climbed on: • *Searsia undulata* (CH) • *Macrostylis hirta* (DO) • No ID (CQ) • Aizoaceae sp (AW) • No ID (CJ) • *Poaceae Sp,*	4
4	17/10/2020	@6:30pm	29° 54' 38.5" S 17° 27' 02.1" E	Succulent Karoo Heuweltjie 7 (KH7) West- Off mound	Sun (29C)	No species recognised during foraging Recognised in stick-pile • *Tetragonia microptera* (Sp10) • *Jordaaniella cuprea* (Sp16)	2
Observations (no collections)							
1	13/10/2020	@5:30	29° 54' 45.4" S 17° 27' 22.0" E	Succulent Karoo @KH1(Q3)-		A few termites seen collecting: • *Ruschia leucosperma* (Sp15) • *Mesembryanthemum deciduum* (Sp46)	1
2	17/10/2020	@5:30	29° 54' 37.6" S 17° 27' 03.5" E	@KH7 (off NE)		• *Oxalis annae* (Sp4) 1 termite collected with an Oxalis annae (Sp4) stem Termites retreated to the hole very quickly and closed it up	1
3	17/10/2020	@6:00	29° 54' 36.5" S 17° 27' 03.0" E	@KH5 (Off SE)		• *Gazania difusa* (Sp44)	1

4.3.2 Isotopes

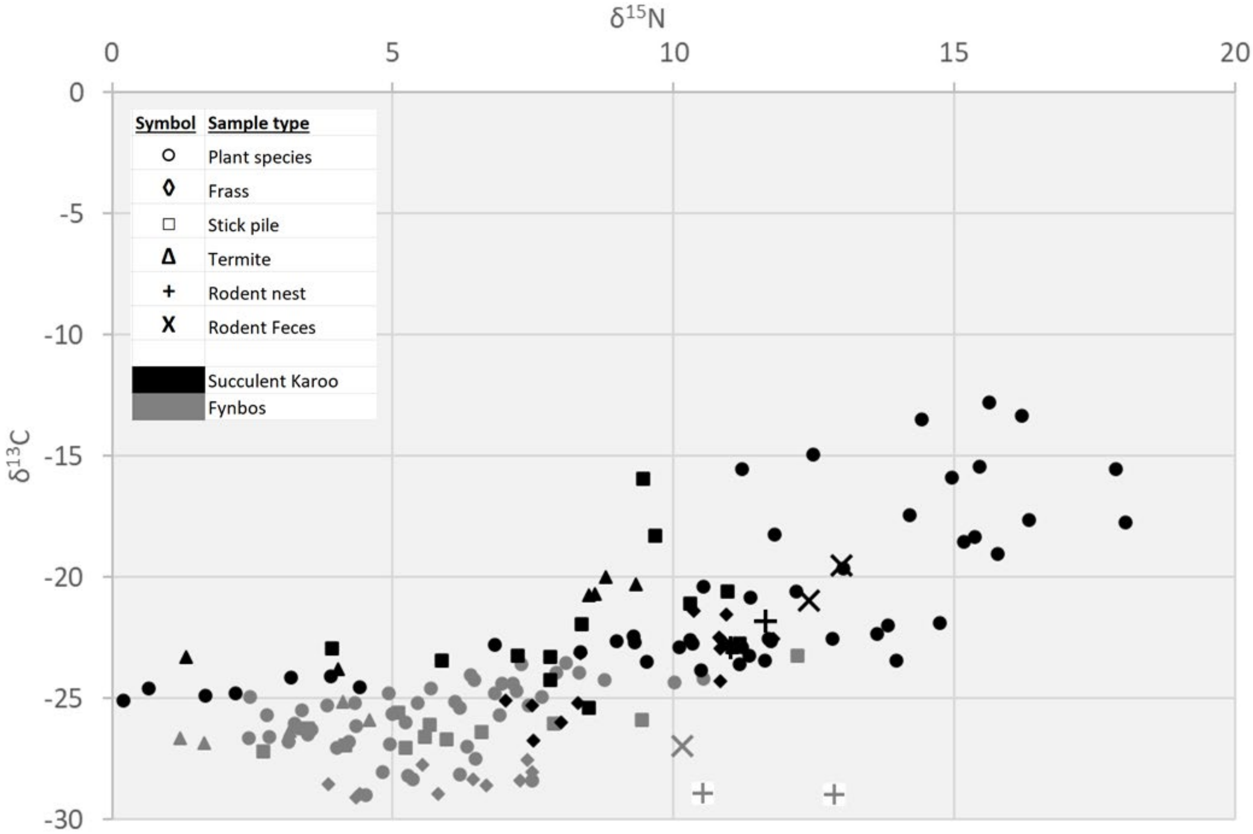


Figure 4.5: $\delta^{13}C$ compared with $\delta^{15}N$ ratios for biological samples collected on heuweltjies at sites in two biomes. The Fynbos site in grey and the Succulent Karoo samples in black.

The Fynbos sites (grey) were found to have lower $\delta^{13}C$ and $\delta^{15}N$ values than the Succulent Karoo (black) sites. When looking at the data as a whole the data was found to be strongly positively correlated ($r=0.73$). The Fynbos data on its own is weakly positively correlated ($r=0.18$) while the Succulent Karoo data shows strong positive correlation ($r=0.66$). ANOVAS were run to compare the $\delta^{13}C$ values for each of the groups. Significant differences were found between the $\delta^{13}C$ ($F=59.15$; $p<0.001$) values for the sample groups. When grouped and averaged by the type of sample and zone collected from (Figure 4.6), $\delta^{13}C$ shows very little variation (standard deviation) in the values of each sample, while there is considerable variation seen in the $\delta^{15}N$ values (Figure 4.6). All the sites were found to have significantly different $\delta^{13}C$ ($p<0.001$) amongst the sample groups.

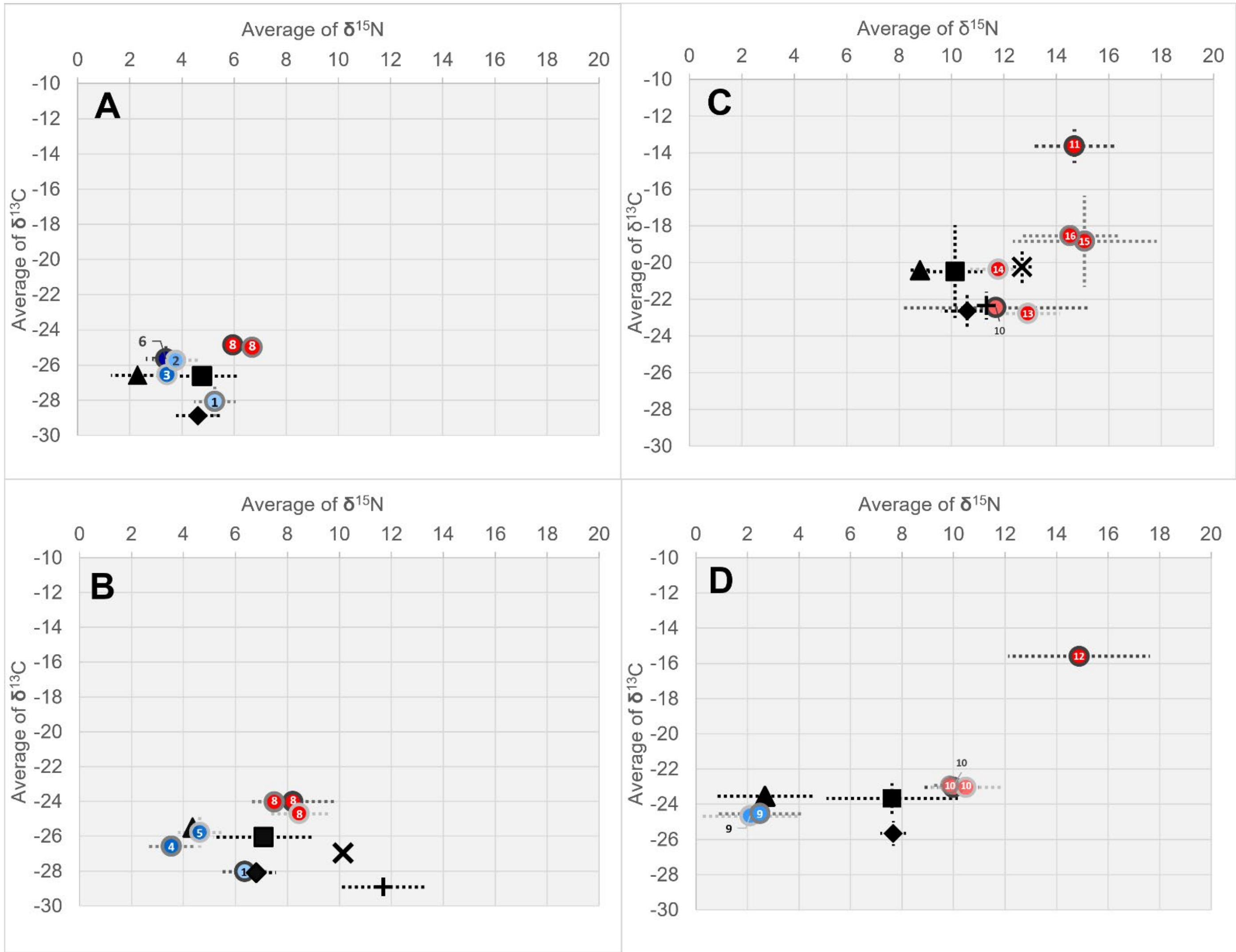
Within the Fynbos LD site (Figure 4.6A): *Chlorophytum triflorum* (FB-1), a geophytic herb, was found to have a significantly lower $\delta^{13}C$ to almost all other samples except for frass. *Chlorophytum triflorum* (FB-1) was the only geophytic herb, that was sampled at this site. *Ruschia lunulata* (CL-7) and *Ruschia suaveolens* (FE-8) both succulent shrubs, also had significantly higher $\delta^{13}C$ values than both the stick-piles, termites and *Metalasia muricata* (CS-3) a low shrub. The termite $\delta^{13}C$ and stick-pile $\delta^{13}C$ was not significantly different ($p>0.05$). *Searsia undulata* (CH- 6) a small tree, *Thamnochortus insignis* (CT-2) a restio, or *Metalasia muricata* (CS-3) are the only species not significantly different ($p>0.05$) to the termite and stick-pile $\delta^{13}C$ values at the Fynbos LD site.

At Fynbos MD subsite (Figure 4.6B) the $\delta^{13}C$: *Chlorophytum triflorum* (FB-1) and frass were again both significantly lower than all the samples except for the rodent samples. Of note the $\delta^{13}C$ signature of

Chlorophytum triflorum (FB-1) is not significantly different to the signature of the rodent nest sample. *Ruschia suaveolens (FE-8)* samples collected from all the mound zones were significantly different (*$p<0.05$*) to the $\delta^{13}C$ values of the *Wiborgia sericea* (FH-4), a low shrub, stick-piles, rodent faeces, and rodent nest. While samples collected from only the slope and off mound were significantly different to *Passerina truncta ssp. Truncata* (FI-5), another low shrub, while the *Ruschia suaveolens (FE-8)* samples collected on the centre were significantly different to the termite $\delta^{13}C$ signature. *Wiborgia sericea* (FH-4), and *Passerina truncta ssp. Truncata* (FI-5), were not different (*$p<0.05$*) to the termite or stick-pile samples. Lastly the rodent nest $\delta^{13}C$ values were significantly different (*$p<0.05$*) to the termite's stick-pile $\delta^{13}C$ signatures.

At the Succulent Karoo LD site (Figure 4.6C): $\delta^{13}C$ of *Mesembryanthemum barklyi* (Sp5 -11) an exceptionally succulent herb, had a significantly different and higher $\delta^{13}C$ signature than all the other samples. *Ruschia centrocapsula?* (Sp40-15) and *Cheirodopsis denticulata* (Sp8-16), both succulent shrubs, also separated out from the rest of the samples. These two plants had significantly different (*$p<0.05$*) $\delta^{13}C$ signatures to the frass, *Tetragonia microptera* (Sp10-10) *and Ruschia leucosperma* (sp15-13), however not to the rodent nest, stick-piles, or termite $\delta^{13}C$ signatures. *Ruschia centrocapsula?* (Sp40-15) and *Cheirodopsis denticulata* (Sp8-16) were not significantly different to each other. There were no differences found among *Tetragonia microptera* (Sp10-10), *Ruschia leucosperma* (sp15-13) and *Jordaaniella cuprea* (Sp16-14), and the rodent samples (nest and faeces) or termite samples (frass, termites , stick-piles).These three plant species were the least succulent species at the site (observation).

At the Succulent Karoo MD site (Figure 4.6D) *Mesembryanthemum hypertrophicum* (Sp99-12) was found to have a significantly higher $\delta^{13}C$ to all the samples while frass $\delta^{13}C$ was significantly lower than the rest of the samples. *Lotononis falcata* (Sp25-9), both the off and slope samples, were also significantly different to the $\delta^{13}C$ of the *Tetragonia microptera* (Sp10-10) collected from all 3 zones of the mound. All the plant species at this site have significantly different $\delta^{13}C$ to each other. There were no significant differences found among the $\delta^{13}C$ values of *Tetragonia microptera* (Sp10-10) and *Lotononis falcata* (Sp25-9), with the termites or stick-piles. Again, *Mesembryanthemum hypertrophicum* (Sp99-12) was the most succulent plant species that was collected while the other two were both herbs.



No.	Code	Species name
1	FB	*Chlorophytum triflorum*
2	CT	*Thamnochortus insignis*
3	CS	*Metalasia muricata*
4	FH	*Wiborgia sericea*
5	FI	*Passerina truncta ssp. Truncata*
6	CH	*Searsia undulata*
7	CL	*Ruschia lunulata*
8	FE	*Ruschia suaveolens*
9	Sp25	*Lotononis falcata*
10	Sp10	*Tetragonia microptera*
11	Sp5	*Mesembryanthemum barklyi*
12	Sp99	*Mesembryanthemum hypertrophicum*
13	Sp15	*Ruschia leucosperma*
14	Sp16	*Jordaaniella cuprea*
15	Sp40	*Ruschia centrocapsula?*
16	Sp8	*Cheirodopsis denticulata*

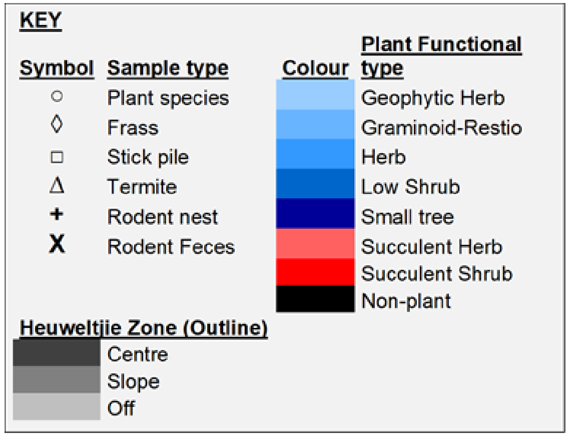


Figure 4.6: $\delta^{13}C$ compared with $\delta^{15}N$ ratios for biological samples collected on heuweltjies at four sub-sites in two biomes. Two based in the Fynbos (A- Fynbos less disturbed (LD); B-Fynbos more disturbed (MD) and the other two in the Succulent Karoo (C-Succulent Karoo less disturbed (LD); D- Succulent Karoo more disturbed (MD)). Each site is plotted on separate graphs above and each point on the graph represents the average of each category with the standard deviation of each value imposed upon the point as dashed lines. Each symbol shape represents a different type of sample while each colour shade shows the variation in plant functional types amongst the plant species samples (circles). The numbers correspond with plant species names. The colour of the outline represents the zone of the heuweltjie from which the samples were taken.

4.4 Discussion

Many plants and animals associated with heuweltjies can be considered ecosystem engineers, due to their ability to change the environment around them. Rodents and termites living in heuweltjie mound soils, forage in the surrounding inter-heuweltjie matrix and bring plant material back into mound. An observational study was conducted to give insight into termite foraging behaviour and was accompanied by an isotopic analysis to investigate what types of plants form part of rodent and termite diets. Isotopic signatures of the plant material were used to gain further insight into the structure of the heuweltjie plant communities.

4.4.1 Termites foraging

Microhodotermes viator are known locally as houtkapper (woodcutter) or stokkiesdraer (stick carrier) termites (Coaton and Sheasby, 1974). They got these names because they forage in groups across the soil surface for vegetation which they then transport into subterranean nests (Dean, 1993). One of the hypotheses proposed in this study, was that termites forage for specific plant species and for specific plant parts or litter types. This was due to the existence of the distinctive stick-piles associated with *M viator*. However, after observing their foraging in the field, this hypothesis does not appear to hold true. Based on the observational analysis conducted in this study, not just one type of plant part was being targeted and collected but rather a mix of all plant parts from a variety of species while they were still fresh. For this reason, it is therefore proposed that *M. viator* are rather generalists and collect all plant material, not just dry sticks.

The termites also did not appear to be targeting any one specific species. This can also be seen from the plant list (Appendix 16) that was produced of all the species that were seen and identified as being used by the termites. It is also evident in the stick-pile photos (Figure 4.4 and Appendix 15). All the stick-piles were made of a different combination of plant species, and this provides evidence that the termites were collecting the plant species that just happened to be close by and were bringing them to these collection points. The termites seemed to show preference for the softer more easily digestible material while foraging (observation), this raised questions about the purpose of the stick-piles. Are they leftover food piles that just never got taken underground because of preference? Or did they get left on the surface because the plant pieces just couldn't fit down the hole fast enough? From personal observations it appeared that there was a possibility that the material was sorted before being taken underground as some plant pieces would get brought down into the hole and immediately brought back out. However, one instance was observed away from a mass foraging event when a few termites were seen slowly bringing one stick down into the hole. Therefore, the question arouse as to whether the stick-piles could be food stores for when food below ground is running low? Another study would be needed, to be able to answer these questions.

Termites are not the only animals to influence heuweltjie ecosystems. However, amongst the animals that are known to occupy heuweltjies, the activities of mole-rats on heuweltjie ecosystem structures, formation, and functioning (Lovegrove and Siegfried, 1986) are worthy of mention and were partially assessed in this study. The more common species associated with heuweltjies in the literature, are the common mole rat *(Cryptomys hottentotuss*), the cape mole-rat (*Georhychus capensis*), or the cape dune mole rat (*Bathyergus suillus*) (Cox et al., 1987; Davies and Jarvis, 1986; Helme, 1990; Lovegrove and Siegfried, 1986). They also forage for plant material, mainly underground bulbs, and tubers, and bring these into their underground nests to be used as bedding or stored as food (Davies and Jarvis, 1986; Lovegrove and Siegfried, 1986). Faeces are deposited on the surface (observation). However, as these are burrowing rodent species, they are much harder to actually observe, identify, and study using behavioural observations. Isotopic analysis was used to investigate the unobservable activities of rodent and termites and show what impact these two species have on the heuweltjie soil and plant communities.

4.4.2 Plants targeted by termites and rodents

Both the isotopes of carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) are naturally occurring stable isotopes and can be used in various ways to study community structure. $\delta^{13}C$ values of plants depend specifically on the method of primary production used by the plant to photosynthesise (e.g., C3, C4, or CAM) and WUE (Water Use Efficiency) (Hartman and Danin, 2010; Spence and Rosenheim, 2005), while $\delta^{15}N$ isotopes of terrestrial plants depend on the source of nitrogen fixed by the plant and also water availability (Gannes et al., 1998; Hartman and Danin, 2010). $\delta^{13}C$ isotopes could be used effectively to trace the termite and rodent diets through the heuweltjie ecosystems using the vast difference in $\delta^{13}C$ signatures of the different photosynthetic pathways in plant species (Fry, 2014; Spence and Rosenheim, 2005). C3 plants have a signature of -34‰ to -22‰ (Gannes et al., 1998), while C4 and CAM plants have $\delta^{13}C$ signatures in the range of -20‰ to -10‰ (Gannes et al., 1998). Because these ranges are so different, resource portioning of primary consumers diets can be seen in the $\delta^{13}C$ isotopic ratios.

It does appear, from the carbon isotope data, that the termites and rodents do show an element of choice in what they forage. Not necessarily in plant species but rather in plant functional types. It was found that termites and rodents do not appear to be collecting and eating the exceptionally succulent plants. To come to this conclusion, a few assumptions had to be made. Firstly $\delta^{13}C$ isotopic ratio of a consumers body should resemble its diet (Fry, 2014; Hyodo, 2015) and secondly the termites stick-piles and rodent nests represent the plants being eaten by these two species. The termite $\delta^{13}C$ and stick-piles $\delta^{13}C$ were found to be statistically the same as each other, at all sites. No rodent tissue was able to be sampled but it was still assumed that the nest samples found were a collection of species that the rodents were consuming. The $\delta^{13}C$ values of the stick-piles and the rodent nests along with the species recoded and identified in the observations (Appendix 16) were used to categorie the sampled plant species according to section 4.2.3 and indicate if they were targeted by termites and rodents or not.

Table 4.3: Showing collected plant species, with their functional types and the zone they were collected from, with the classification of whether they were targeted by termites and rodents, or not. This classification was done by assessing if the plant species met three conditions (shown using a Yes/No) 1) if they were observed being carried into the mound or seen in a termite stick-piles (Table 4.2 and Appendix 16), 2) if the $\delta^{13}C$ values were not significantly different to the termite stick-piles or termites and 3) if the plant species $\delta^{13}C$ values were not significantly different to the rodent nest sample. If the plant species met all conditions (all Yes), it was classified as: Targeted, No conditions (all No's): Not Targeted, and a mix of conditions: Maybe

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						Termite: Observation	Termite $\delta^{13}C$			Rodent $\delta^{13}C$		
Site	Zone collected	Species Code	Species name	$\delta^{13}C$: Plant species	Functional type	Yes/No	Termite bodies ($\delta^{13}C$)	Stick-pile ($\delta^{13}C$)	Yes/No	Rodent nest ($\delta^{13}C$)	Yes/No	Targeted Classifications
Fynbos -LD							-26.58±0.43 (4)	-26.62±0.22 (7)		No rodent nest samples		
	Centre	CH	Searsia undulata	-25.65 ± 0.73(4)	Small tree	Yes			Yes			Targeted
		FE	Ruschia suaveolens	-24.85 ± 0.52(4)	Succulent Shrub	No	**	***	No			Not Targeted
	Slope	CL	Ruschia lunulata	-24.99 ± 0.55(4)	Succulent Shrub	Yes	**	***	No			Maybe
		FB	Chlorophytum triflorum	-28.08 ± 0.84 (4)	Geophytic Herb	Yes	**	**	No			Maybe
	Off	CS	Metalasia muricata	-26.53 ± 0.37 (4)	Low Shrub	Yes			Yes			Targeted
		CT	Thamnochortus insignis	-25.73 ± 0.43 (4)	Graminoid-Restio	Yes			Yes			Targeted
Fynbos -MD							-25.50 ± 0.53 (2)	-26.05 ± 0.45 (4)		-28.92 ± 0.04 (2)		
	Centre	FE	Ruschia suaveolens	-24.02 ± 0.35 (4)	Succulent Shrub	No	*	***	No	***	No	Not Targeted
		FB	Chlorophytum triflorum	-28.04 ± 0.38 (4)	Geophytic Herb	Yes	***	***	No		Yes	Maybe
	Slope	FE	Ruschia suaveolens	-24.04 ± 0.47 (4)	Succulent Shrub	No		***	No	***	No	Not Targeted
		FH	Wiborgia sericea	-26.59 ± 0.31 (4)	Low Shrub	No			Yes	***	No	Maybe
	Off	FE	Ruschia suaveolens	-24.70 ± 0.51 (4)	Succulent Shrub	No		*	No	***	No	Not Targeted
		FI	Passerina truncta ssp. Truncata	-25.79 ± 0.88 (4)	Low Shrub	No			Yes	***	No	Maybe
Succulent Karoo -LD							-20.40 ± 0.35 (4)	-20.49 ± 2.54 (7)		-22.34 ± 0.77 (2)		
	Centre	Sp10	Tetragonia microptera	-22.45 ± 0.65 (4)	Succulent Herb	Yes			Yes		Yes	Targeted
		Sp5	Mesembryanthemum barklyi	-13.64 ± 0.92 (4)	Succulent Herb	No	***	***	No	***	No	Not Targeted
	Slope	Sp40	Ruschia centrocapsula?	-18.84 ± 2.48 (4)	Succulent Shrub	No			Yes		Yes	Maybe
		Sp8	Cheirodopsis denticulata	-18.53 ± 0.34 (4)	Succulent Shrub	No			Yes		Yes	Maybe
	Off	Sp15	Ruschia leucosperma	-22.78 ± 0.48 (4)	Succulent Shrub	Yes			Yes		Yes	Targeted
		Sp16	Jordaaniella cuprea	-20.37 ± 0.51 (4)	Succulent Shrub	Yes			Yes		Yes	Targeted
Succulent Karoo -MD						no termite observations	-23.55 ± 0.35 (2)	-23.67 ± 0.86 (4)		No rodent nest samples		
	Centre	Sp10	Tetragonia microptera	-23.03 ± 0.41 (4)	Succulent Herb				Yes			Targeted
		Sp99	Mesembryanthemum hypertrophicum	-15.60 ± 0.20 (4)	Succulent Herb		***	***	No			Not Targeted
	Slope	Sp10	Tetragonia microptera	-22.93 ± 0.63 (4)	Succulent Herb				Yes			Targeted
		Sp25	Lotononis falcata	-24.53 ± 0.33 (3)	Herb				Yes			Targeted
	Off	Sp10	Tetragonia microptera	-23.03 ± 0.48 (4)	Succulent Herb				Yes			Targeted
		Sp25	Lotononis falcata	-24.66 ± 0.52 (4)	Herb				Yes			Targeted

The rodent nest $\delta^{13}C$ values is significantly different (*p<0.05*) to the termite's stick-pile $\delta^{13}C$ signatures in the Fynbos which suggests a complete split in diets of these two animals in the Fynbos. However, there were no differences amongst the rodent nest, termite stick-piles and actual termites $\delta^{13}C$ values from the samples in the Succulent Karoo, which suggests they are targeting the same plant species in this biome. The interpretation of the results from the $\delta^{13}C$ isotope analysis and the plant species list produced of plants seen collected by termites (Appendix 16) were used to classify each plant species according to if it was targeted, maybe targeted, or not targeted by termite and rodents (Table 3.2). The logic behind these classifications can be found in Section 4.2.3. In the Fynbos at the less disturbed site, the termites appeared to be targeting, *Searsia undulata* (CH- 6) a small tree, *Thamnochortus insignis* (CT-2) a graminoid and *Metalasia muricata* (CS- 3) a low shrub. While at the more disturbed site, *Wiborgia sericea* (FH-4); and *Passerina truncta ssp. Truncata* (FI-5) -both low shrubs - were targeted. *Chlorophytum triflorum* (FB-1), a geophytic herb, was the only species sampled that did not have significantly different $\delta^{13}C$ signature to the rodent nest sample. *Chlorophytum triflorum* (FB-1) was the only geophytic herb that was sampled at this site and the Fynbos rodent nest sample, mostly consisted of bulbs (observations), however it was classified as only maybe targeted as the plant had a significantly different $\delta^{13}C$ signature to both the termites and the stick-piles at both Fynbos sites. The plants species being targeted by the termites and rodents in the Succulent Karoo was not as clearly split between the animal species as in the Fynbos. At the less-disturbed Succulent Karoo site and recognised in the stick-piles across the site, *Tetragonia microptera* (Sp10-10), *Ruschia leucosperma* (Sp15-13) and *Jordaaniella cuprea* (Sp 16-14), a succulent herb and two succulent shrubs respectively, were found to have the same isotopic $\delta^{13}C$ signature as the rodent nest and termite stick-piles and termites. These three plant species all had succulent leaves but were only leaf succulents. At the more disturbed site *Tetragonia microptera* (Sp10-10) a succulent herb and *Lotononis falcata* (Sp25-9) an herb, had the same $\delta^{13}C$ signatures as the termites and stick-piles.

$\delta^{13}C$ values of plants, which are used to make the classifications are not solely the results of differences in photosynthetic pathways. Values closer to 0 can also indicate greater water - use efficiency of plant species. The greater variability in the $\delta^{13}C$ of the Succulent Karoo might indicate that a greater diversity of photosynthetic pathways exists in the plant species of the Succulent Karoo, however the communities were found to consist of a smaller number of functional groups, so less variability in the photosynthetic types was to be expected. Influences in water use efficiency might also very likely play a role in the Succulent Karoo communities. As the Figure 4.6 shows these plants experience more stressful water conditions and increased aridity than the plants in the Fynbos biome experience (Farquhar et al., 1989; Hartman and Danin, 2010).

$\delta^{15}N$ isotope signatures in the plant samples indicate from where the nitrogen gets sourced. Plants can either obtain their nitrogen from the soil or from the atmosphere (Fry, 2014; Hyodo, 2015). As there is almost no fractionation when $\delta^{15}N$ is taken into the plant tissues the $\delta^{15}N$ signature will remain the same as the source (Gannes et al., 1998). So, the $\delta^{15}N$ signature of a plant could be because of a plant specific species nitrogen fixation ability, or because of soil nitrogen concentrations. The $\delta^{15}N$ of atmospheric nitrogen is 0. Therefore, plants with signatures close to 0 indicate the ability to fix atmospheric N_2. However as most of the plants are not close to 0 but still variable this indicated that other influences would be causing the variability seen (Hyodo, 2015).The average $\delta^{15}N$ values for each of the different samples showed more variation than the $\delta^{13}C$ values, with the greatest variation in the samples at the more disturbed sites. This is likely due to the more disturbed sites having greater agricultural inputs and varying degrees of denitrification in the soils. It further shows what effect grazing and other anthropogenic disturbances can have on an ecosystem, along with lower levels of diversity, and even plant communities as seen in Chapter 2. Values of the $\delta^{15}N$ isotopes should be interpreted with caution as the Acacia standard for one of the sample batches was >0.2, which means there could have been instrumental error in the values of the data. This could have also been one of the reasons for the high variation in these results.

4.5 Conclusion

At the time that this study took place, the rodents and termites appeared to not be collecting the exceptionally succulent plant species. The most succulent plant species at all four sites were found to have significantly different $\delta^{13}C$ values to the rodent nests and termite and stick-piles. At the Fynbos the only succulent species were *Ruschia lunulata* (CL-7) and *Ruschia suaveolens* (FE-8) which were both succulent shrubs that had different $\delta^{13}C$ values ($p<0.05$) than both the stick-piles, termites, and rodent nest. Although the Succulent Karoo, as the name suggests, hosts a higher quantity of succulent plant species, the same pattern was seen in this biome. Here the stem-and leaf succulent plant species, which can be considered the most succulent of all the samples, had significantly different $\delta^{13}C$ to the termite and rodent samples. These plants were *Mesembryanthemum barklyi* (Sp5-11) from the less-disturbed site and *Mesembryanthemum hypertrophicum* (Sp99-12), found at the more disturbed site.

There could be a few reasons why exceptionally succulent plant species are not being targeted by termites or rodents: 1) mechanics- the succulent leaves were often much larger and heavier than the other plant species and possibly were too big for the termites to carry and fit down the holes into the mounds 2) succulence is a life strategy of plants to conserve water and deal with salt stress (Males, 2017; Ogburn and Edwards, 2010), there is the possibility that these species have too much salt in their leaves for the termite diet. Chapter 5 will in part try to investigate this last reason by trying to combine the classifications in this chapter and the plant ion concentrations of Chapter 3, to investigate what impact the foraging activities of termites and rodents might have on the heuweltjie salt dynamics.

Chapter 5: Termites as a vector for salt movement in the heuweltjie landscape

5.1 Introduction

Heuweltjie mounds have been found to host different plant communities on the different zones of the mound (Chapter 2). When the plants die or drop their leaves, they will contribute any plant material to the soil where they grew along with the ions held in their tissues (Chapter 3). However, animals such as termites, living in the mound and foraging for plant material, add an extra layer of complexity to this story. Targeted plant material will get moved from the zone where its parent plant established and brought into mound soils (Chapter 4). It is this that has been hypothesised to have a concentrating effect on centre mound ion concentrations and will be investigated in this discussion.

Termites have a large influence on heuweltjies, but they are not the only animal that lives in and uses heuweltjie ecosystems: ants, scorpions, earthworms, different rodent species, livestock, aardvarks, bat-eared foxes, reptiles like tortoises and snakes, as well as various antelope species were all observed on and around heuweltjies during this study. Each of these species would have a part to play and would impact inputs and outputs of the heuweltjie ion and nutritional cycles. However, this current study's main focus was the termite species *Microhodotermes viator*. Ion accumulation in the plant material brought into mounds by *M viator* have been suggested before as a manner in which heuweltjie soils increase salinity, with evidence of magnesium and calcium accumulation being a result of termite foraging (Francis et al., 2013; Francis and Poch, 2019; McAuliffe et al., 2019b, 2014). This was suggested due to similarity amongst ion profiles in termite nest soils and frass, with heuweltjie soils (Cornell, 2014; Kunz et al., 2012; McAuliffe et al., 2019b, 2014). Using the data collected in the above Chapters, this Chapter will further this investigation by linking the plant species targeted by termites to various ions in and on the plant material. This will be structured by a series of three questions, where each question will have its own mini methods, results, and discussion sections. These questions will assess the last objective, whether termites, rodents and plants play a role in the salinity distributions in the heuweltjie ecosystems by discussing the possible impacts of the foraging activities of *M viator* on salt and ion dynamics in heuweltjies soils.

5.2 Are salts and oxalates (internal) being brought into heuweltjies through the foraging activities of termites and rodents?

5.2.1 Methods: Reclassification of Ions and minerals in internal plant tissues.

The concentration of ions in the plant material (Table 3.1) was reclassified according to the groups produced in Table 4.3 and shown in Table 5.1. The groups "targeted,", "maybe targeted" and "not targeted" were compared statistically using Kruskal-Wallis tests, followed by post hoc Wilcoxon rank sum tests. All of the ions analysed did exist in various concentrations, in the plant material brought in by termites and rodents. Low and high concentrations of each ion in the targeted plant material were compared, relative to the non-targeted plant material, as the ion concentrations of centre mound soils would be more influenced by the targeted plant material than the non-targeted, due to the foraging actions of rodents and termites. The minerals identified in the XRD analysis (Table 3.2), were also reclassified according to Table 4.3, and shown in Table 5.2.

It is assumed that rodents and termites bring any plant material that gets collected, into centre mound soils where their nests are located. The classifications of zone and targeted or not targeted are self-assigned groups. So, to bring further insight and to act as an objective reference, the ion concentrations of the stick-piles and rodents' nests were quantified (Table 3.1) and were compared with the ion concentrations of the targeted plants. This was done by looking at whether the ranges of the targeted plant ion concentration (means and standard deviations) overlapped with the ion concentrations ranges (means and standard deviations) of the termite stick-pile and rodent nest samples. This was used to assess and give more validity to the ion concentrations in the targeted and non-targeted groups.

5.2.2 Results: Cations, anions, and minerals in the targeted plant material

At the time of sampling, foraging herbivores, termites, and rodents were found to be foraging for and bringing in plants with significantly lower concentrations of sodium into the centre mound soils of both biomes, compared to plants not likely targeted by these animals (Table 5.1). Low concentrations of calcium, magnesium, manganese, and zinc were found in plants being brought into the Fynbos centre mound soils (Table 5.1). Magnesium and zinc in the plant material of the Succulent Karoo, and aluminium, iron, and copper in the plant material of the Fynbos, were found to exist in higher concentrations in the targeted plant material. No differences were found in concentrations of boron and iron in the Succulent Karoo, nor in phosphorus in the plant material targeted by termites and rodents and not targeted at the Fynbos sites. Potassium and boron concentrations in the fynbos plants and calcium, aluminium, and manganese in the Succulent Karoo, were found to be significantly higher in the maybe targeted plants of the Succulent Karoo.

Sodium in both the stick-piles and rodent nest were found to be comparable but less than the targeted sodium concentrations from the samples in both biomes. With most of the contribution coming from termites in the Fynbos, and rodents in the Succulent Karoo. Calcium concentrations in the Fynbos samples were comparable, with very similar concentrations between the termite stick-pile and the targeted calcium concentrations in the Fynbos as well as with lower calcium concentrations in the rodent nest. In the Succulent Karoo, the calcium concentrations were again comparable but higher in the stick-pile and nest than the targeted plants. Magnesium was found to be comparable in both biomes, with the stick-piles of the Fynbos having a higher concentration of magnesium than the targeted plant species and the rodent nest having a lower concentration. A lower concentration of magnesium was also found in both the rodent nests and stick-piles of the Succulent Karoo. Phosphorous was also comparable in the Fynbos with very similar concentrations amongst all the samples. In the Succulent Karoo the termite stick-pile and rodent nest phosphorus concentrations were found to be lower, but the mean and standard deviation ranges did just overlap with the targeted plant range. Potassium was the last major ion investigated, and the stick-pile and rodent nest concentrations were found to be not comparable to the targeted plant species. It was found that the potassium concentration of the targeted plants were much higher than the stick-pile and nest amounts for both biomes.

Table 5.1: Ion concentrations of internal plant tissues grouped by classifications in Table 4.3 whether the plant species are targeted, maybe targeted or not targeted by termites and rodents.

Ions	Biome	Targeted (Fynbos: n=32 S Karoo n=60)	Maybe Targeted (Fynbos: n=32 S Karoo n=16)	Not Targeted (Fynbos: n=32 S Karoo n=16)
Sodium (%)	Fynbos	0.29±0.18a	0.40±0.52a	1.73±0.63b
	S. Karoo	4.37±3.23a	5.22±2.34a	23.71±9.05b
Calcium (%)	Fynbos	0.86±0.83a	1.13±1.01a	1.99±0.55b
	S. Karoo	1.62±1.45a	4.05±1.12b	1.94±1.31a
Magnesium (%)	Fynbos	0.15±0.07a	0.26±0.31a	1.31±0.52b
	S. Karoo	0.71±0.21a	2.15±1.86a	0.42±0.10b
Phosphorus (%)	Fynbos	0.05±0.02a	0.05±0.03a	0.05±0.02a
	S. Karoo	0.12±0.05b	0.08±0.05a	0.08±0.02a
Potassium (%)	Fynbos	0.83±0.26a	0.67±0.44b	1.69±0.48c
	S. Karoo	1.36±0.45a	1.21±0.45a	1.58±0.61a
Aluminium (mg/kg)	Fynbos	309.17±264.46a	555.73±788.68a	157.72±118.46b
	S. Karoo	857.67±610.49a	1492.59±899.57b	2043.13±2448.10ab
Boron (mg/kg)	Fynbos	28.88±15.82ab	24.48±7.56a	32.75±4.55b
	S. Karoo	38.47±22.59a	33.61±11.63a	32.88±13.81a
Copper (mg/kg)	Fynbos	6.24±4.05ab	5.47±2.4a	4.38±2.77b
	S. Karoo	4.81±1.10a	3.07±0.91b	3.12±1.98ab
Iron (mg/kg)	Fynbos	303.09±223.22a	669.64±1199.76a	151.70±107.89b
	S. Karoo	901.84±736.82a	1035.29±671.59a	1653.29±1862.20a
Manganese (mg/kg)	Fynbos	249.67±140.49a	652.95±575.46b	2141.21±999.45c
	S. Karoo	157.85±104.54a	372.07±273.94b	103.59±35.20c
Zinc (mg/kg)	Fynbos	13.76±6.16b	19.44±9.25a	17.38±7.62a
	S. Karoo	14.59±4.89b	8.81±2.42a	10.76±4.35a

Aluminium and iron concentrations in the stick-pile and rodent nests were both found to be higher but comparable to the targeted plant species in the Fynbos. The Succulent Karoo samples however were higher and not comparable. Boron concentrations were comparable and lower in the stick-pile and rodent nests in both biomes. Copper in Fynbos samples showed the same patterns as boron, but the Succulent Karoo stick-pile and rodent nests were comparable but higher than the targeted plant copper concentrations. Manganese concentrations in the rodent nests and stick-piles were comparable but higher than the targeted plants, except for the rodent nest in the Succulent Karoo. Because this had a higher range than the targeted plants, it was considered not comparable. Lastly zinc in the samples had comparable ranges with the stick-piles in both biomes, having very similar means and standard deviations to the targeted plants. Rodent nests had lower concentrations of zinc than the targeted plants.

Previous studies have assumed that termites were bringing calcium-rich plant material into the heuweltjie through their foraging activities, as a part of the process in the formation of the calcite layers in heuweltjies.

In these studies, the link has not been shown, but rather just assumed that calcium oxalate is being brought into mounds through the actions of termites (Francis et al., 2012; Francis and Poch, 2019; McAuliffe et al., 2019a). Table 5.2 regroups the identified minerals according to whether they were targeted by termites and rodents or not. Calcium oxalate in the form of whewellite could be found present in the targeted plants in both biomes, and sodium oxalate was found in the targeted plant material of the Succulent Karoo. Calcium oxalate in the form of weddellite was not in the targeted plant material for either biome. Pentahydrite, halite and sylvite could be found in the targeted plant material of the Succulent Karoo.

*Table 5.2: The presence of various minerals in plant samples, identified using XRD analysis, grouped by if they were targeted or not. *'s represents if a mineral was found present in the plant species in the sample group.*

Biome	Grouping	Weddellite $Ca(C_2O_4)\cdot 2H_2O)$	Whewellite $(CaC_2O_4\cdot H_2O)$	Sodium oxalate	Pentahydrite $(MgSO_4.5H_20)$	Halite (NaCl)	Sylvite (KCl)
Targeted vs Not							
Fynbos	Targeted (n=4)		*				
	Not Targeted (n=4)	*	*		*	*	*
	Maybe (n=4)	*	*		*		*
Succulent Karoo	Targeted (n=8)		*	*	*	*	*
	Not Targeted (n=2)		*	*		*	*
	Maybe (n=2)		*	*		*	*

5.2.3 Discussion on whether salts and oxalates are being brought into heuweltjies through the foraging activities of termites and rodents

The stick-pile and rodent nest ion concentration (Table 3.1) appeared to be slightly different to the targeted plant ion concentrations (Table 5.1). Both sample groups have their pros and cons as a measure of what concentration of salts in the plant material that can be attributed to the termites and rodents foraging. The targeted plant material consisted of plant species physically seen being carried into mounds by termites, and visually seen in the stick-piles, as well as plant species with the same carbon isotope signatures as termites stick pile and rodent nests (as these two types of samples were assumed to be plant material collected by the respective animal species). Therefore, the targeted grouping is a categorisation to show what plant material is being brought into the mound, no matter the animal species. The stick-pile samples were possibly a more accurate grouping to assess termite plant ion input. However, when observing the termites actually forage, it was noted that they collected fresh plant material, – in fact much more than just the twigs and sticks that the stick-piles are characteristically made up of. It was deduced that the stick-piles are discarded food, left unwanted on the surface. Alternatively, they could be food stores used to top up whatever had been taken underground. The rodent nests samples were also questionable as food sources, the Fynbos samples were collected from underground within the heuweltjies soils during an excavation of a heuweltjie for another master project (Hattingh, 2021) (submitted for examination), where plant material was found stored in an underground chamber. These were most likely winter food stores as they contained bulbs and were collected just before the start of spring. On the other hand, they could have also been bedding which was used as part of a nest. Whatever the case, it can be safely concluded that this plant material would eventually have decomposed in the centre mound soils. The Succulent Karoo nest sample was found on the centre surface of heuweltjies by what appeared to be a discarded hole. Plant material and faecal pellets were mixed together and appeared to have been pushed out of a hole, forming part of a rodent burrow system (see Appendix 14 for photos of these plant samples). Due to this uncertainty and to give an indication of what plants and therefore what ions were being brought into the mounds by termites, both the targeted plant material and the stick-piles ion concentration values need to be considered in the following discussions.

5.3 What other factors influence the external plant washes?

5.3.1 Methods: Reclassification of sodium wash results

Sodium concentrations measured from the plant washes were investigated with greater detail to see if any groups could be found within the plant data categories (Figure 5.1). Sodium was used because it was the most dominant ion and behaves conservatively in reactions, i.e., it does not have many other factors influencing the concentrations. Kruskal-Wallis tests, followed by post hoc Wilcoxon rank sum tests were used to compare groups

5.3.2 Results: Differences in sodium external plant ions deposits, amongst biomes, zones, and species

Plant internal tissues were not the only source of ions which were identified to be moved around and concentrated in mound soils due to actions by termites. Any ion deposits on the external surface of the plant material also followed this pathway into mound soils. Due to the nature of aerosols and fog, no differences amongst any of the groups (targeted, zone, species) was expected to be found, but rather just a general and even spread of the marine aerosols on all plant surfaces was expected. Na^+ concentrations between biomes were found to be different ($p<0.05$), which would be expected due to the vast spatial and climatic differences between the sites. No differences however were found amongst the zones when looking at the data grouped by zone, providing evidence for the uniform nature of the aerosol deposition across the mound. Differences were also found ($p<0.05$) amongst the different plant species. In the Fynbos this was between species *Chlorophytum triflorum* (FB) and *Passerina truncta ssp. Truncata* (FI), which occurred on the bottom and top of the ranges of sodium but neither separated out from the group. However, in the Succulent Karoo *Mesembryanthemum barklyi* (Sp5), *Tetragonia microptera* (Sp10), and *Mesembryanthemum hypertrophicum* (Sp99), were found to be different to most of the plant species. This can be seen in the separated group that had higher sodium concentrations than the rest of the plant samples. These three plants were the species with bladder cells which were removed from the previous analysis. Lastly differences were found ($p<0.05$) amongst the external salts when categorised by targeted and non-targeted plants. However, this was just between the maybe targeted and the not targeted categories and could possibly be because of the inclusion *Mesembryanthemum barklyi* (Sp5), *Tetragonia microptera* (Sp10), and *Mesembryanthemum hypertrophicum* (Sp99).

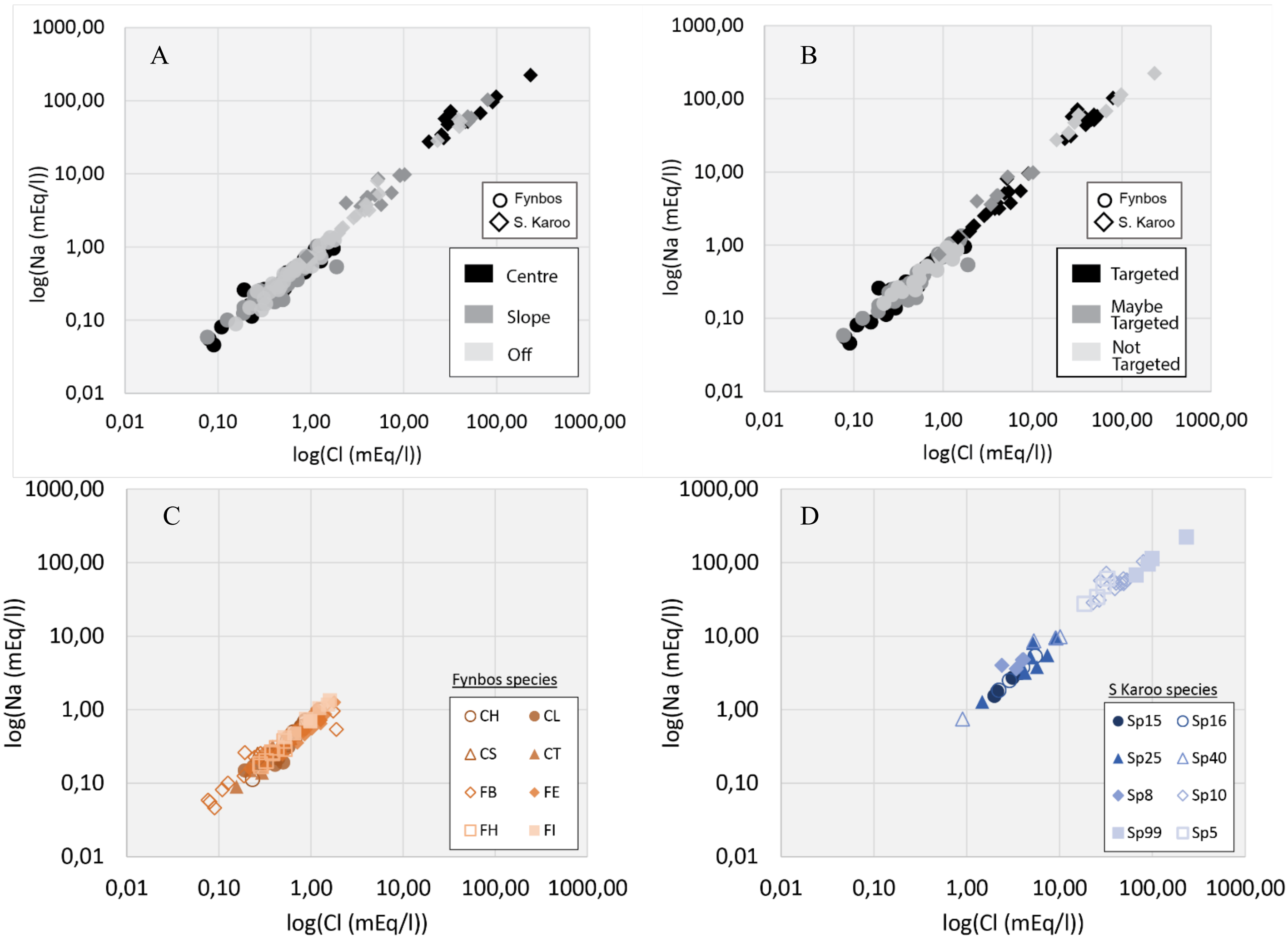

Figure 5.1: Graph showing the ionic ratio of the log of Na⁺ (mEq/l) vs Cl (mEq/l), for the solutions washed off the external surface of the different plant species found growing on Heuweltjies. Each graph shows the same samples coloured with different categories. A: The plants are coloured according to which zone they were found growing on; B: The plants are coloured according to the categories if they were targeted by termites and rodents or not; C and D have different colours and symbols showing each of the individual plant species (See Appendix 7 for names). C shows just the Fynbos plant species and D shows the Succulent Karoo plant species.

5.3.3 Discussion: Source of external plant deposits

After investigating the external salt depositions, in terms of the plant distributions on the mounds and behaviour of termites- using sodium as a subject, no significant deviations could be found to change the conclusions from the previous analysis (Chapter 3). There were slight plant species differences but only in the species that were considered outliers and were removed from the previous analysis while the others were still part of the main ion distribution. By eliminating these other factors as influences, we can show that the dominant contribution to the plant washes must indeed be the marine aerosols factor.

5.4 Which ions (external and internal) are being brought into centre mound soils by termites, and in what quantity?

5.4.1 Methods: Calculations of ion inputs

Ions found in and on plant material were reassessed above to see what ions could be associated with termite foraging. Even though it was found that the plants that formed part of termite diets, were not the plants which held exceptionally high salts, over time even these small amounts could accumulate in the mound soils and become a significant input. Calculations were done to investigate the rate of plant ion input by termites into mound soils (See Table 5.3 for references of input data). *Microhodotermes viator* have been found to forage for an average of an hour a day (Dean, 1993) and are active all year round with higher activity levels in summer (Dean, 1993). For the purposes of this calculation, it will be assumed that termites forage for 1 hour a day for every day of the year. Termite foraging collection was done for an average of 4 minutes (Chapter 4), with an average of 0.3266 grams being collected per foraging hole (n=14) (Chapter 4). The collection of plants being carried into the mounds is definitely an underestimate since some plant pieces were taken into the mounds by termites before they could be collected. Moreover, some termites dropped their plant material prematurely, as they were not habituated to the collectors and were more skittish than usual. In spite of this however, an average per minute rate of plant material being brought into the mound could be worked out. The amount of plant material brought into the mound from one foraging hole on one heuweltjie was calculated using the equation below (Equation 5). The weight of plant material being brought into one foraging hole per minute was estimated to be 0.0817 g per min, 4.899 g per hour and 1788.135 g per year

$$Input\ plant\ material\ (per\ year) = \frac{0.327\ g}{4\ mins} \times 60\ minutes \times 365\ days \quad \text{(Equation 5)}$$

Table 5.3: Values and references for input data of flux calculations

Input data	Value	units	Reference
Collected plant material (average for 1 hole)	0.327	grams	Section 4.3.1
Time collected	4	minutes	Section 4.2.1.1
Average foraging time per day	60	minutes	(Dean, 1993)
Targeted average ions			Table 5.1
Stick-pile ion concentrations			Table 3.1

Ion concentrations of the plant surface washes were tabularised and grouped according to the zone the plant was found growing on. Categorisations of targeted vs non-targeted were also done to assess what the inputs of these groups of plants were, from external sources.

5.4.2 Results: What quantity of ions are being brought in by termites?

The average concentration of ions in the targeted plant material (Table 5.1) and the stick-piles (Table 3.2) was used to calculate an estimated average weight of various ions, brought into the mound by termites (Table 5.4). An estimated 4.0 g of sodium in the Fynbos and 44.9 g in the Succulent Karoo is possibly being added into the mound soils every year from one foraging hole. Calcium inputs per year are estimated to be 18.4 g (Fynbos) and 41.5 g (S. Karoo). Estimated inputs of magnesium were 4.15 g in the Fynbos and 10.6 g in the Succulent Karoo and potassium ranged from 9.69 g in the Fynbos and 15.0 g in the Succulent Karoo. The remaining ions were all lower than 4 g per year and can be found in Table 5.4.

Table 5.4: Results of calculations of the estimated amounts of ions being brought in the plant material from one foraging hole, on one heuweltjie mound, based on ion concentrations from targeted plant species and the stick-piles. The estimated average amount of each ion per year is the average of the targeted plants and stick-pile estimations

Ion	Biome	Targeted plants			Stick-pile			Estimated average: per year (g)
		Per minute (mg)	Per hour (mg)	Per year (g)	Per minute (mg)	Per hour (mg)	Per year (g)	
Sodium	Fynbos	0.24	14.38	5.25	0.13	7.53	2.75	4.00
	Succulent Karoo	3.57	214.00	78.11	0.54	32.46	11.85	44.9
Calcium	Fynbos	0.70	42.03	15.3	0.98	58.8	21.48	18.4
	Succulent Karoo	1.32	79.3	28.9	2.47	148	54.1	41.5
Magnesium	Fynbos	0.13	7.51	2.74	0.25	15.22	5.56	4.15
	Succulent Karoo	0.58	34.57	12.62	0.39	23.6	8.62	10.6
Phosphorus	Fynbos	0.04	2.27	0.83	0.03	1.70	0.62	0.72
	Succulent Karoo	0.09	5.70	2.08	0.07	4.36	1.59	1.84
Potassium	Fynbos	0.68	40.6	14.8	0.21	12.5	4.57	9.69
	Succulent Karoo	1.11	66.9	24.4	0.26	15.5	5.67	15.0
Aluminium	Fynbos	2.52×10^{-2}	1.51	0.55	0.19	11.2	4.09	2.32
	Succulent Karoo	7.0×10^{-2}	4.20	1.53	0.27	15.9	5.84	3.68
Boron	Fynbos	2.40×10^{-3}	0.14	0.05	1.80×10^{-2}	0.11	0.04	4.50×10^{-2}
	Succulent Karoo	3.10×10^{-3}	0.19	0.07	1.90×10^{-2}	0.12	0.04	5.56×10^{-2}
Copper	Fynbos	5.10×10^{-4}	3.06×10^{-2}	1.12×10^{-2}	3.71×10^{-4}	2.23×10^{-2}	0.81×10^{-2}	9.6×10^{-3}
	Succulent Karoo	3.93×10^{-4}	2.36×10^{-2}	0.86×10^{-2}	5.01×10^{-4}	3.00×10^{-2}	1.10×10^{-2}	9.8×10^{-3}
Iron	Fynbos	2.47×10^{-2}	1.48	0.54	0.20	11.9	4.37	2.46
	Succulent Karoo	7.36×10^{-2}	4.42	1.61	0.28	16.	6.08	3.84
Manganese	Fynbos	2.04×10^{-2}	1.22	0.45	8.37×10^{-2}	5.02	1.83	1.14
	Succulent Karoo	1.29×10^{-2}	0.77	0.28	2.10×10^{-2}	1.26	0.46	0.37
Zinc	Fynbos	1.12×10^{-3}	6.74×10^{-2}	2.46×10^{-2}	1.4×10^{-3}	8.25×10^{-2}	3.01×10^{-2}	2.73×10^{-2}
	Succulent Karoo	1.19×10^{-3}	7.15×10^{-2}	2.61×10^{-2}	1.2×10^{-3}	7.00×10^{-2}	2.55×10^{-2}	2.58×10^{-2}

Table 5.4 only shows the termite ion inputs of the plant internal tissues. There are also the external plant salts to consider. Table 5.5 shows the average concentrations of the ions from the external plant washes grouped according to the location the plants were found growing in, for all the plants per zone and then for just the targeted plants that identified in each zone. For example, calcium concentrations are 3.53 mg/L on the Fynbos plants and 22.01 mg/L for the S. Karoo plant species. While the average concentration for just the targeted plants in the Fynbos was 3.42 and 28.11mg/L in the S. Karoo

Table 5.5: Average concentration of each ion, from the external surfaces of the plants split by biome, zone and if the plants were targeted. (Averages of just the targeted plants can be seen in brackets). The data does not include the averages of the plants, that were found to have high EC.

Categories	Ca (mg/L)	Na (mg/L)	SO4 (mg/L)	Mg (mg/L)	Cl (mg/L)
Fynbos	3.53 (3.42)	10.06 (9.67)	3.58 (3.97)	7.44 (3.28)	22.63 (19.98)
Centre	3.63 (4.03)	10.76 (13.09)	4.14 (6.32)	8.21 (2.90)	23.90 (26.24)
Slope	3.18 (0)	7.38 (0)	2.76 (0)	8.61 (0(	18.25 (0)
Off	3.79 (3.12)	12.04 (7.97)	3.85 (2.79)	5.49 (3.46)	25.75 (16.85)
Succulent Karoo	22.01 (28.11)	104.41 (90.75)	32.63 (40.25)	59.89 (65.02)	157.43 (148.92)
Slope	33.18 (79.88)	133.86 (138.15)	37.14 (76.62)	81.20 (144,34)	195.91 (238.84)
Off	10.85 (10.85)	74.95 (74.95)	28.13 (28.13)	38.59 (38.59)	118.95(118.95)

The above estimations of ion inputs are only for 1 foraging hole on one heuweltjie. From observations of foraging colonies, 2-7 foraging holes (section 4.3.1) were active per foraging session. In data collection for Chapter 2 a systematic sampling of the number of holes per quadrat was undertaken and can be found in Appendix 4. The relevant sections of the table have been summarised below which shows the number of holes per m^2 on the different zones of the mounds. The number of holes per heuweltjie that termites use will depend on the size of the heuweltjie this was not measured in this study. However, given that a 10 m wide circle has an area of around 78 m^2, based off a broad estimation from numbers in Table 5.6 there could be over 80 extra small holes (~1 hole per m^2) and 40 small holes (~0.5 holes per m^2) per heuweltjie. Not all of these holes would belong to termites as many other insects also reside in the heuweltjie soils. The number of stick-piles would be a more accurate estimation of foraging holes (~0.05 stick-piles per m^2). These numbers appear to be an under estimation compared to visual observations of the site.

Table 5.6: Number of extra small (~<0.5 cm), small (~1 cm-5 cm) holes, and stick-piles per m^2 from heuweltjie surface characteristic in Table B in Appendix 4. And the estimated no. of stick-piles per heuweltjie

Biome Zone	Fynbos Centre	Fynbos Slope	Fynbos Off	S. Karoo Centre	S. Karoo Slope	S. Karoo Off
No per m^2						
XS hole	1.02±1.3	2.2±2.1	1.3±1.7	0.5±0.68	1.03±1.0	0.98±1.2
S hole	0.4±0.5	0.5±0.7	0.65±0.9	0.3±0.4	0.3±0.6	0.6±1.01
Stick piles	0.01±0.05	0.04±0.12	0.12±0.29	0.00	0.02±0.07	0.02±0.07
Estimated no. per heuweltjie (~10m in diameter)						
Stick-piles	0.78	3.12	9.36	0	1.56	1.56

5.4.3 Discussion: What could the impacts of these input rates be, over the time heuweltjies have been predicted to exist for?

Over the lifespan that heuweltjie have been predicted to have formed (4000-30000 years (Midgley et al., 2002; Moore and Picker, 1991; Potts et al., 2009)) even the smaller amounts of ions brought in by termites will accumulate to significant numbers. Take sodium and calcium for example, using the average yearly input rates (Table 5.3) and multiplying them each respectively by 4000 or 30000 years a total input can be calculated. Sodium input due to termites overall in the Fynbos could be: 16 kg to 120 kgs while total inputs over the time frame in the Succulent Karoo could be: 180 kg to 1.35 tons of just sodium. Ranges of calcium input were estimated to be similar, with the Fynbos estimated inputs ranging from: 74 kg to 522 kg at either 4000 or 30000 years, and the Succulent Karoo: 166 kg to 1.25 tons . Table 5.7 shows the estimate for the other ions.

Table 5.7: Estimates of ion input by termites for one foraging hole, as an average per year (based off of estimates from targeted and stick-pile ion concentrations), projected for the estimated age of heuweltjies.

Ion	Biome	Average per year (g)	4000 years (Kg)	30000 years (Kg)
Sodium	Fynbos	~ 4	~ 16	~ 120
	Succulent Karoo	~ 45	~ 180	~ 1349
Calcium	Fynbos	~ 18	~ 74	~ 552
	Succulent Karoo	~ 42	~ 166	~ 1246
Magnesium	Fynbos	~ 4	~ 17	~ 125
	Succulent Karoo	~ 11	~ 42	~ 319
Phosphorus	Fynbos	~ 1	~ 3	~ 22
	Succulent Karoo	~ 2	~ 7	~ 55
Potassium	Fynbos	~ 10	~ 39	~ 291
	Succulent Karoo	~ 15	~ 60	~ 451
Aluminium	Fynbos	~ 2	~ 9	~ 70
	Succulent Karoo	~ 4	~ 15	~ 111
Boron	Fynbos	~ 5 x10^{-2}	~ 2 x10^{-1}	~ 1.4
	Succulent Karoo	~ 5 x10^{-2}	~ 0.2 x10^{-1}	~ 1.7
Copper	Fynbos	~ 9.7 x10^{-3}	~ 3.9 x10^{-2}	~ 0.3
	Succulent Karoo	~ 9.8 x10^{-3}	~ 3.9 x10^{-2}	~ 0.3
Iron	Fynbos	~ 3	~ 10	~ 74
	Succulent Karoo	~ 4	~ 15	~ 115
Manganese	Fynbos	~ 1.1	~ 4.6	~ 34.2
	Succulent Karoo	~ 0.4	~ 1.5	~ 11.1
Zinc	Fynbos	~ 2.7 x10^{-2}	~ 0.1	~ 0.8
	Succulent Karoo	~ 2.6 x10^{-2}	~ 0.1	~ 0.8

The importance of the role of termites in the functioning of heuweltjie mounds and contribution to salt accumulation has often just been assumed. These estimates are the first of its kind to give a quantifiable estimate of just how much of an impact termites can have on an ecosystem in terms of ion inputs from plant material. All of these estimates are for just 1 foraging hole per mound. The number of foraging holes per foraging event seen in this study ranged from 2-7 holes (over 4 events). For estimation purposes it will be assumed there are 5 foraging holes per heuweltjie. For the purposes of yearly estimations, this does not increase ion input too significantly. However, over the lifetime a heuweltjie is estimated to exist, upper input estimates of sodium and calcium would be over 5 tons per heuweltjie.

Calcium has been highlighted in the above results because of the calcrete plates associated with heuweltjies. The parent rock materials in the areas where heuweltjies and calcrete plates are found do not hold calcium, therefore the calcium needed to form this calcrete plate must come from external sources as shown above such as the plant tissues. As mentioned previously, the ion deposits on the external surface of plants are another viable source. Midgely et at. (2012) used similar data and calculated estimates of how long it could take for the calcrete layer to be created if the input concentration of Ca^{2+} only came from rainfall. Concentrations of 0.77 mg/l (=38.5 μeq/l) (Wyk et al., 1992) and 7 μeq/l (Soderberg and Compton, 2007) were used to determine how long it could take to form a calcrete plate of minimum thickness (0.25 m) using 600 mm of rainfall and deposition rates between 0.8 (based on Soderberg and Compton 2007) and 4.4 kg $ha^{-1}yr^{-1}$ (based on van Wyk et al., 1992) and found that it will take 3600 to 50500 years for calcrete plate to form at these inputs. This study found that the concentration of calcium on the surface of targeted plants is 3.66 mg/l in the Fynbos and 18.11 mg/l on the plants of the Succulent Karoo (Table 5.4). This can be further broken down into plant growing on the centre, targeted plants remaining on the centre and targeted plants growing off mound and being moved into the centres (Table 5.4). The Fynbos the input concentrations are around 4 times higher than the 0.77 mg/l determined by Midgely et al. (2012) and the Succulent Karoo concentrations are 20 times higher.

This suggests that the external salt input on the heuweltjie plants found in this study exists in a high enough concentration to be a viable source in the formation of the calcite layers, in less than 3600 years. Along with the contribution of the plant tissue calcium, the calcrete layer could possibly be created in a much shorter timeframe than has been previously suggested. This is quantitative information highlights how important the role is that termites have in concentrating ions into heuweltjie mounds soils.

Chapter 6: Summary

6.1 General Conclusions

Ion inputs from the external surfaces of plant communities, as well as internal tissue ions from plants growing in centre mound soils were identified and investigated. It was found that centre mound plants have high concentrations of sodium, magnesium, and potassium (Table 3.1) where they are also sources of calcium oxalate (whewellite ($CaC_2O_4 \cdot H_2O$)), pentahydrite ($MgSO_4.5H_20$), sylvite (KCl) and halite (NaCl) in both biomes.

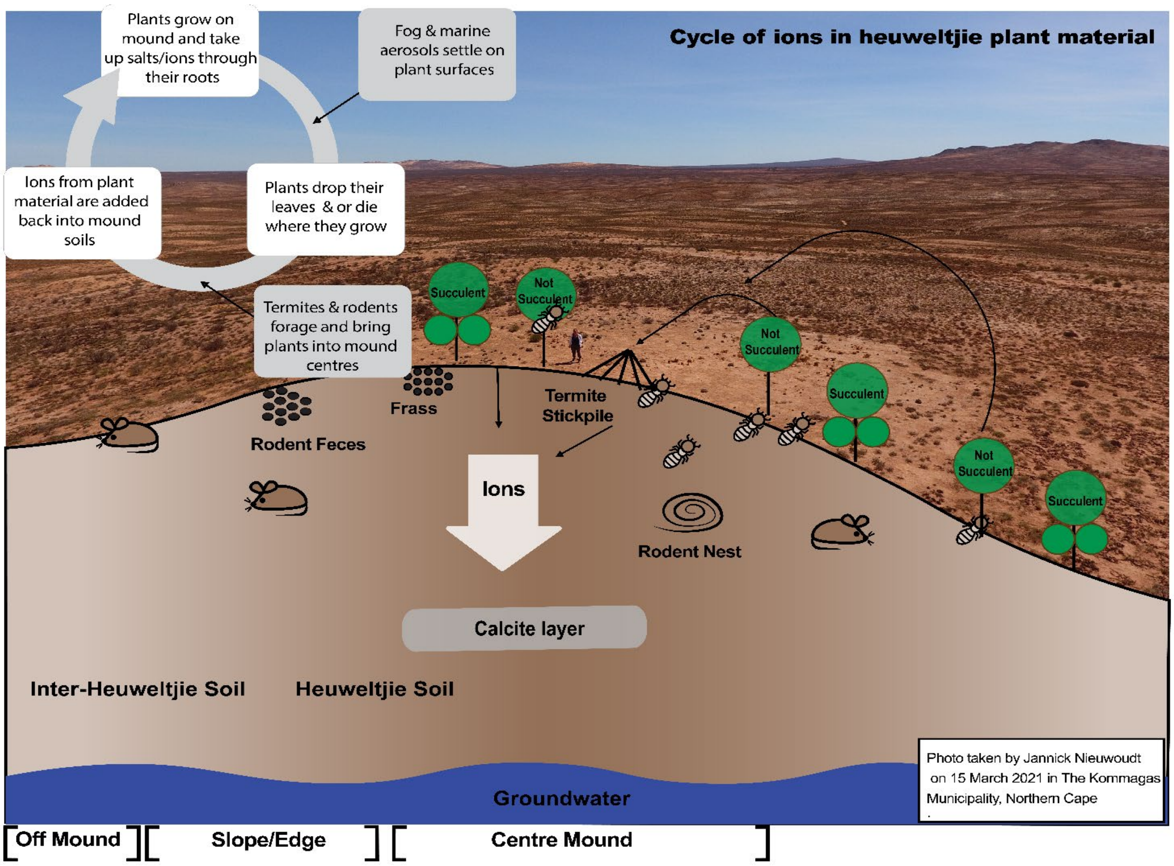

Figure 6.1: A simplified diagram of a heuweltjie ecosystem with the cycle of ions in heuweltjie plant material that was explored in this thesis.

These mound growing plants most likely create a feedback loop with the soils and help to maintain the heuweltjie centres as areas of higher salts, with unique plant communities in the landscape. Two external inputs into this feedback loop were identified 1) marine blown aerosols deposited on plant surfaces which exist in high enough concentrations to be a significant input into mound soils and 2) termites that bring plant material into their nests. Ions were found to exist in low concentrations in these plants but over time even these inputs could become a viable source of salt that can be used to explain the highly saline soils found on heuweltjie mounds. This cycle has been summarised and depicted in Figure 6.1. Dry deposition of marine aerosols on the land surface and plant material, is not a new idea and has been assumed by many previous studies. However, this is possibly the first work to provide conclusive evidence that dry deposition of marine aerosols on plant surfaces, most likely occurs across the entire western coastal system. This is because the results were consistent on all plant species, and across all the sampling locations, which ranged from the southern end of the western coast in the Fynbos Biome, to the northern end in the Succulent Karoo. The yearly estimates of ion input for both tissue inputs and the external plant washes would very likely change from year to year. Annual precipitation does change - with some years being very dry while some are very wet – and together with the presumably fluctuating size of a termite colony, will impact the amount of material being brought into mounds. As a result, the plant communities will change, and that in turn will alter the available ion pool that can be collected. Despite this, the hypothesis that the plants growing on heuweltjie mounds are the primary biological source of salts can be accepted. Furthermore, the activities of termites and rodent species living in the mound do indeed contribute significant amounts of ions into heuweltjie soils and can be considered a viable means of increasing salinity in heuweltjie centre mound soils.

6.2 Conservation implications

Research on heuweltjie vegetation has been undertaken in many previous studies. The results of this particular study add to the growing information on how these ecosystems function, by looking at heuweltjies in two different biomes. The worth of a descriptive study like this, is that it can exist as a baseline assessment of what heuweltjieveld should look like before a landscape becomes degraded or before land-use changes such as mining begin. Mining, particularly in the Succulent Karoo, has become a major economic activity (Francis et al., 2007). If restoration is needed, a study such as this can be referred back to in order to aid restoration efforts and understand what the ecosystem structure would need to be returned to. Grazing in both biomes has been shown to degrade heuweltjieveld. The plant functional type changes, between sites of differing disturbances, could also be used by managers as a fore-warning of what to look for to prevent their land from becoming degraded. Arid and semi-arid ecosystems in general, do not typically support very high biodiversity because of limiting resources such as water. However, heuweltjies are a feature known to enhance the biodiversity of many arid ecosystems, as they host unique plant and animal communities when compared with the surrounding off mound areas (Desmet, 2007; Francis et al., 2013; Knight et al., 1989; Kunz et al., 2012; McAuliffe et al., 2014; Midgley and Musil, 1990; Moore and Picker, 1991; Picker et al., 2007). Both the Succulent Karoo and Fynbos biomes are arid to semi-arid biomes recognised, with exceptionally high diversity, despite their aridity. The presence of heuweltjies should be considered an important feature contributing to the high levels of biodiversity. They should be conserved to maintain strong biodiversity links, which ultimately keep ecosystems more resilient to change.

The oxalate - carbonate pathway can cycle carbon from the atmosphere through an ecosystem into the soil where it is stored. Ecosystems where this pathway exists act as significant carbon sinks and long-term carbon storage. This means the oxalate - carbonate pathway could be important for atmospheric CO_2 reduction, which would help slow down the effect of climate change. If this process exists in heuweltjies, it will have significant conservation implications for south Africa's arid and semi-arid ecosystems. As currently they are not seen as exceptionally productive or important for carbon storage, due to the significant lack of surface plant biomass. This thesis shows that termites could form an important part of this pathway by bringing in calcium oxalate into mound soils through the plants. When heuweltjies are ploughed for agricultural use, termite colonies will most likely be removed or die off and their function as concentrators of ions will cease. Therefore, for heuweltjies to hold the possible role of carbon sinks in the landscape they will need to be conserved in their natural states. Lastly as mentioned in the beginning, many arid catchments are commonly affected by saline groundwater, which is influenced by the presence of heuweltjies. Groundwater is a precious resource for human communities in the area. Ultimately by understanding the role heuweltjie have as a part of the landscape and where and how the salts move into heuweltjie soils, people will be able to make better management decisions when putting water provisions in place. Saline heuweltjie soils have been found to be one of the contributors to the salinity of groundwater system, however this study suggests that accumulation and concentration of salts in heuweltjie soils is a natural process that can't be mitigated without risk of disturbing the structure and functions of heuweltjie ecosystems. Particularly in their role as hotspots of biodiversity or as potential carbon sinks. Alternative sources of water such as mist, or fog could provide the answer to supplying fresh water to human communities without the need to rely on groundwater. These non-rainfall moisture inputs such as fog or mist can be collected using simple set ups such as screens or mist nets with buckets beneath, and could provide a viable, less saline water alternative.

6.3 Recommendations for further work

Research on heuweltjie plant communities have mostly occurred at a small number of sites in one biome, in areas influenced by grazing, with variable results. I would recommend that a large-scale vegetation assessment would be useful to assess heuweltjie vegetation community findings to be able to identify vegetation patterns common to all heuweltjies. This should occur across multiple sites (more than two) in both the Fynbos and Succulent Karoo biomes in areas not influenced by grazing.

Another recommendation for future research would be to find and specifically analyse the bulbs of geophytic herbs found growing on heuweltjies. Plant species FB (*Chlorophytum triflorum*) was the only geophytic herb analysed for ions and isotopes in this study and was found to have no statistical differences in $\delta^{13}C$ to the rodent nests $\delta^{13}C$ (which had visible bulbs in it). Samples analysed in this plant species were also found to be sources of whewellite, halite and sylvite. A larger number of samples and analysis of multiple other species with bulbs would provide greater insight into the inputs of rodents to the ion cycle in heuweltjie plant material.

Due to the skittish nature of *M. viator* termite colonies, another recommendation would be to record more repeats of foraging observations and collections in all seasons of the year, to provide more accurate plant input rates. Secondly the use of less intrusive methods would be needed to understand exactly which species gets taken into mound soils, and if there are any sorting processes that occur. I would recommend video recording of an entire foraging session as well as the day afterwards. Lastly, I would recommend investigations to be conducted into the amount of precipitation that could be collected from non-rainfall precipitation sources, particularly in the more arid areas where heuweltjies can be found. This would allow the concentration of ions in marine aerosols deposits to be quantified more accurately and provide important information for human communities on the viability of mist and fog as an alternative water source.

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Appendix

Appendix 1: Figures of rank abundance curves

Appendix 2: Figures of box plots and accompanying t-tests comparing variables by biomes and subsites

Appendix 3: Boxplots of vegetation data grouped by zone – for site and sub-sites. Followed by r-code of Kruskal-Wallis tests and Pairwise comparisons using Wilcoxon rank sum test.

Appendix 4: Heuweltjie surface characteristics across biomes and zones

Appendix 5: Top 10 most common species by cover

Appendix 6: Photos of sampling locations on heuweltjies

Appendix 7: Plant species list

Appendix 8: Collected plant species list and photos

Appendix 9: Bar graphs of each ion showing the averages concentrations of each type of plant sample collected and r-code of statistical tests run.

Appendix 10: Boxplots and table of plant tissues ions groups by mound location and targeted plants

Appendix 11: XRD results showing mineral presence in all species

Appendix 12: Correlations: Surface washes

Appendix 13: Photos of foraging termites foraging

Appendix 14: Photos of rodent nest and faeces samples

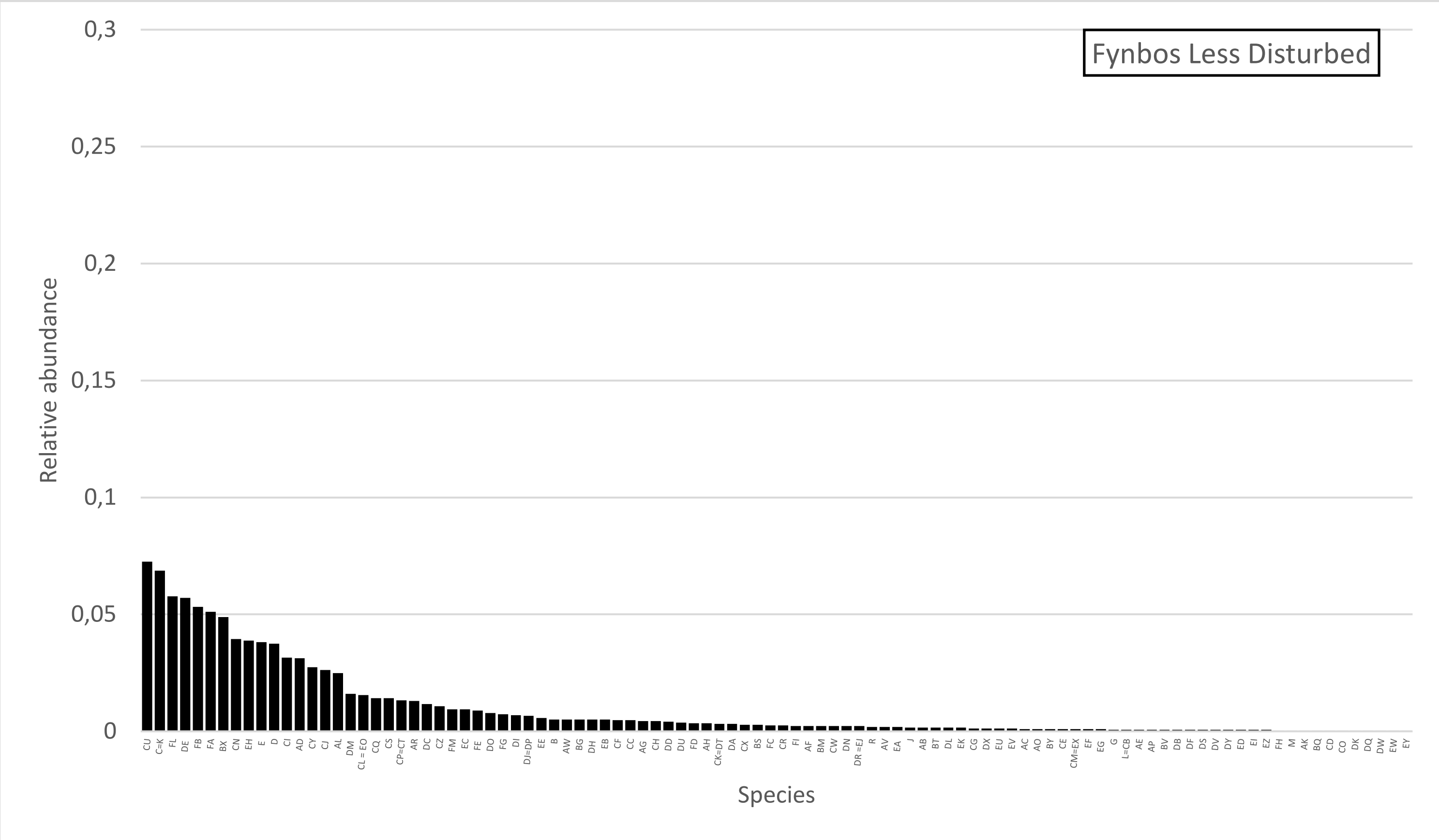
Appendix 15: Photos of different termite stick-piles

Appendix 16: Termite observations plant species list

Appendix 17: Diversity Variables pooled and averaged across the study sites

Appendix 18: Figure 3.1 without the plants with bladder cells

Appendix 1: Figures of Rank Abundance Curves



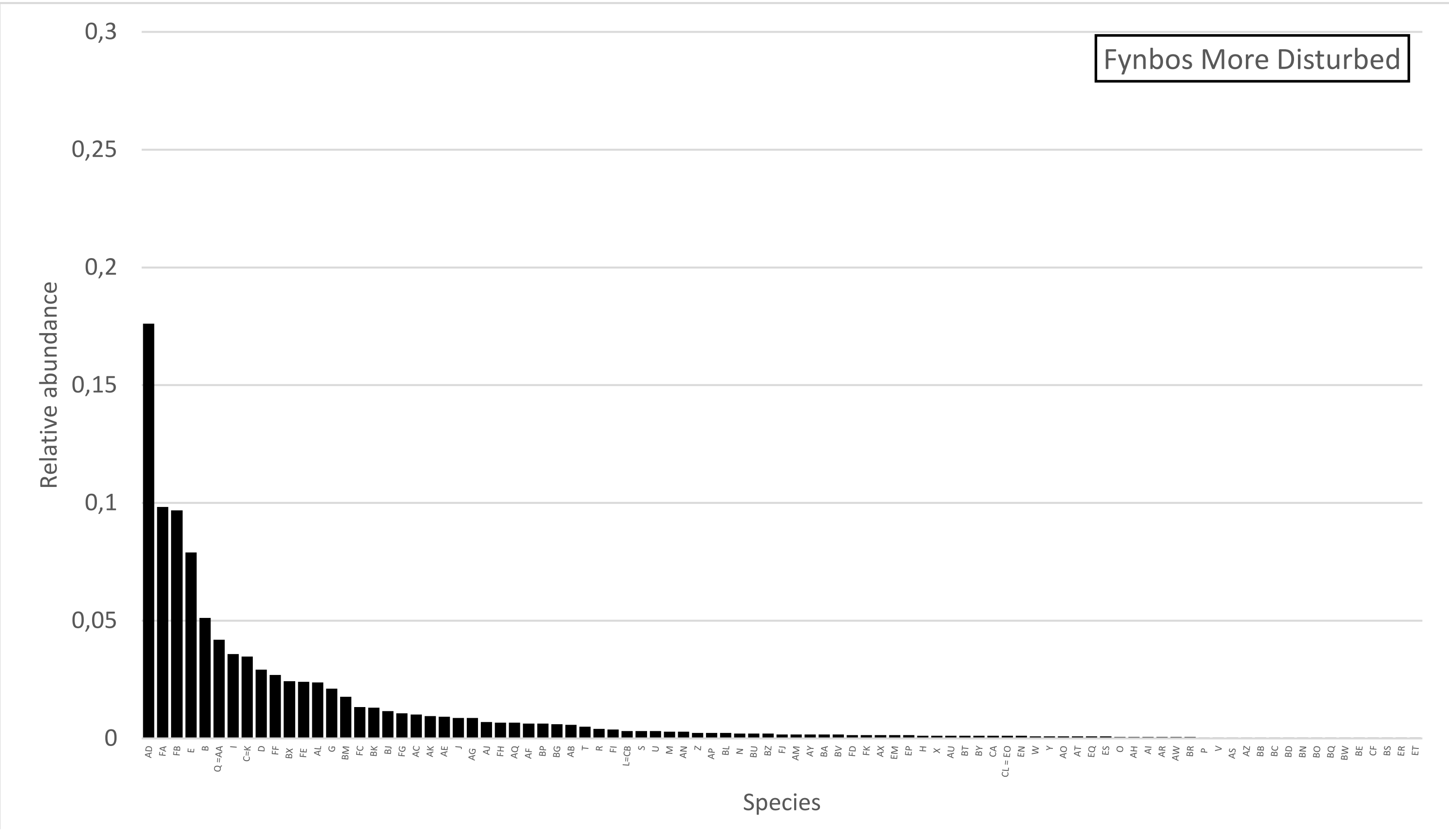


Figure G: Fynbos rank abundance curves

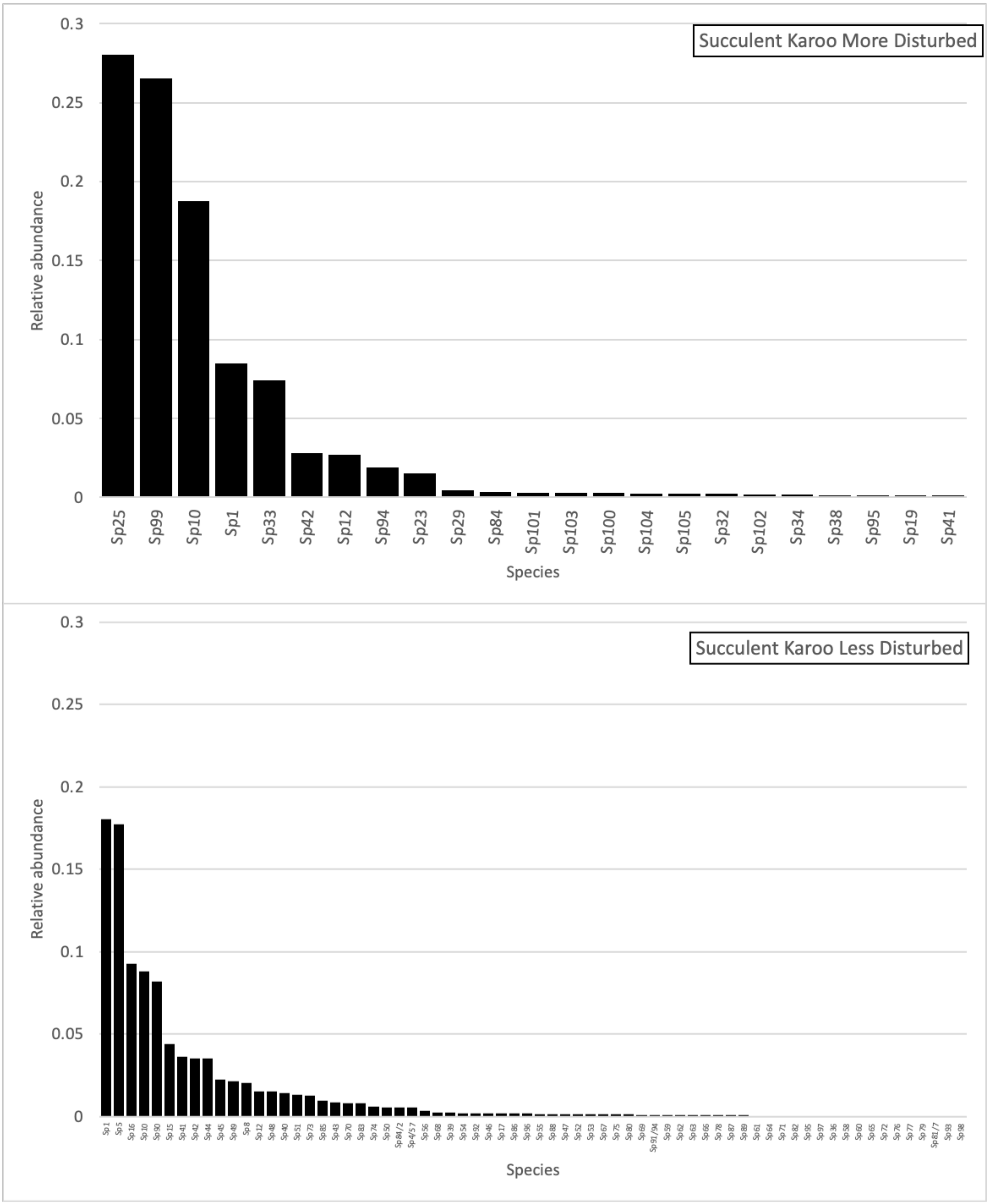


Figure HSucculent Karoo Rank abundance curves

Appendix 2: Figures of Box plots and accompanying t-tests comparing variables by biomes and subsites

Biomes

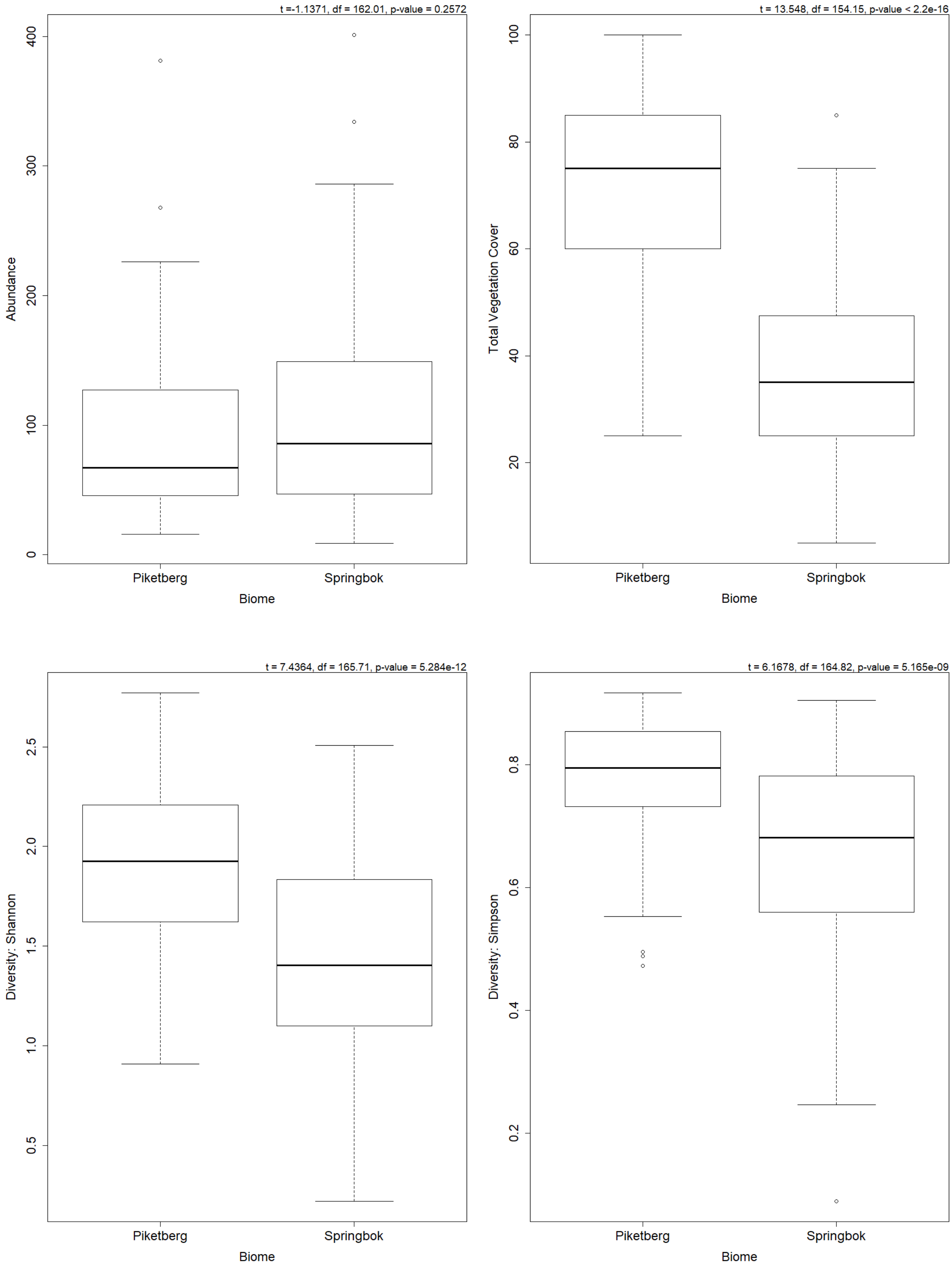


Figure E: Boxplots of abundance, vegetation cover, Simpsons and Shannon's diversity by biome

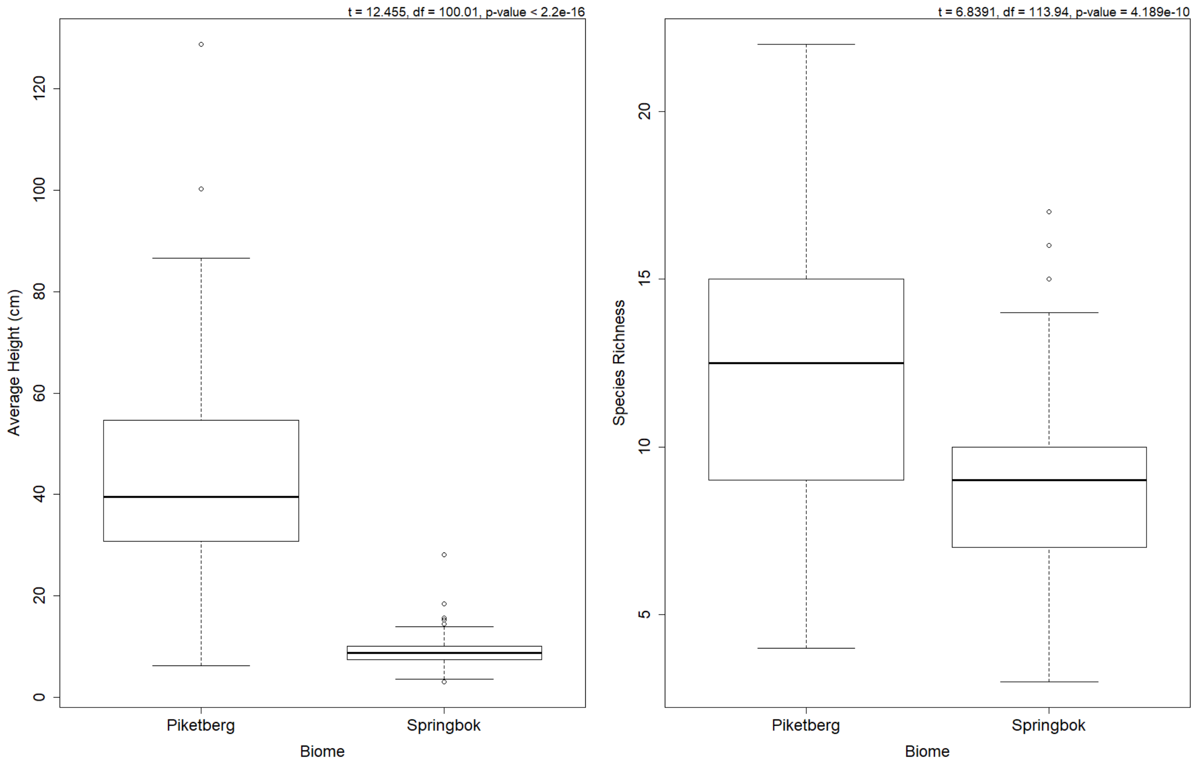


Figure F: Average Height and Specie richness by biome

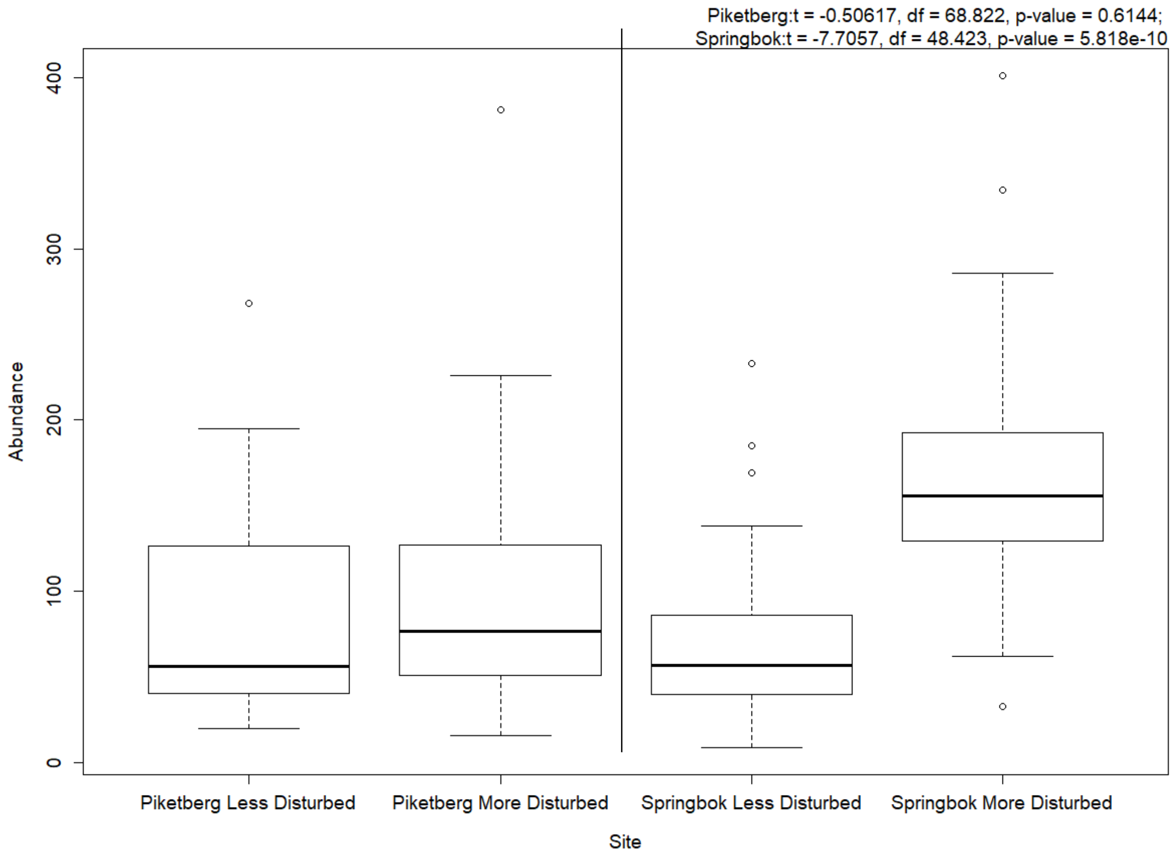
<u>R code – Welch Two Sample t-test by biome</u>

```
> t.test(Total.Vegetation.cover~Biome,data=vegstats)#t = 13.548, df = 154.15, p-
value < 2.2e-16

        Welch Two Sample t-test

data: Total.Vegetation.cover by Biome
t = 13.548, df = 154.15, p-value < 2.2e-16
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 30.23309 40.55479
sample estimates:
mean in group Piketberg mean in group Springbok
               72.77778                37.38384
> t.test(Average.height~Biome,data=vegstats)#t = 12.455, df = 100.01, p-value <
2.2e-16

        Welch Two Sample t-test

data:  Average.height by Biome
t = 14.005, df = 73.586, p-value < 2.2e-16
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 30.11111 40.10182
sample estimates:
mean in group Piketberg mean in group Springbok
              44.370953                9.264489


> t.test(Abundance~Biome,data=vegstats)#t = -1.0391, df = 162.02, p-value = 0.30
03

        Welch Two Sample t-test

data:  Abundance by Biome
t = -1.1371, df = 162.01, p-value = 0.2572
alternative hypothesis: true difference in means is not equal to 0
```

```
95 percent confidence interval:
 -33.484721   9.012499
sample estimates:
mean in group Piketberg mean in group Springbok
               92.09722               104.33333

> t.test(Abundance.m2~Biome,data=vegstats)

        Welch Two Sample t-test

data:  Abundance.m2 by Biome
t = -1.1371, df = 162.01, p-value = 0.2572
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 -8.371180  2.253125
sample estimates:
mean in group Piketberg mean in group Springbok
               23.02431                26.08333

> t.test(Species.richness~Biome,data=vegstats)#t = 6.8391, df = 113.94, p-value
= 4.189e-10

        Welch Two Sample t-test

data:  Species.richness by Biome
t = 6.8391, df = 113.94, p-value = 4.189e-10
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 2.806380 5.095135
sample estimates:
mean in group Piketberg mean in group Springbok
              12.486111                8.535354
> t.test(Species.richness.m2~Biome,data=vegstats)

        Welch Two Sample t-test

data:  Species.richness.m2 by Biome
t = 6.8391, df = 113.94, p-value = 4.189e-10
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 0.701595 1.273784
sample estimates:
mean in group Piketberg mean in group Springbok
               3.121528                2.133838

> t.test(Diversity.Shannon~Biome,data=vegstats)#t = 7.4364, df = 165.71, p-value
= 5.284e-12

        Welch Two Sample t-test

data:  Diversity.Shannon by Biome
t = 7.3765, df = 165.3, p-value = 7.474e-12
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 0.3443387 0.5960443
sample estimates:
mean in group Piketberg mean in group Springbok
               1.899515                1.429324


> t.test(Diversity.Simpson~Biome,data=vegstats)#t = 6.1678, df = 164.82, p-value
= 5.165e-09

        Welch Two Sample t-test

data:  Diversity.Simpson by Biome
t = 6.1447, df = 164.99, p-value = 5.803e-09
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 0.08458482 0.16467935
sample estimates:
mean in group Piketberg mean in group Springbok
```

```
              0.7767308                 0.6520988

> t.test(Pielous.eveness~Biome,data=vegstats)#

        Welch Two Sample t-test

data:  Pielous.eveness by Biome
t = 4.5214, df = 168.59, p-value = 1.153e-05
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 0.05447799 0.13891763
sample estimates:
mean in group Piketberg mean in group Springbok
              0.7752403                 0.6785424
```

Box plots and t-tests comparing variables by subsites



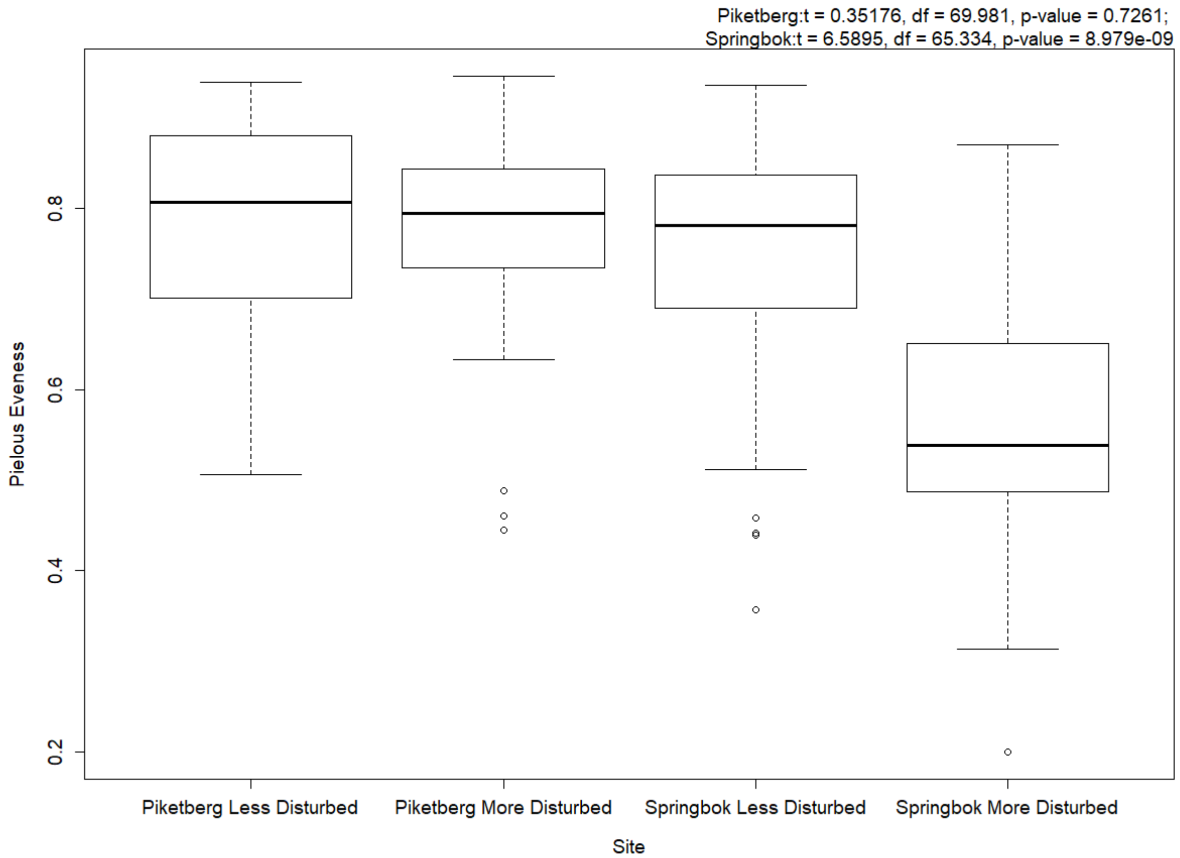
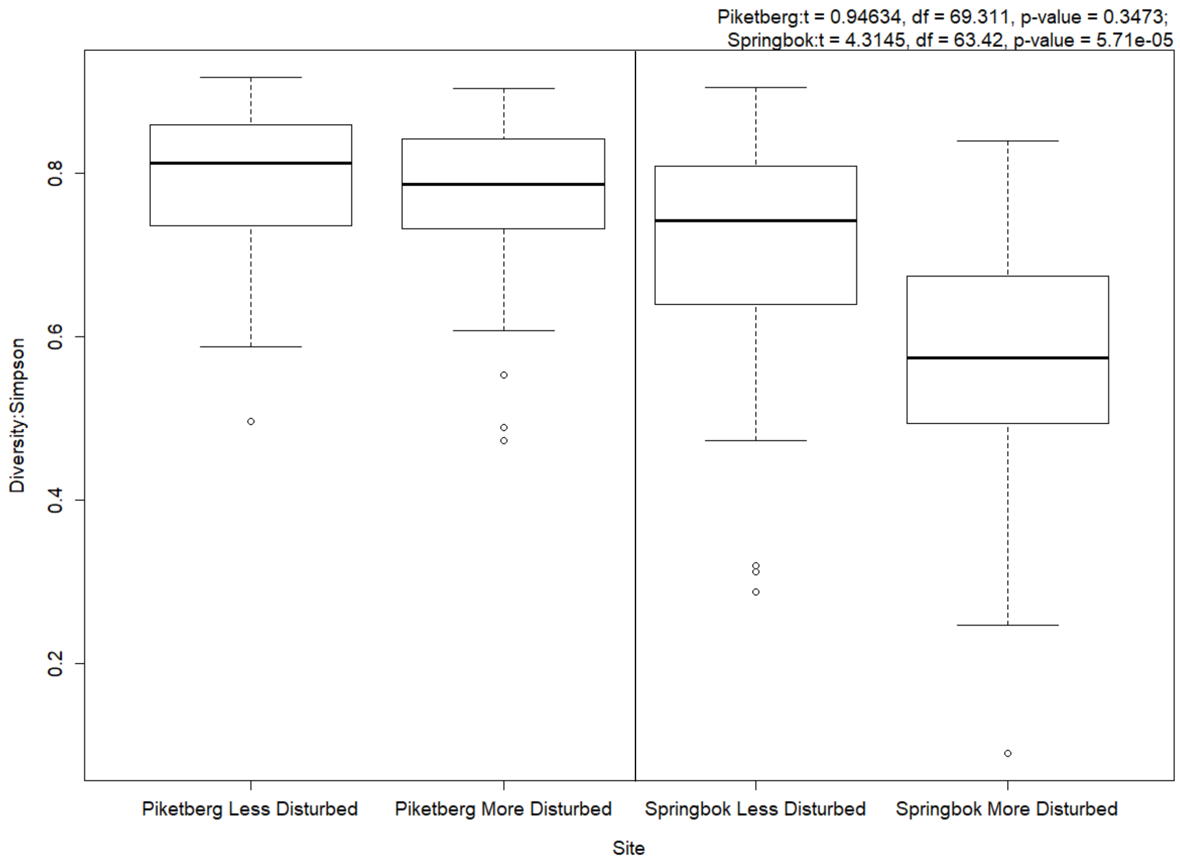


Figure G: Boxplots of Abundance and Evenness by Subsite



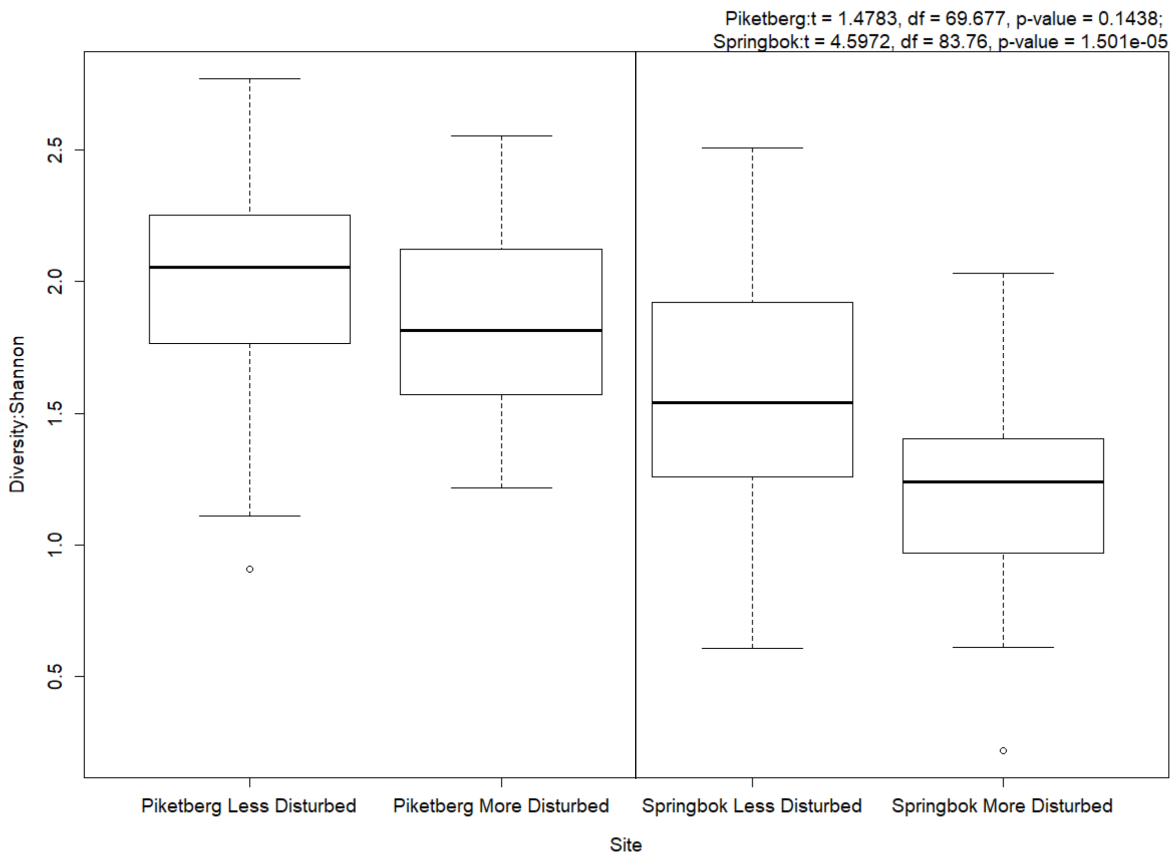
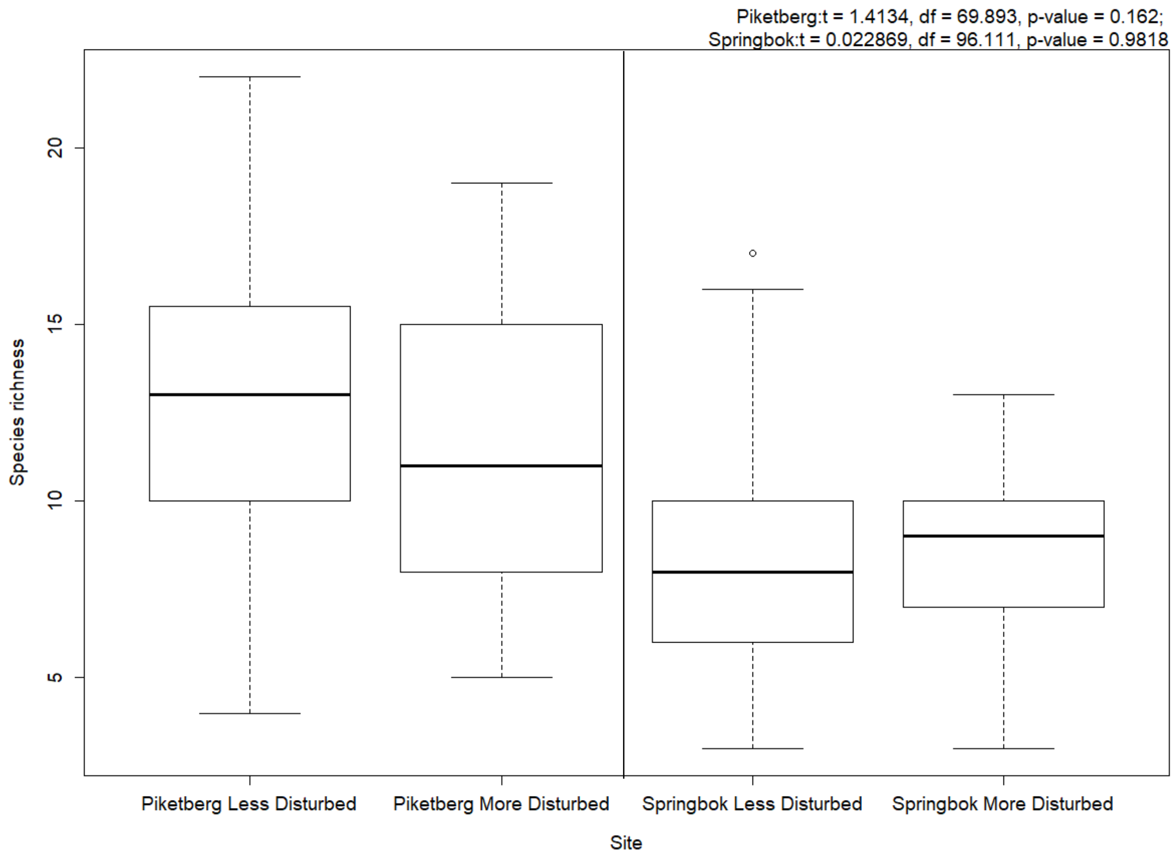


Figure H: Boxplots of Shannon and Simpsons diversity by Subsite



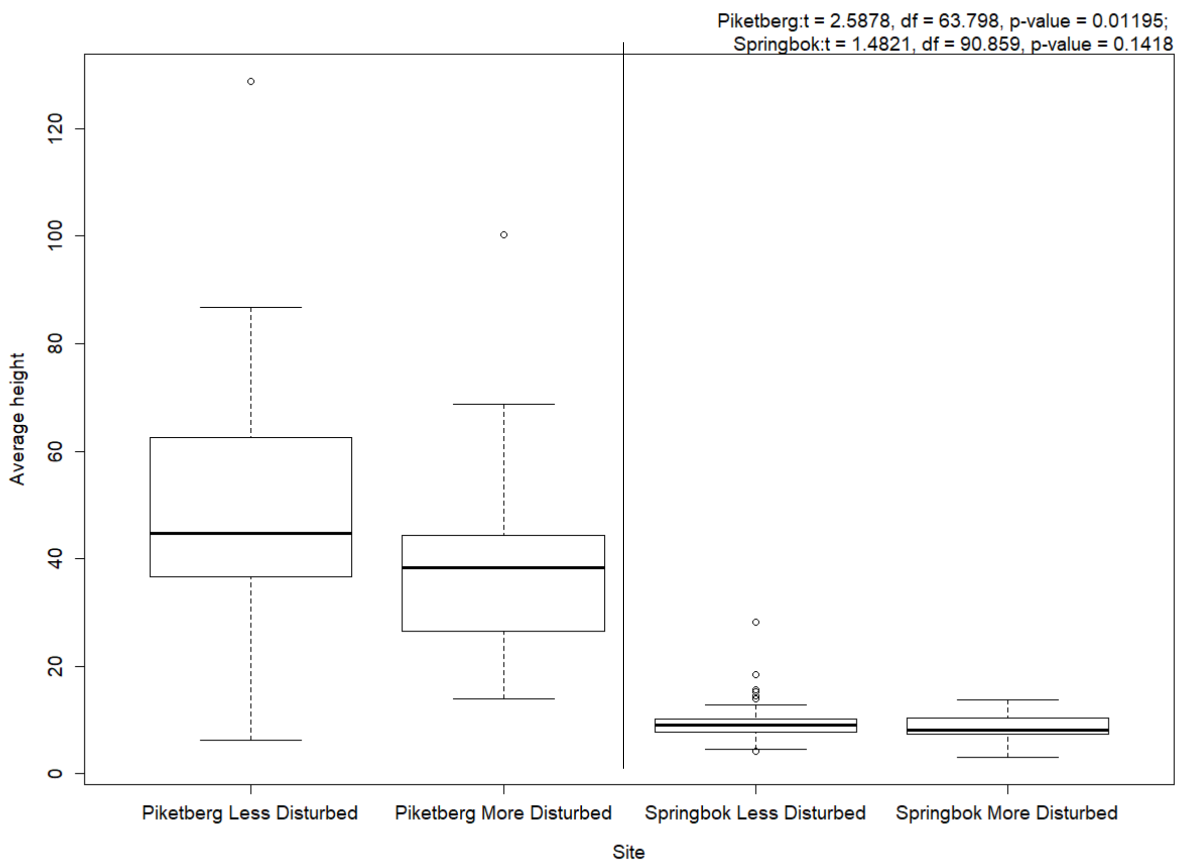


Figure I: Boxplots of species richness and average plant height by subsite

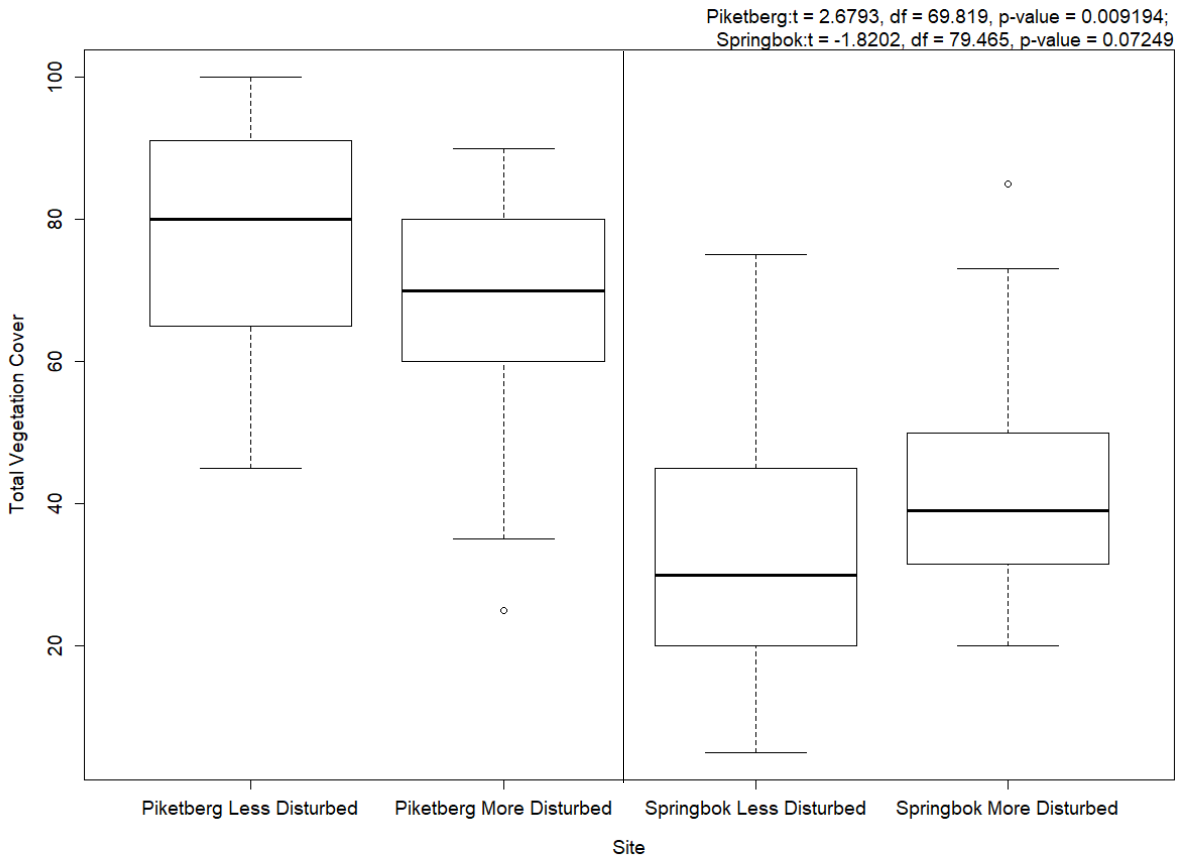


Figure J: Boxplots of Total vegetation cover by subsite

R code – Welch Two Sample t-test by Subsite(disturbance)

```
> #disturbed
> t.test(Total.Vegetation.cover~Site,data=Piketbergdata)

        Welch Two Sample t-test

data:  Total.Vegetation.cover by Site
t = 2.6793, df = 69.819, p-value = 0.009194
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
  2.598395 17.734939
sample estimates:
mean in group Piketberg Less Disturbed mean in group Piketberg More Disturbed
                              77.86111                               67.69444

> t.test(Total.Vegetation.cover~Site,data=Springbokdata)

        Welch Two Sample t-test

data:  Total.Vegetation.cover by Site
t = -1.8202, df = 79.465, p-value = 0.07249
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 -12.9924659   0.5797675
sample estimates:
mean in group Springbok Less Disturbed mean in group Springbok More Disturbed
                              35.12698                               41.33333


> #disturbed
> t.test(Average.height~Site,data=Piketbergdata)
```

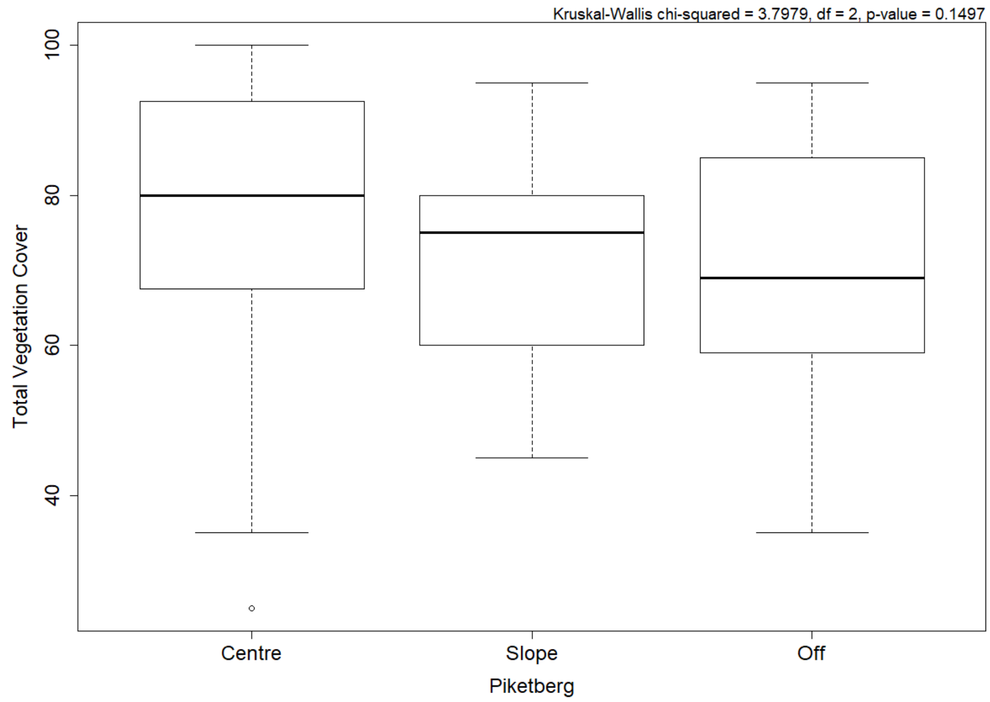
```
        Welch Two Sample t-test

data:  Average.height by Site
t = 2.5878, df = 63.798, p-value = 0.01195
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
  2.820485 21.922143
sample estimates:
mean in group Piketberg Less Disturbed mean in group Piketberg More Disturbed
                              50.55661                               38.18530

> t.test(Average.height~Site,data=Springbokdata)

        Welch Two Sample t-test

data:  Average.height by Site
t = 1.4821, df = 90.859, p-value = 0.1418
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 -0.3218541  2.2137041
sample estimates:
mean in group Springbok Less Disturbed mean in group Springbok More Disturbed
                              9.608461                               8.662536

> #disturbed
> t.test(Abundance.m2~Site,data=Piketbergdata)

        Welch Two Sample t-test

data:  Abundance.m2 by Site
t = -0.50617, df = 68.822, p-value = 0.6144
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 -9.779967  5.821634
sample estimates:
mean in group Piketberg Less Disturbed mean in group Piketberg More Disturbed
                              22.03472                               24.01389

> t.test(Abundance.m2~Site,data=Springbokdata)

        Welch Two Sample t-test

data:  Abundance.m2 by Site
t = -7.7057, df = 48.423, p-value = 5.818e-10
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 -32.26602 -18.91454
sample estimates:
mean in group Springbok Less Disturbed mean in group Springbok More Disturbed
                              16.77778                               42.36806

> #disturbed
> t.test(Species.richness.m2~Site,data=Piketbergdata)

        Welch Two Sample t-test

data:  Species.richness.m2 by Site
t = 1.4134, df = 69.893, p-value = 0.162
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 -0.1456143  0.8539477
sample estimates:
mean in group Piketberg Less Disturbed mean in group Piketberg More Disturbed
                              3.298611                               2.944444

> t.test(Species.richness.m2~Site,data=Springbokdata)

        Welch Two Sample t-test

data:  Species.richness.m2 by Site
t = 0.022869, df = 96.111, p-value = 0.9818
```

```
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 -0.2553466  0.2612990
sample estimates:
mean in group Springbok Less Disturbed mean in group Springbok More Disturbed
                              2.134921                               2.131944


> #disturbed
> t.test(Diversity.Shannon~Site,data=Piketbergdata)

        Welch Two Sample t-test

data:  Diversity.Shannon by Site
t = 1.4783, df = 69.677, p-value = 0.1438
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 -0.04587801  0.30861795
sample estimates:
mean in group Piketberg Less Disturbed mean in group Piketberg More Disturbed
                               1.96520                                1.83383

> t.test(Diversity.Shannon~Site,data=Springbokdata)

        Welch Two Sample t-test

data:  Diversity.Shannon by Site
t = 4.5972, df = 83.76, p-value = 1.501e-05
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 0.2152140 0.5433645
sample estimates:
mean in group Springbok Less Disturbed mean in group Springbok More Disturbed
                              1.567247                               1.187958

> #disturbed
> t.test(Diversity.Simpson~Site,data=Piketbergdata)

        Welch Two Sample t-test

data:  Diversity.Simpson by Site
t = 0.94634, df = 69.311, p-value = 0.3473
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 -0.02493407  0.06994529
sample estimates:
mean in group Piketberg Less Disturbed mean in group Piketberg More Disturbed
                             0.7879837                              0.7654780

> t.test(Diversity.Simpson~Site,data=Springbokdata)

        Welch Two Sample t-test

data:  Diversity.Simpson by Site
t = 4.3145, df = 63.42, p-value = 5.71e-05
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 0.07572446 0.20636137
sample estimates:
mean in group Springbok Less Disturbed mean in group Springbok More Disturbed
                             0.7033871                              0.5623442

> #disturbed
> t.test(Pielous.eveness~Site,data=Piketbergdata)

        Welch Two Sample t-test

data:  Pielous.eveness by Site
t = 0.35176, df = 69.981, p-value = 0.7261
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
```

```
 -0.04448191  0.06353215
sample estimates:
mean in group Piketberg Less Disturbed mean in group Piketberg More Disturbed
                             0.7800028                              0.7704777

> t.test(Pielous.eveness~Site,data=Springbokdata)

        Welch Two Sample t-test

data:  Pielous.eveness by Site
t = 6.5895, df = 65.334, p-value = 8.979e-09
alternative hypothesis: true difference in means is not equal to 0
95 percent confidence interval:
 0.1359076 0.2540987
sample estimates:
mean in group Springbok Less Disturbed mean in group Springbok More Disturbed
                             0.7494527                              0.5544495


>
```

Appendix 3: Boxplots of Vegetation data grouped by Zone – for Site and Sub-Sites- followed by r-code of Kruskal-Wallis tests and Pairwise comparisons using Wilcoxon rank sum test.

Total Vegetation cover



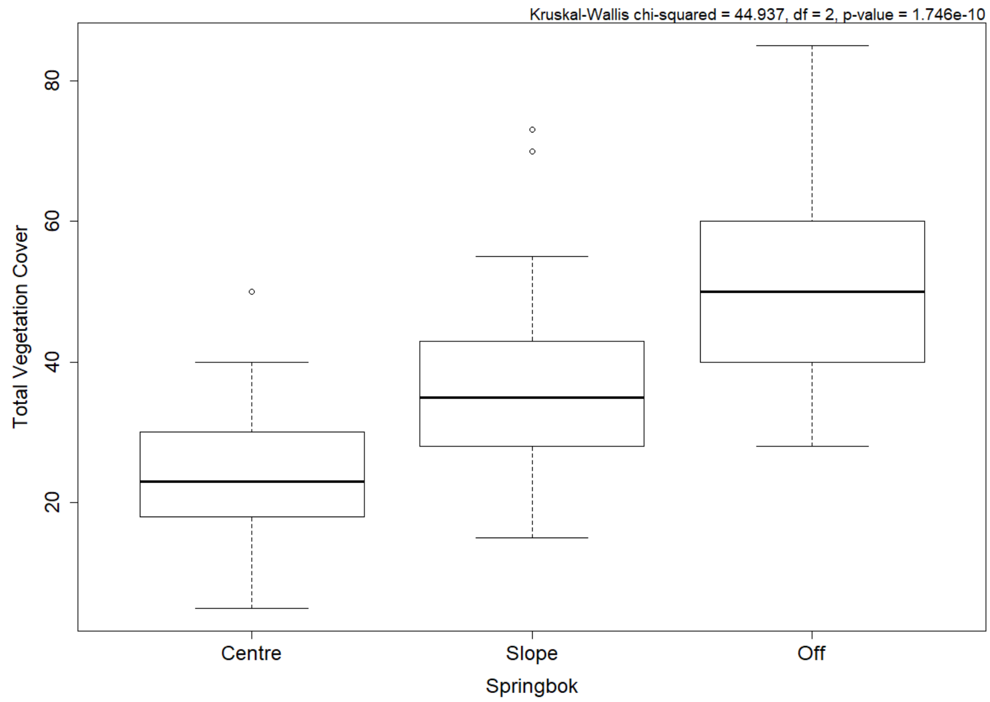


Figure K: Boxplots of vegetation cover by zone in each biome

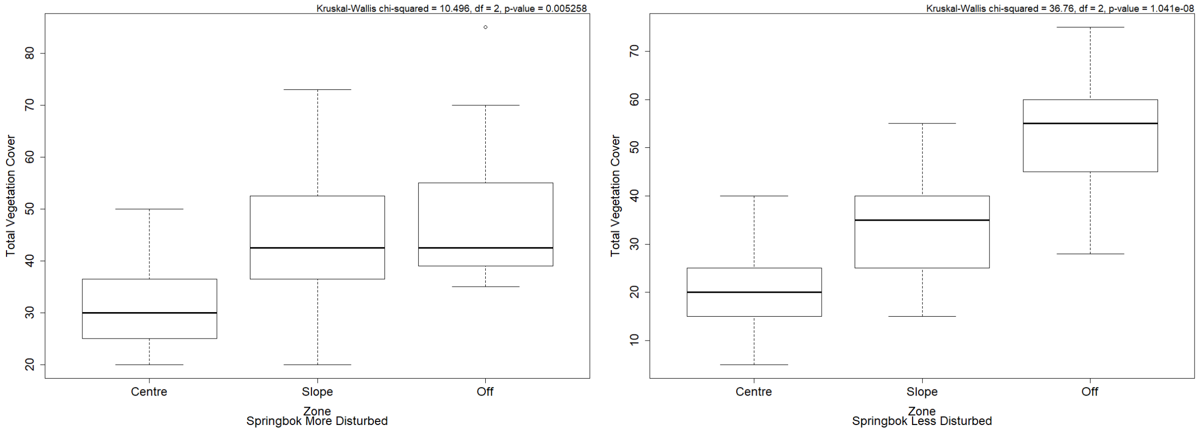
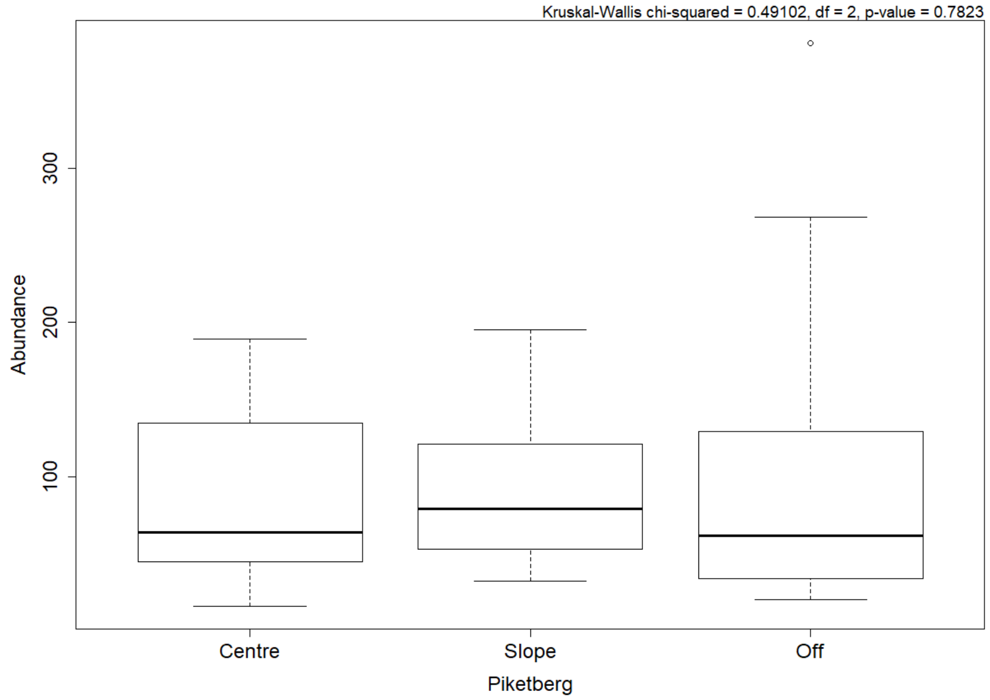


Figure L: Boxplots of total vegetation cover by zone in each subsite with significant differences

```
> kruskal.test(Total.Vegetation.cover~Zone,data=vegstats,subset=Site=="Piketbe
rg Less Disturbed")#Kruskal-Wallis chi-squared = 5.5325, df = 2, p-value = 0.0
629

	Kruskal-Wallis rank sum test

data:  Total.Vegetation.cover by Zone
Kruskal-Wallis chi-squared = 5.5325, df = 2, p-value = 0.0629

> kruskal.test(Total.Vegetation.cover~Zone,data=vegstats,subset=Site=="Piketbe
rg More Disturbed")#Kruskal-Wallis chi-squared = 1.3321, df = 2, p-value = 0.5
137

	Kruskal-Wallis rank sum test

data:  Total.Vegetation.cover by Zone
Kruskal-Wallis chi-squared = 1.3321, df = 2, p-value = 0.5137



Pairwise comparisons using Wilcoxon rank sum test

data:  Springbokdata$Total.Vegetation.cover and Springbokdata$Zone 
      Centre  Off    
Off   1e-09   -      
Slope 0.00046 0.00014


data:  SpringbokMD$Total.Vegetation.cover and SpringbokMD$Zone 
      Centre Off   
Off   0.0038 -     
Slope 0.0636 0.5810


data:  SpringbokLD$Total.Vegetation.cover and SpringbokLD$Zone 
      Centre  Off    
Off   3.8e-07 -      
Slope 0.0017  4.0e-05
```

Abundance



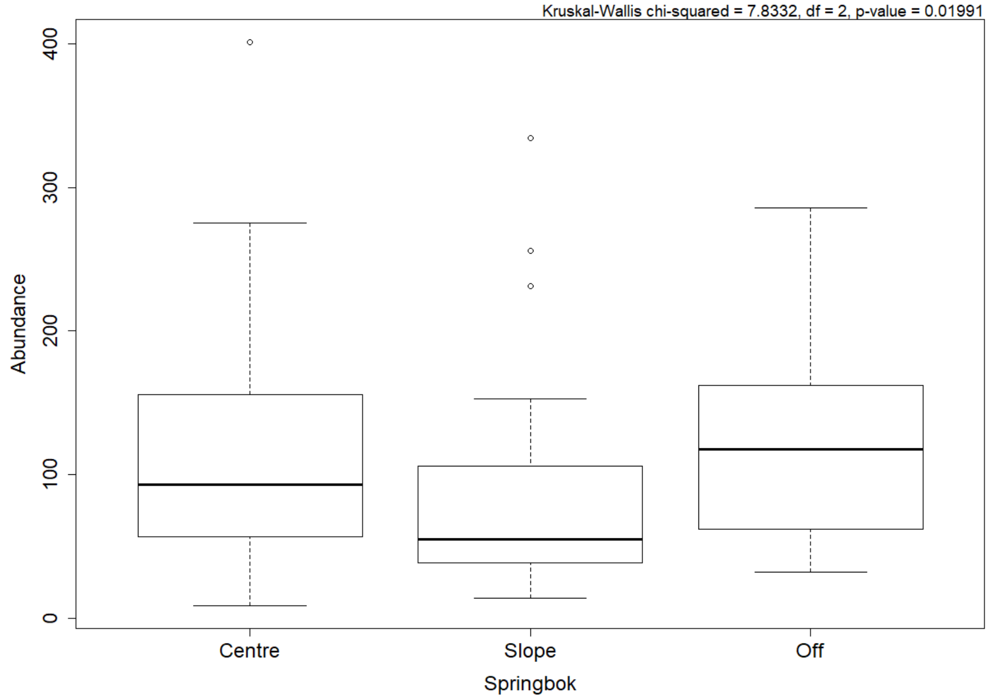


Figure M: Boxplots of abundance by zone in each biome

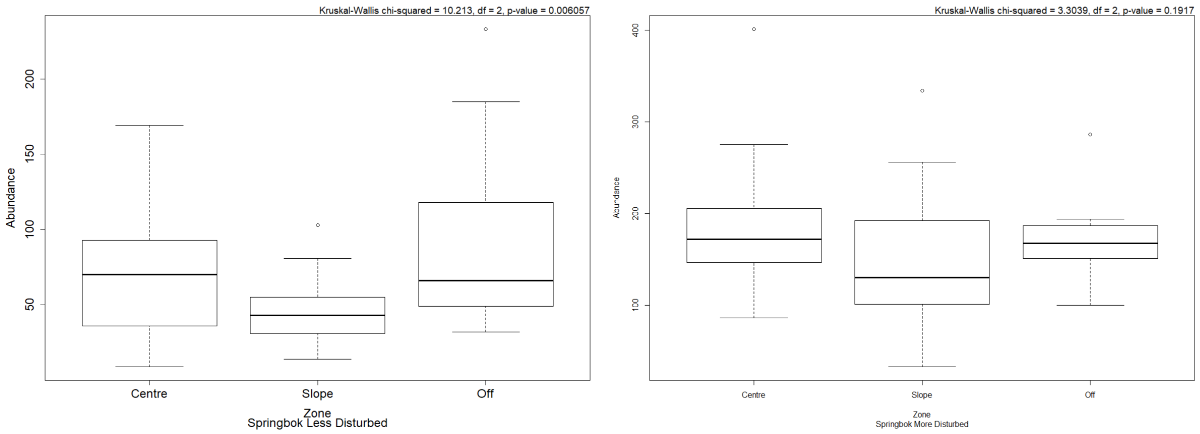


Figure N: Boxplots of abundance by zone in each subsite with significant differences

```
> kruskal.test(Abundance~Zone,data=vegstats,subset=Site=="Piketberg Less Distu
rbed")#Kruskal-Wallis chi-squared = 4.0959, df = 2, p-value = 0.129

        Kruskal-Wallis rank sum test

data:  Abundance by Zone
Kruskal-Wallis chi-squared = 4.0959, df = 2, p-value = 0.129


> kruskal.test(Abundance~Zone,data=vegstats,subset=Site=="Piketberg More Distu
rbed")#Kruskal-Wallis chi-squared = 1.4844, df = 2, p-value = 0.4761

        Kruskal-Wallis rank sum test

data:  Abundance by Zone
Kruskal-Wallis chi-squared = 1.4844, df = 2, p-value = 0.4761


Pairwise comparisons using Wilcoxon rank sum test

data:  Springbokdata$Abundance and Springbokdata$Zone
      Centre Off
Off   0.577  -
Slope 0.089  0.021


data:  SpringbokMD$Abundance and SpringbokMD$Zone
      Centre Off
Off   0.84   -
Slope 0.36   0.36


data:  SpringbokLD$Abundance and SpringbokLD$Zone
      Centre Off
Off   0.4428 -
Slope 0.0519 0.0062
```

Simpsons Diversity (D)

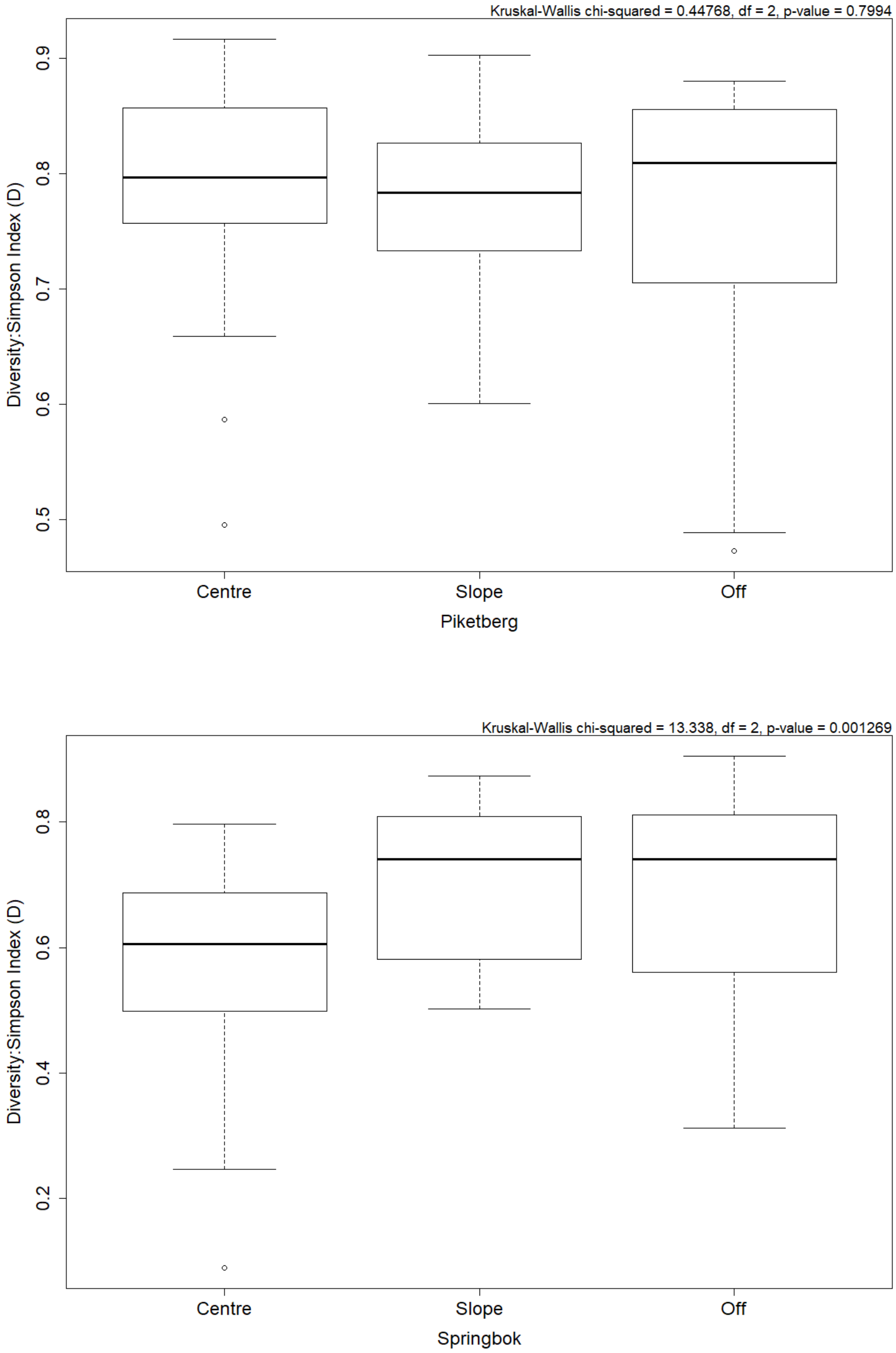


Figure O: Boxplots of Simpson's diversity by zone in each biome.

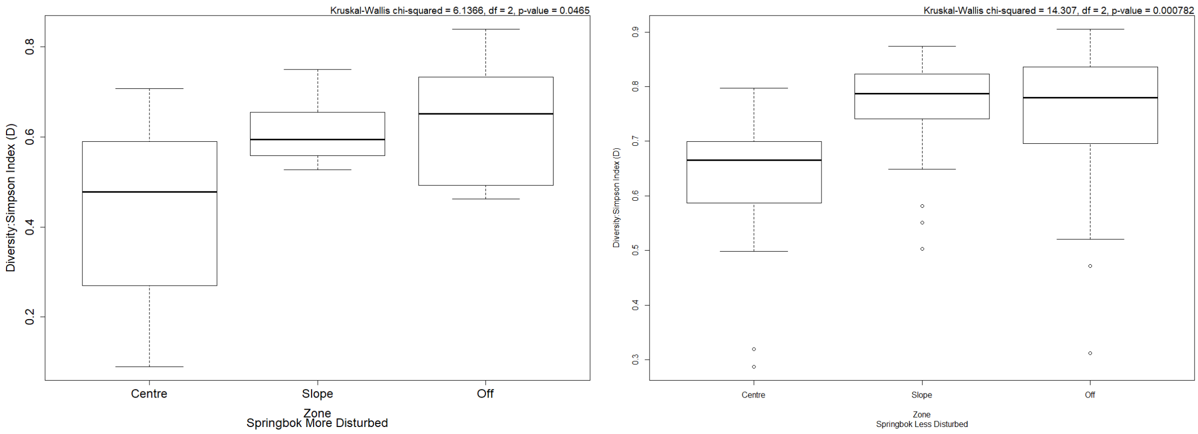
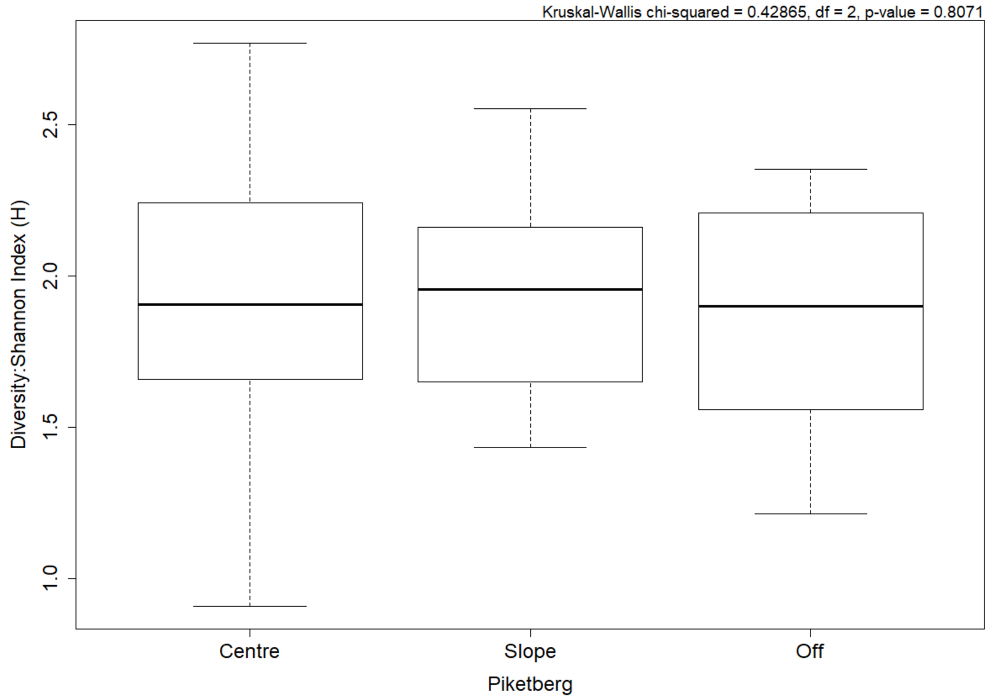


Figure P: Boxplots of Simpson's diversity by zone in each subsite with significant differences.

```
> kruskal.test(Diversity.Simpson~Zone,data=vegstats,subset=Site=="Piketberg Le
ss Disturbed")#Kruskal-Wallis chi-squared = 0.7988, df = 2, p-value = 0.6707

        Kruskal-Wallis rank sum test

data:  Diversity.Simpson by Zone
Kruskal-Wallis chi-squared = 0.9024, df = 2, p-value = 0.6369

> kruskal.test(Diversity.Simpson~Zone,data=vegstats,subset=Site=="Piketberg Mo
re Disturbed")#Kruskal-Wallis chi-squared = 1.2716, df = 2, p-value = 0.5295

        Kruskal-Wallis rank sum test

data:  Diversity.Simpson by Zone
Kruskal-Wallis chi-squared = 1.8213, df = 2, p-value = 0.4023
```

```
Pairwise comparisons using Wilcoxon rank sum test

data:  Springbokdata$Diversity.Simpson and Springbokdata$Zone
      Centre Off
Off   0.0033 -
Slope 0.0033 0.9949

data:  SpringbokMD$Diversity.Simpson and SpringbokMD$Zone
      Centre Off
Off   0.073  -
Slope 0.104  0.713

data:  SpringbokLD$Diversity.Simpson and SpringbokLD$Zone
      Centre  Off
Off   0.00680 -
Slope 0.00052 0.83068
```

Shannon Diversity (H)



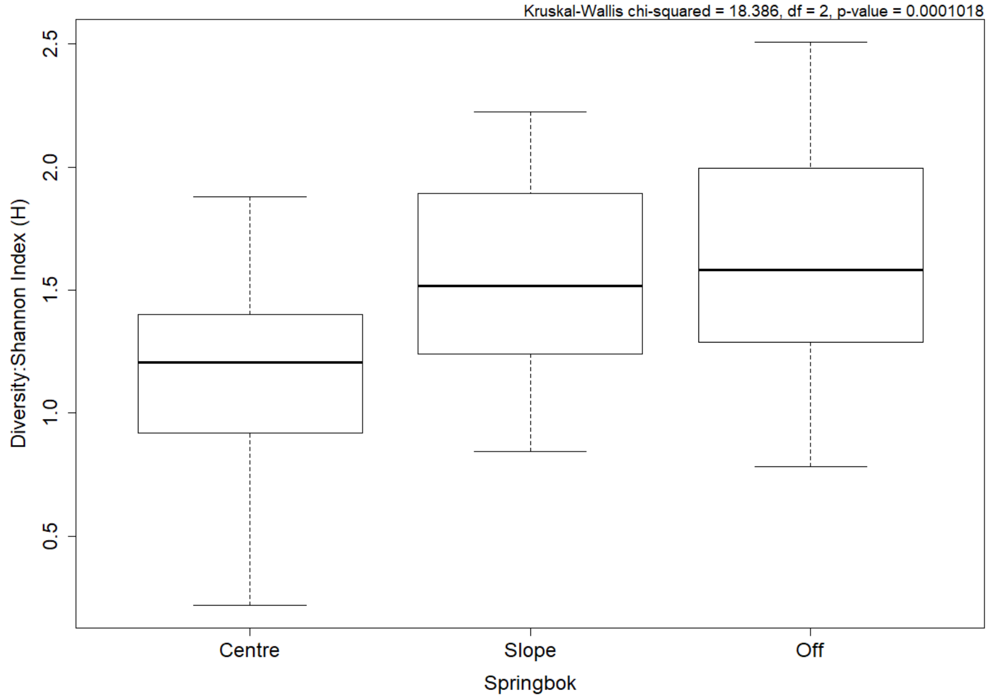


Figure Q: Boxplots of Shannon's diversity by zone in each biome.

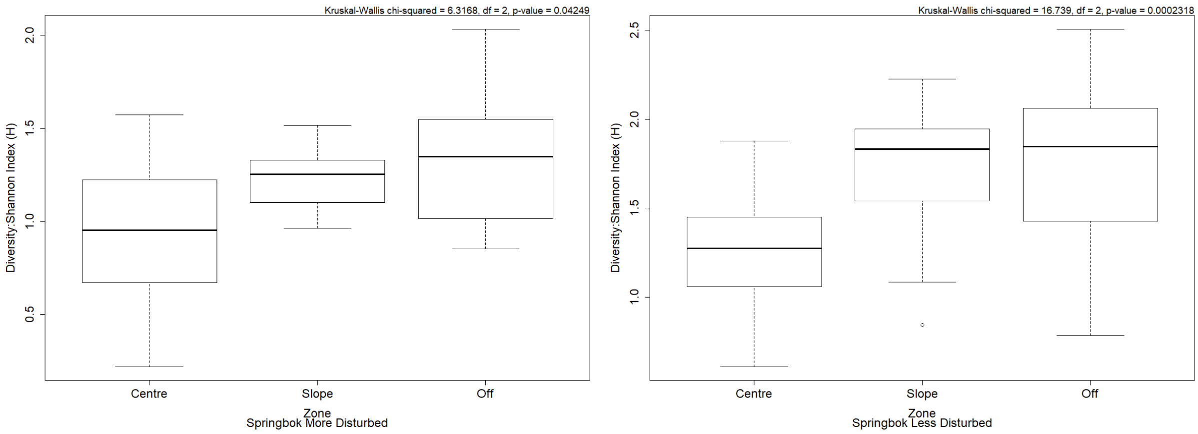
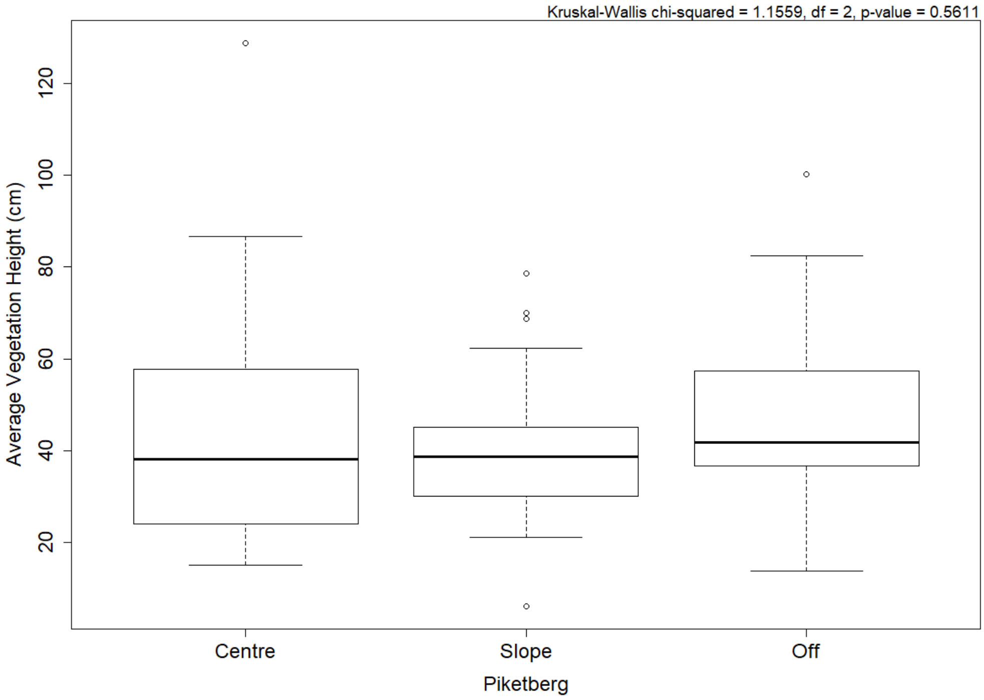


Figure R: Boxplots of Simpson's diversity by zone in each subsite with significant differences.

```
> kruskal.test(Diversity.Shannon~Zone,data=vegstats,subset=Site=="Piketberg Le
ss Disturbed")#Kruskal-Wallis chi-squared = 0.073574, df = 2, p-value = 0.9639

        Kruskal-Wallis rank sum test

data:  Diversity.Shannon by Zone
Kruskal-Wallis chi-squared = 0.046547, df = 2, p-value = 0.977

> kruskal.test(Diversity.Shannon~Zone,data=vegstats,subset=Site=="Piketberg Mo
re Disturbed")#Kruskal-Wallis chi-squared = 1.2716, df = 2, p-value = 0.5295

        Kruskal-Wallis rank sum test

data:  Diversity.Shannon by Zone
Kruskal-Wallis chi-squared = 1.2716, df = 2, p-value = 0.5295



Pairwise comparisons using Wilcoxon rank sum test

data:  Springbokdata$Diversity.Shannon and Springbokdata$Zone
      Centre  Off
Off   0.00026 -
Slope 0.00061 0.50736

data:  SpringbokMD$Diversity.Shannon and SpringbokMD$Zone
      Centre Off
Off   0.085  -
Slope 0.085  0.630

data:  SpringbokLD$Diversity.Shannon and SpringbokLD$Zone
      Centre  Off
Off   0.00066 -
Slope 0.00066 0.67208
```

Vegetation Height



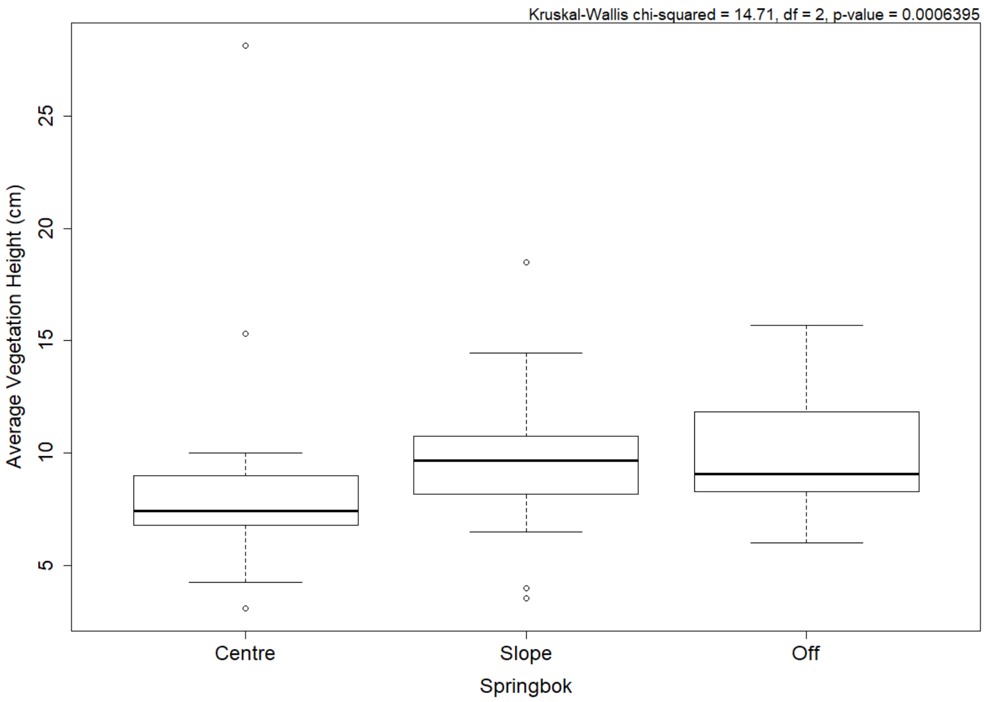


Figure S: Boxplots of vegetation height by zone in each biome.

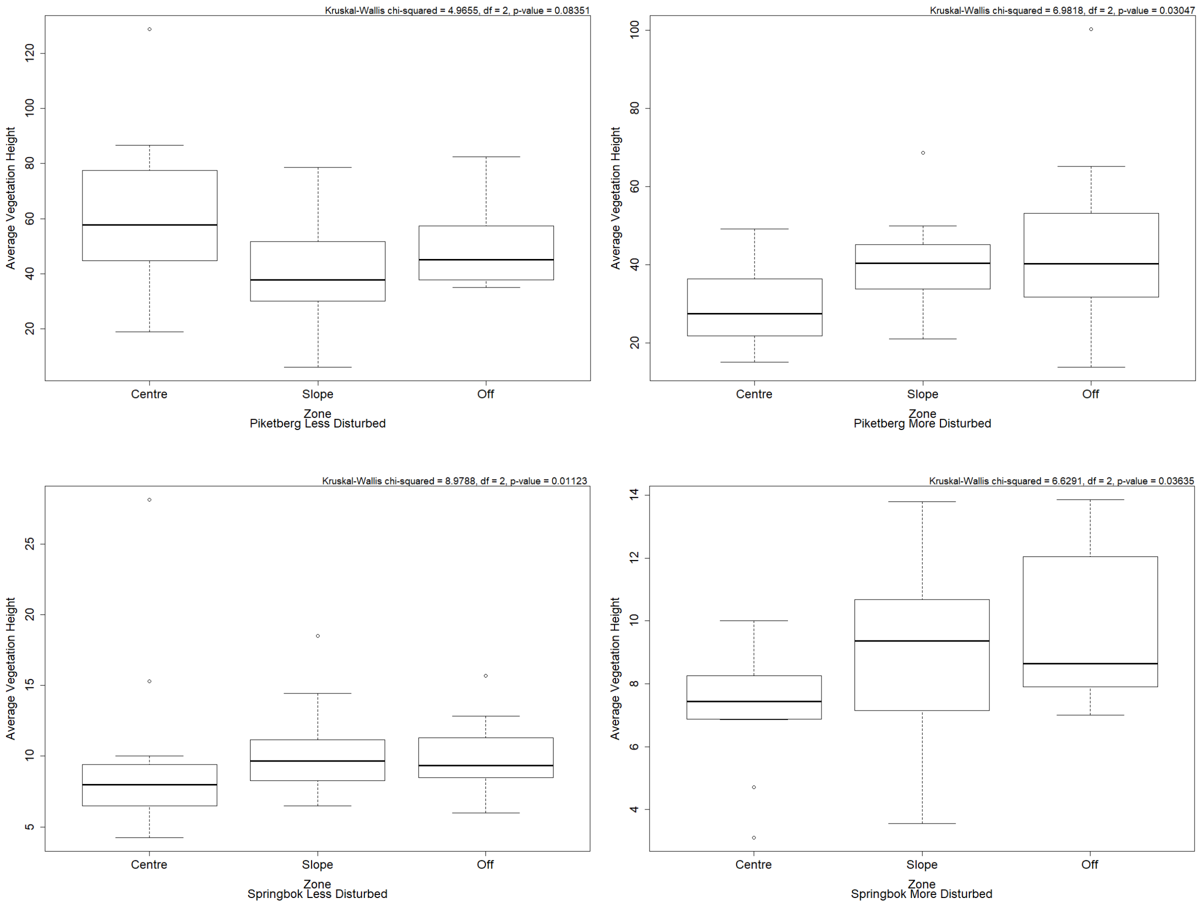
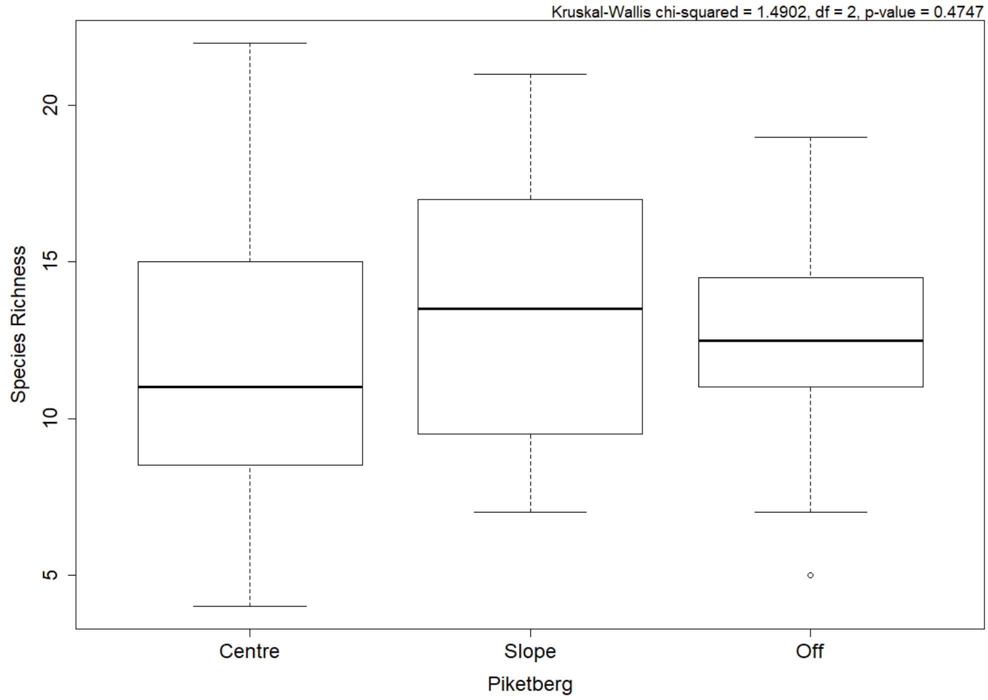


Figure T: Boxplots of vegetation height by zone in each at each subsite with significant differences

```
Pairwise comparisons using Wilcoxon rank sum test

data:  Piketbergdata$Average.height and Piketbergdata$Zone
      Centre Off
Off   0.93   -
Slope 0.93   0.93

data:  Springbokdata$Average.height and Springbokdata$Zone
      Centre Off
Off   0.0015 -
Slope 0.0034 0.9693
.......................................................................

data:  PiketbergMD$Total.Vegetation.cover and PiketbergMD$Zone
      Centre Off
Off   0.87   -
Slope 0.87   0.77
P value adjustment method: holm

data:  SpringbokMD$Average.height and SpringbokMD$Zone
      Centre Off
Off   0.033  -
Slope 0.188  0.590

data:  SpringbokLD$Average.height and SpringbokLD$Zone
      Centre Off
Off   0.027  -
Slope 0.024  0.744
```

Species Richness



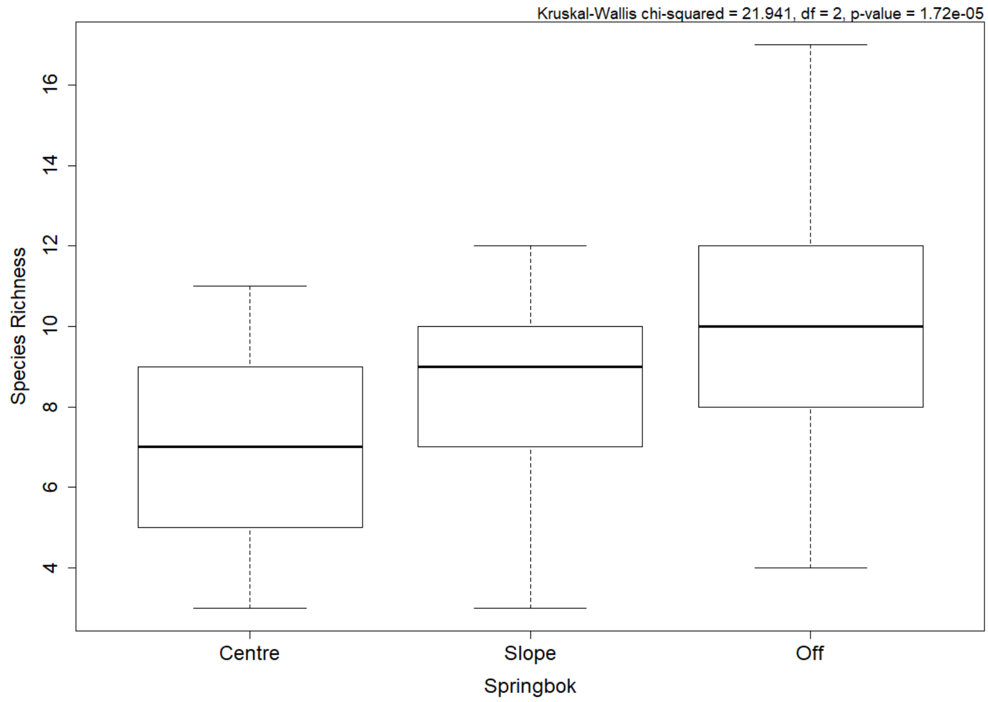


Figure U: Boxplots of species richness by zone in each biome.

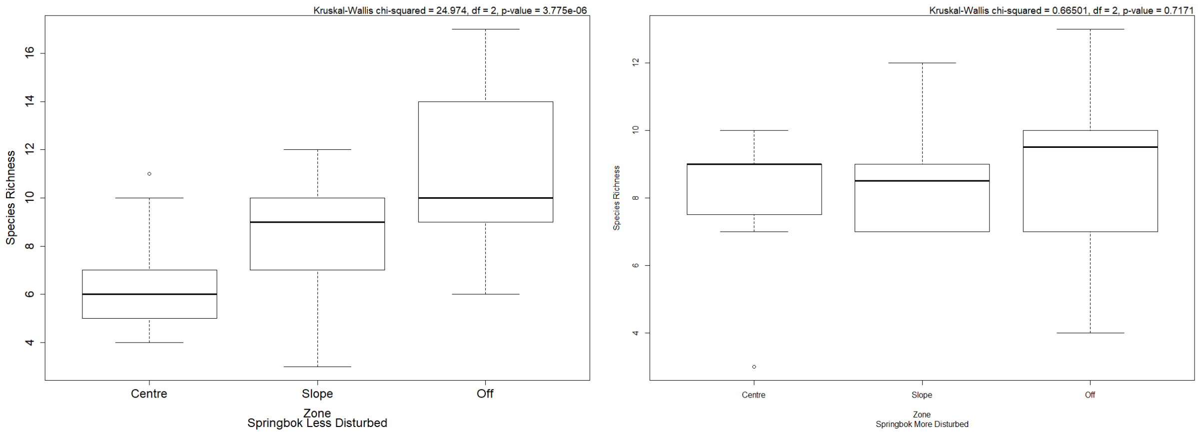
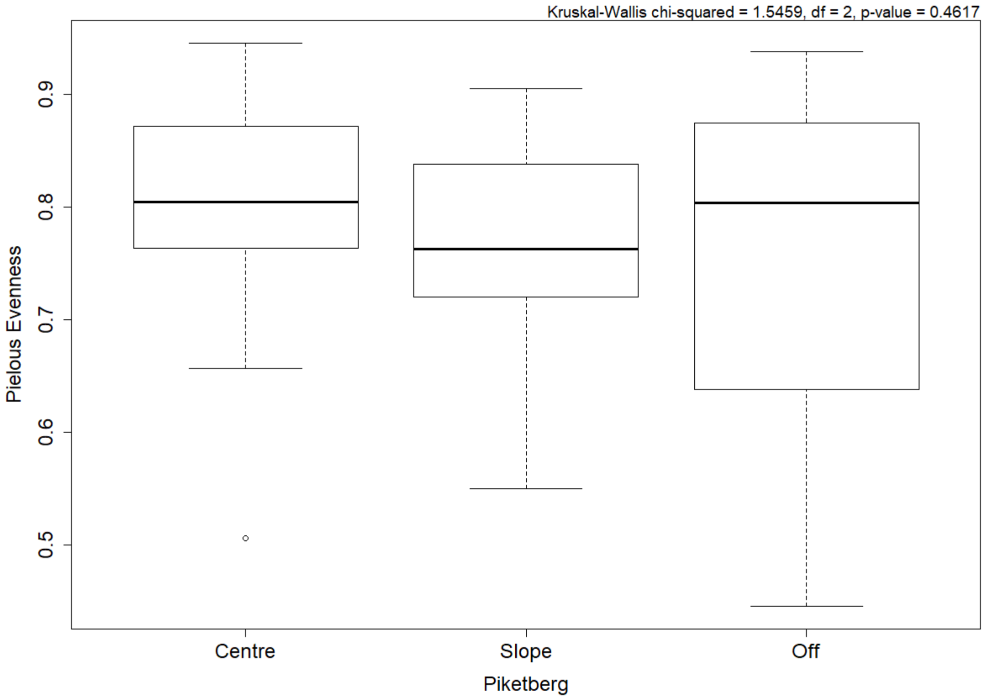


Figure V: Boxplots of species richness by zone in each biome.

```
> kruskal.test(Species.richness~Zone,data=vegstats,subset=Site=="Piketberg Les
s Disturbed")#Kruskal-Wallis chi-squared = 3.635, df = 2, p-value = 0.1624

        Kruskal-Wallis rank sum test

data:  Species.richness by Zone
Kruskal-Wallis chi-squared = 3.635, df = 2, p-value = 0.1624

> kruskal.test(Species.richness~Zone,data=vegstats,subset=Site=="Piketberg Mor
e Disturbed")#Kruskal-Wallis chi-squared = 1.656, df = 2, p-value = 0.4369

        Kruskal-Wallis rank sum test

data:  Species.richness by Zone
Kruskal-Wallis chi-squared = 1.656, df = 2, p-value = 0.4369


Pairwise comparisons using Wilcoxon rank sum test

data:  Springbokdata$Species.richness and Springbokdata$Zone

      Centre  Off
Off   3.1e-05 -
Slope 0.016   0.012

data:  SpringbokMD$Species.richness and SpringbokMD$Zone

      Centre Off
Off   1      -
Slope 1      1


data:  SpringbokLD$Species.richness and SpringbokLD$Zone

      Centre  Off
Off   1.1e-05 -
Slope 0.0081  0.0081
```

Pielou's Evenness



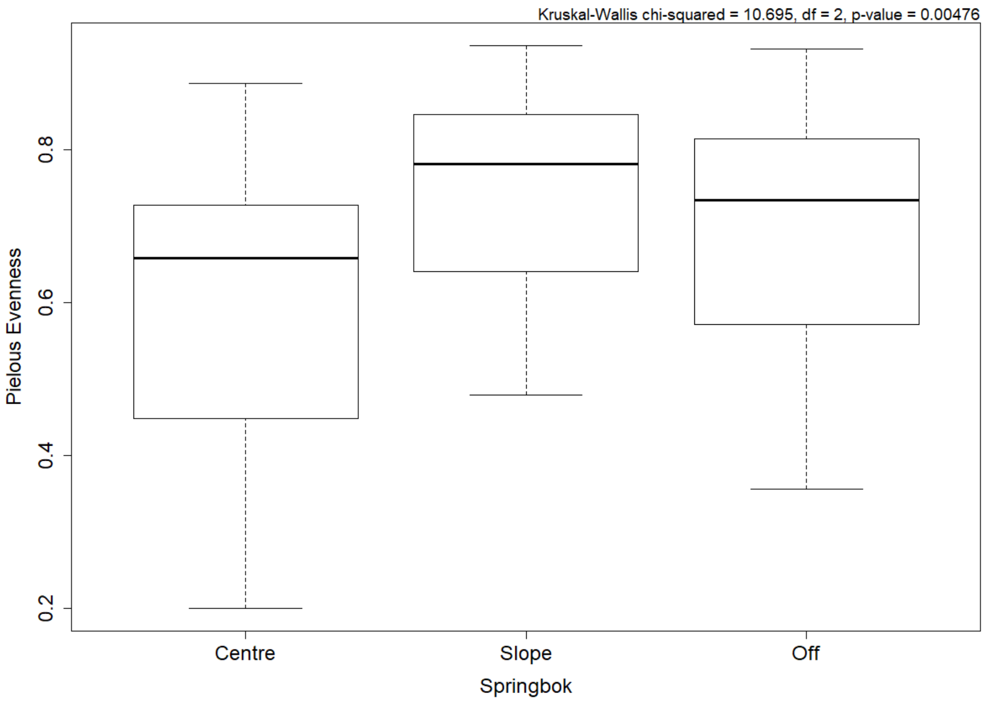


Figure W: Boxplots of evenness by zone in each biome.

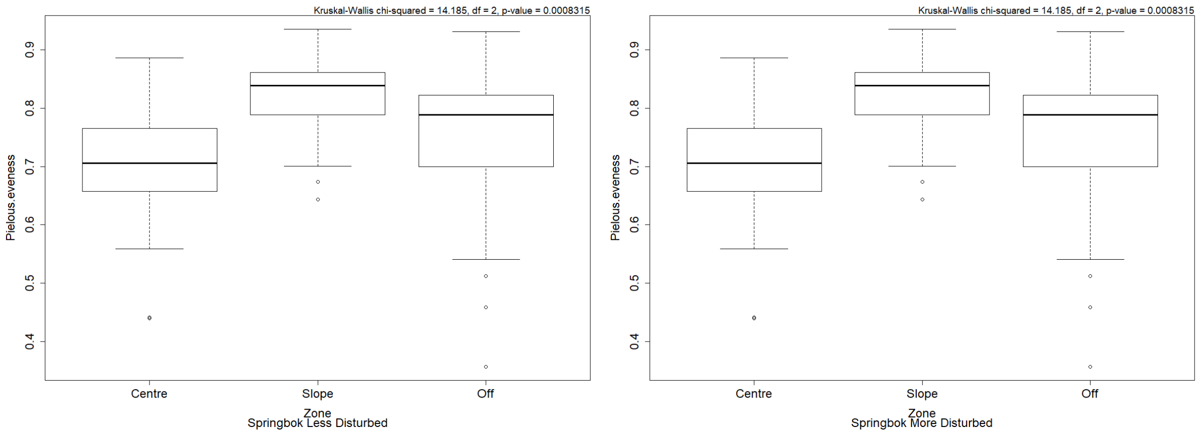


Figure X: Boxplots of evenness by zone in each subsite with significant results

```
> kruskal.test(Pielous.eveness~Zone,data=vegstats,subset=Site=="Piketberg More
Disturbed")

        Kruskal-Wallis rank sum test

data:  Pielous.eveness by Zone
Kruskal-Wallis chi-squared = 3.2988, df = 2, p-value = 0.1922


> kruskal.test(Pielous.eveness~Zone,data=vegstats,subset=Site=="Piketberg Less
Disturbed")

        Kruskal-Wallis rank sum test

data:  Pielous.eveness by Zone
Kruskal-Wallis chi-squared = 2.2027, df = 2, p-value = 0.3324



Pairwise comparisons using Wilcoxon rank sum test

data:  Springbokdata$Pielous.eveness and Springbokdata$Zone

      Centre Off
Off   0.0450 -
Slope 0.0046 0.3132


data:  SpringbokMD$Pielous.eveness and SpringbokMD$Zone

      Centre Off
Off   0.020  -
Slope 0.048  0.291


data:  SpringbokLD$Pielous.eveness and SpringbokLD$Zone

      Centre  Off
Off   0.09802 -
Slope 0.00022 0.09802
```

Appendix 4: Heuweltjie Surface Characteristics across Biomes and Zones

Table A: Showing Average values for data collected on the heuweltjie Surface Characteristics at each of the Sites across the Heuweltjie zones per quadrats (4m^2). p-value: p<0,05; p<0,01**; p<0,001***; different letters show groups that are significantly different from each other (p < 0.05)) in posthoc tests amongst zones.*

Biome **Zone**	**Fynbos** **Centre**	**Fynbos** **Slope**	**Fynbos** **Off**	*Zone (p-value)*	**S. Karoo** **Centre**	**S. Karoo** **Slope**	**S. Karoo** **Off**	*Zone (p-value)*	*Site (t-tests)*
Holes per qudrat (no.)	**6.21±6.53**	**11.83±9.92**	**8.75±8.28**	.	**4.52±4.85**	**5.61±5.28**	**6.27±6.79**		**
XS hole	4.08±5.06	8.88±8.30	5.08±6.93		2.15±2.72	4.12±4.02	3.94±4.96		
S hole	1.67±1.88	1.96±2.61	2.63±3.65		1.12±1.75	1.33±2.56	2.30±4.04		
M hole	0.46±0.88	1.00±1.67	1.04±2.37		1.18±2.42	0.12±0.55	0.03±0.17		
L hole	0.00	0.00	0.00		0.06±0.35	0.03±0.17	0.00		
Surface disturbance (no)	**0.50±1.10**	**0.96±1.68**	**0.79±1.10**		**0.45±0.75**	**0.24±0.44**	**0.06±0.24**	*	**
M pit	0.04±0.20	0.63±1.53	0.21±0.59			0.00	0.00		
L dug pit	0.04±0.20	0.21±0.41	0.04±0.20		0.03±0.17	0.00	0.00		
Mole hill	0.21±1.02	0.04±0.20	0.04±0.20		0.00	0.00	0.00		
Raised mounds	0.21±0.51	0.08±0.28	0.50±0.93		0.42±0.75	0.24±0.44	0.06±0.24		
Litter (Cover)	**18.42±21.96**	**5.17±10.77**	**5.21±10.99**	**	**0.09±0.52**	**2.12±6.00**	**0.67±2.85**		***
Stick piles	**0.04±0.20**	**0.17±0.48**	**0.46±1.14**		**0.00**	**0.09±0.29**	**0.09±0.29**		
Frass (cover)	**11.79±19.41**	**1.25±4.23**	**0.46±2.04**	***	**1.03±3.34**	**0.12±0.70**	**0.45±1.79**		*
Cryptogram cover	**3.96±6.40**	**3.63±7.97**	**3.54±7.82**		**1.18±4.63**	**3.15±7.85**	**1.61±8.70**		***
Lichen (Cover)	0.42±2.04	0.00	0.00		0.67±2.23	0.33±1.22	0.39±1.80		
Crust (cover)	0.63±3.06	0.42±2.04	1.79±7.16		0.00	0.00	0.00		
Crust (moss) (cover)	2.92±5.47	3.21±7.88	1.75±3.90		0.52±2.62	2.82±7.21	1.21±6.96		
Earthworm casts (presence/absence)	**0.17±0.38**	**0.33±0.48**	**0.29±0.46**		**0.00**	**0.00**	**0.00**		***

Appendix 5: Top 10 most common species by cover

TableB: Species list showing the top 10 species by cover at each of the sites and Zones within each Site See appendix for remaining list

Site	Succulent Karoo More Disturbed Site											
Zone	**SITE**			**Centre**			**Slope**			**Off**		
	Species	**Average % Cover**	**Area (m2)**	**Species**	**Average % Cover**	**Area (m2)**	**Species**	**Average % Cover**	**Area (m2)**	**Species**	**Average % Cover**	**Area (m2)**
1	**Sp10**	11.778	0.471	**Sp99**	7,056	0,282	**Sp10**	4,389	0,176	**Sp10**	4,722	0,189
2	**Sp99**	10,056	0,402	**Sp10**	2,667	0,107	**Sp25**	3,250	0,130	**Sp25**	4,222	0,169
3	**Sp25**	9,583	0,383	**Sp25**	2,111	0,084	**Sp1**	3,139	0,126	**Sp1**	3,722	0,149
4	**Sp1**	8,472	0,339	**Sp1**	1,611	0,064	**Sp42**	2,694	0,108	**Sp33**	2,167	0,087
5	**Sp42**	4,667	0,187	**Sp101**	1,583	0,063	**Sp99**	2,667	0,107	**Sp23**	1,306	0,052
6	**Sp33**	3,361	0,134	**Sp42**	1,194	0,048	**Sp104**	1,778	0,071	**Sp12**	1,250	0,050
7	**Sp101**	2,861	0,114	**Sp33**	0,333	0,013	**Sp33**	0,861	0,034	**Sp101**	0,889	0,036
8	**Sp104**	2,000	0,080	**Sp12**	0,278	0,011	**Sp94**	0,806	0,032	**Sp42**	0,778	0,031
9	**Sp12**	1,972	0,079	**Sp94**	0,222	0,009	**Sp103**	0,500	0,020	**Sp94**	0,722	0,029
10	**Sp94**	1,750	0,070	**Sp23**	0,222	0,009	**Sp100**	0,500	0,020	**Sp84**	0,389	0,016
Site	Succulent Karoo less disturbed site											
Zone	**Site**			**Centre**			**Slope**			**Off**		
	Species	**Average % Cover**	**Area (m2)**	**Species**	**Average % Cover**	**Area (m2)**	**Species**	**Average % Cover**	**Area (m2)**	**Species**	**Average % Cover**	**Area (m2)**
1	**Sp16**	5,444	0,218	**Sp5**	3,048	0,122	**Sp40**	2,159	0,086	**Sp16**	5,349	0,214
2	**Sp5**	4,619	0,185	**Sp10**	2,397	0,096	**Sp8**	1,825	0,073	**Sp15**	3,460	0,138
3	**Sp10**	4,349	0,174	**Sp1**	2,048	0,082	**Sp1**	1,778	0,071	**Sp44**	3,317	0,133
4	**Sp1**	4,206	0,168	**Sp56**	1,048	0,042	**Sp10**	1,603	0,064	**Sp45**	2,730	0,109
5	**Sp15**	4,206	0,168	**Sp68**	0,905	0,036	**Sp5**	1,556	0,062	**Sp48**	1,381	0,055
6	**Sp44**	3,492	0,140	**Sp8**	0,540	0,022	**Sp83**	1,111	0,044	**Sp41**	0,730	0,029
7	**Sp45**	2,746	0,110	**Sp42**	0,317	0,013	**Sp15**	0,746	0,030	**Sp74**	0,730	0,029
8	**Sp8**	2,714	0,109	**Sp84=2**	0,317	0,013	**Sp42**	0,683	0,027	**Sp90**	0,730	0,029
9	**Sp40**	2,429	0,097	**Sp81=7**	0,317	0,013	**Sp41**	0,651	0,026	**Sp54**	0,714	0,029

10	**Sp56**	1,810	0,072	**Sp40**	0,270	0,011	**Sp56**	0,444	0,018	**Sp70**	0,714	0,029
Site	Fynbos More Disturbed Site											
Zone	**Site**			**Centre**			**Slope**			**Off**		
	Species	**Average % Cover**	**Area (m2)**	**Species**	**Average % Cover**	**Area (m2)**	**Species**	**Average % Cover**	**Area (m2)**	**Species**	**Average % Cover**	**Area (m2)**
1	**FE**	30,667	1,227	**FE**	12,639	0,506	**FE**	10,278	0,411	**FE**	7,750	0,310
2	**FH**	8,750	0,350	**Q =AA**	3,472	0,139	**FH**	5,639	0,226	**FI**	3,528	0,141
3	**FB**	7,597	0,304	**FB**	2,667	0,107	**FB**	3,361	0,134	**I**	2,556	0,102
4	**FI**	4,778	0,191	**FK**	2,361	0,094	**B**	1,556	0,062	**AD**	2,361	0,094
5	**AD**	4,306	0,172	**FH**	2,083	0,083	**FI**	1,250	0,050	**FB**	1,569	0,063
6	**Q =AA**	3,500	0,140	**FA**	1,903	0,076	**BX**	1,250	0,050	**D**	1,472	0,059
7	**FA**	3,181	0,127	**BK**	1,528	0,061	**AD**	1,139	0,046	**FJ**	1,389	0,056
8	**E**	2,833	0,113	**E**	1,139	0,046	**AL**	1,111	0,044	**BE**	1,111	0,044
9	**B**	2,681	0,107	**AD**	0,806	0,032	**BM**	1,111	0,044	**FH**	1,028	0,041
10	**I**	2,556	0,102	**FF**	0,764	0,031	**FA**	1,111	0,044	**B**	0,972	0,039
Site	Fynbos Less Disturbed Site											
Zone	**Site**			**Centre**			**Slope**			**Off**		
	Species	**Average % Cover**	**Area (m2)**	**Species**	**Average % Cover**	**Area (m2)**	**Species**	**Average % Cover**	**Area (m2)**	**Species**	**Average % Cover**	**Area (m2)**
1	**CH**	17,917	0,717	**CH**	17,917	0,717	**BS**	5,417	0,217	**CS**	6,944	0,278
2	**CS**	9,667	0,387	**FE**	5,611	0,224	**AR**	4,194	0,168	**CP=CT**	4,389	0,176
3	**AR**	8,556	0,342	**AR**	4,083	0,163	**CL = EO**	4,167	0,167	**DO**	3,806	0,152
4	**CP=CT**	7,333	0,293	**CX**	3,194	0,128	**CQ**	3,833	0,153	**CL = EO**	2,222	0,089
5	**CL = EO**	6,611	0,264	**DJ=DP**	2,361	0,094	**CP=CT**	2,944	0,118	**DJ=DP**	1,944	0,078
6	**FE**	6,000	0,240	**FB**	2,028	0,081	**CS**	2,722	0,109	**DX**	1,611	0,064
7	**BS**	5,694	0,228	**FA**	1,500	0,060	**FM**	2,500	0,100	**CF**	1,528	0,061
8	**CQ**	5,667	0,227	**FM**	1,472	0,059	**DM**	2,417	0,097	**FI**	1,528	0,061
9	**DJ=DP**	5,556	0,222	**DE**	0,972	0,039	**FB**	2,306	0,092	**CQ**	1,361	0,054
10	**FB**	4,694	0,188	**BX**	0,972	0,039	**DE**	2,222	0,089	**CG**	1,306	0,052

Appendix 6: Photos of sampling locations on Heuweltjies

Fynbos More Disturbed Site

Figure Y *PH1 (Piketberg (Fynbos) Heuweltjie 1)*

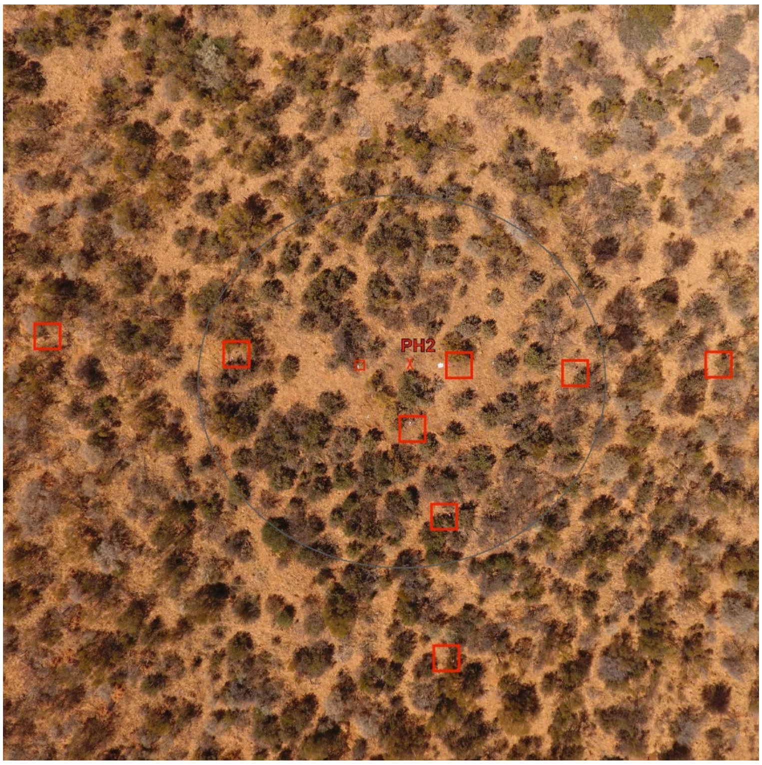


Figure Z *PH2 (Piketberg (Fynbos) Heuweltjie 2)*

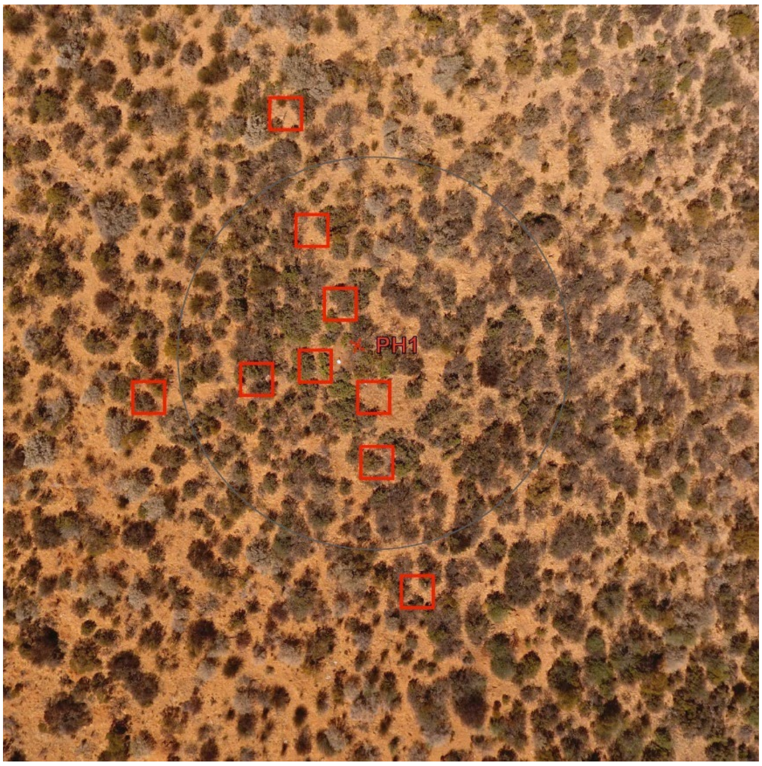


*Figure AA*PH3 (Piketberg (Fynbos) Heuweltjie 3)

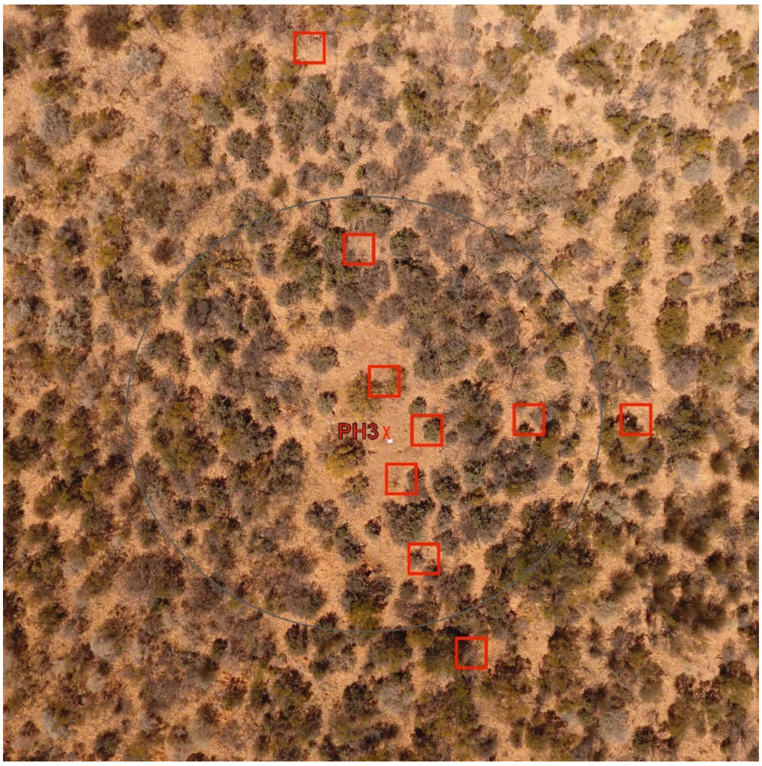
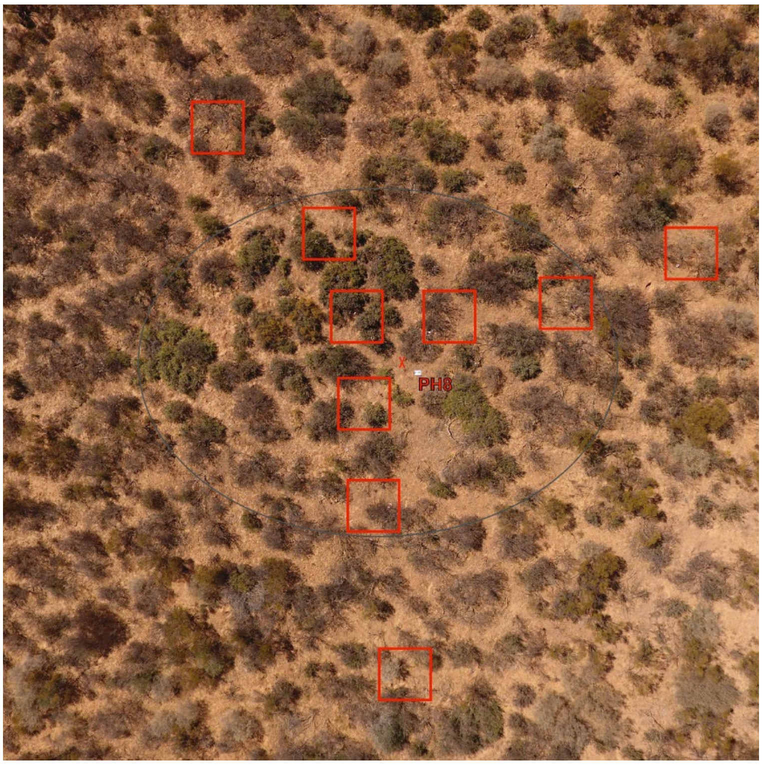


*Figure BB*PH8 (Piketberg (Fynbos) Heuweltjie 8)



Fynbos Least Disturbed Site

Figure CC PH4 (Piketberg (Fynbos) Heuweltjie 4)

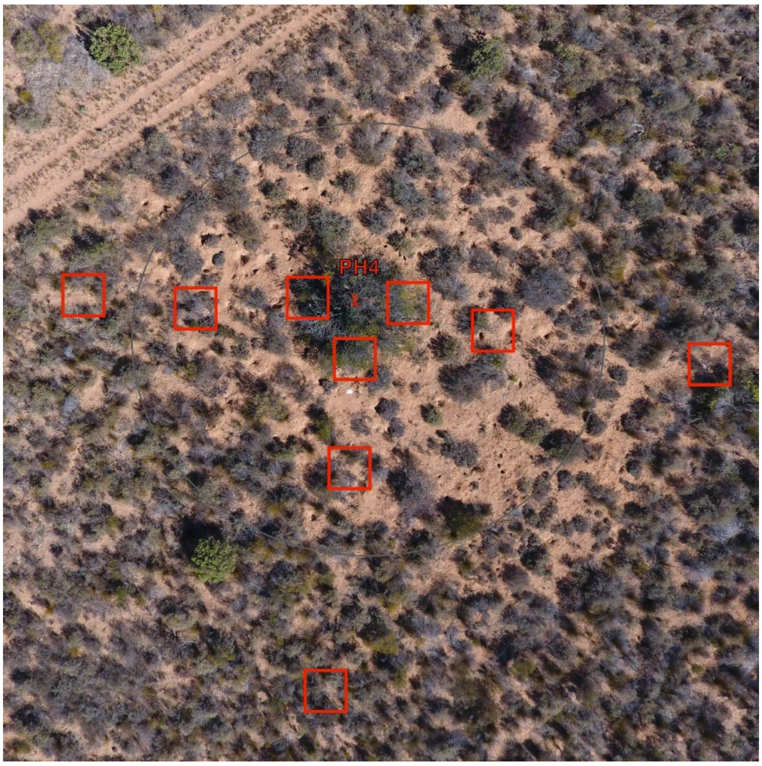


Figure DD PH5 (Piketberg (Fynbos) Heuweltjie 5)

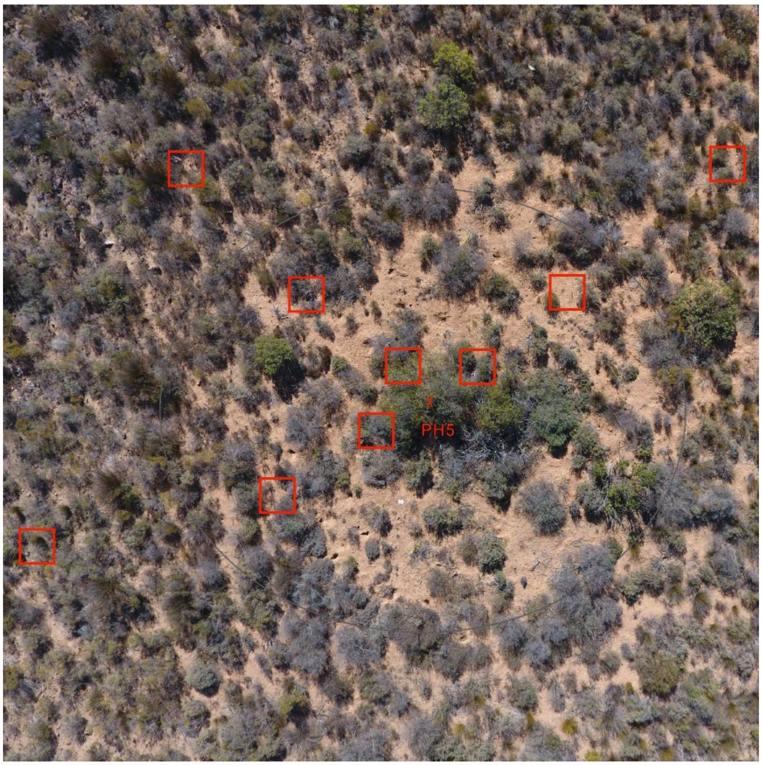


*Figure EE*PH6 (Piketberg (Fynbos) Heuweltjie 6)

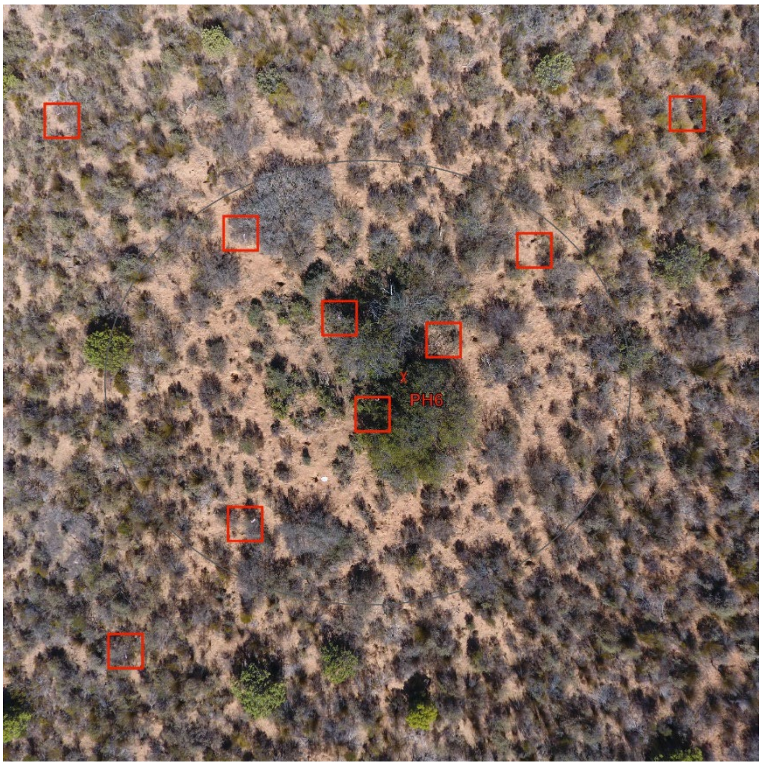
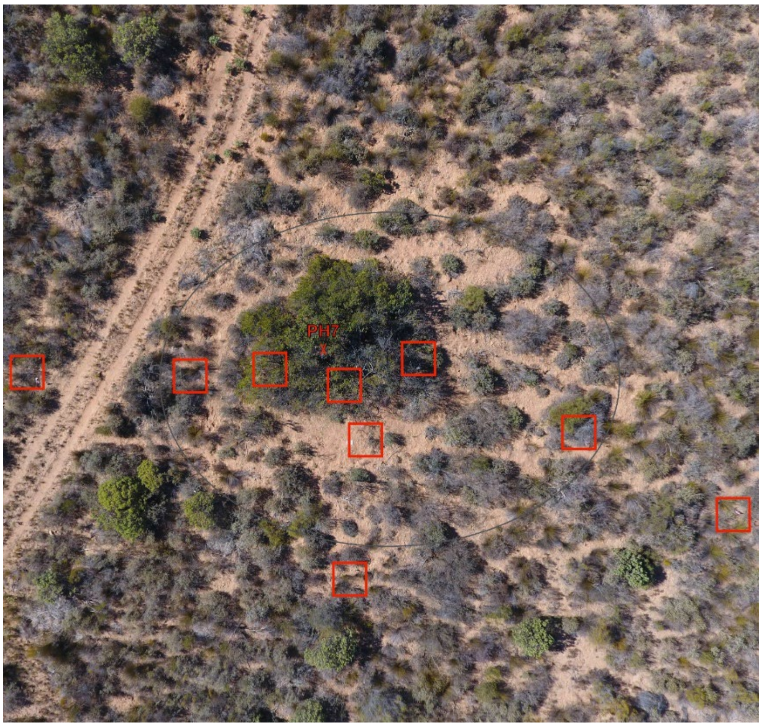


Figure FF PH7 (Piketberg (Fynbos) Heuweltjie 7)



Succulent Karoo Least Disturbed Heuweltjies

Figure GG *KH1 (Kommagas (Succulent Karoo) Heuweltjie 1)*

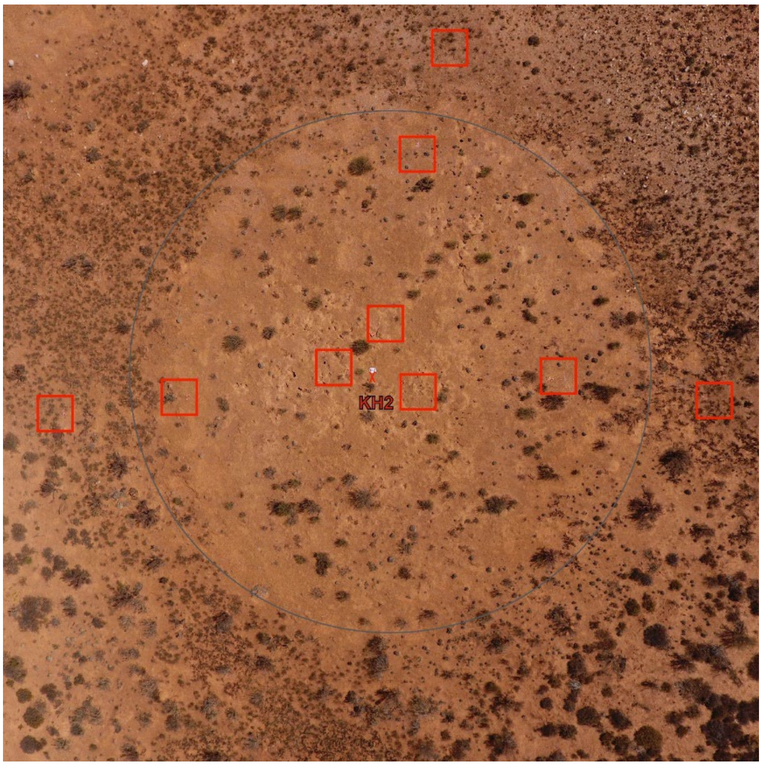


Figure HH *KH2 (Kommagas (Succulent Karoo) Heuweltjie 2)*

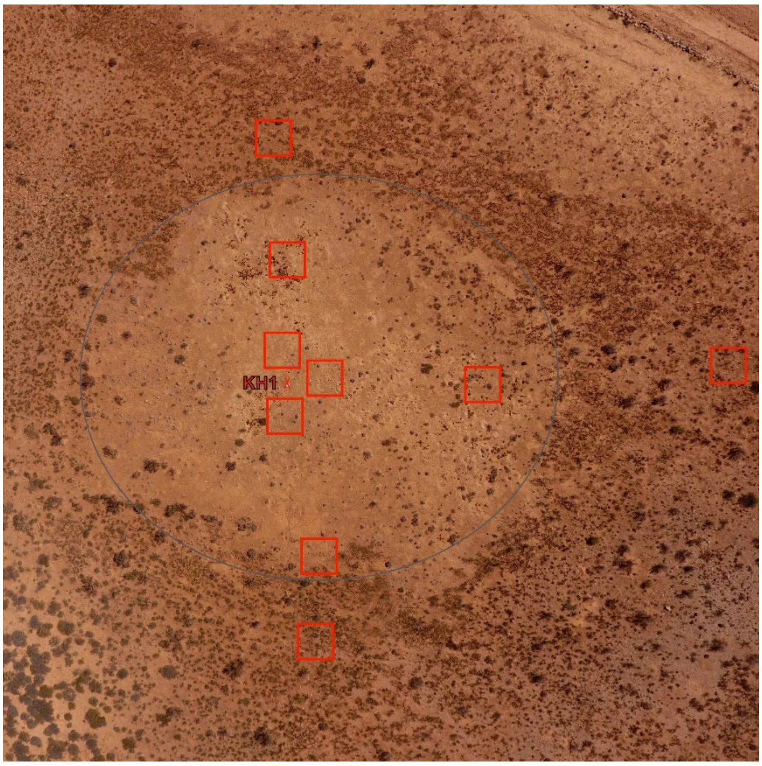


Figure II<u>*KH3 (Kommagas (Succulent Karoo) Heuweltjie 3)*</u>

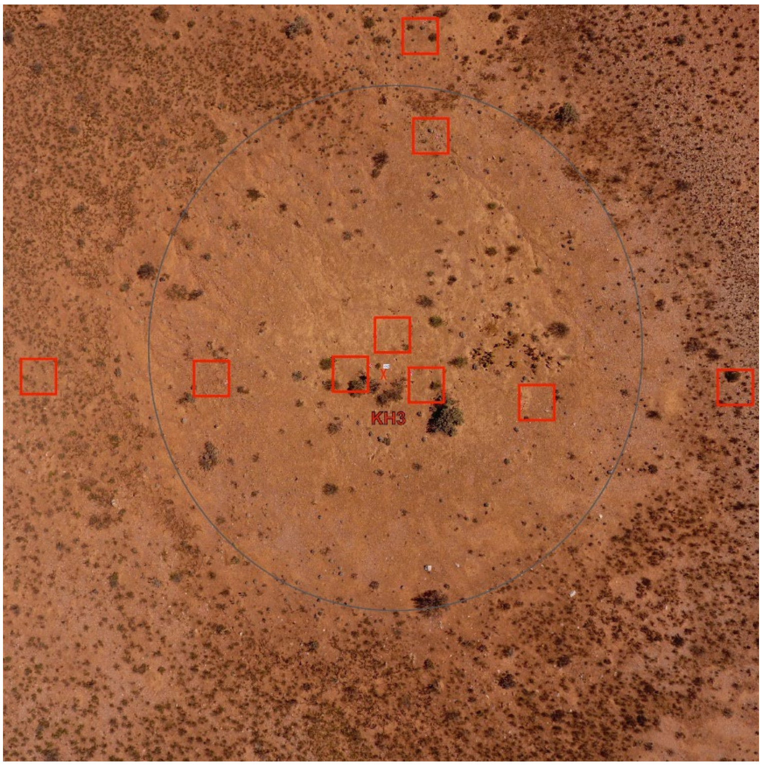


Figure JJ KH4 (Kommagas (Succulent Karoo) Heuweltjie 4)

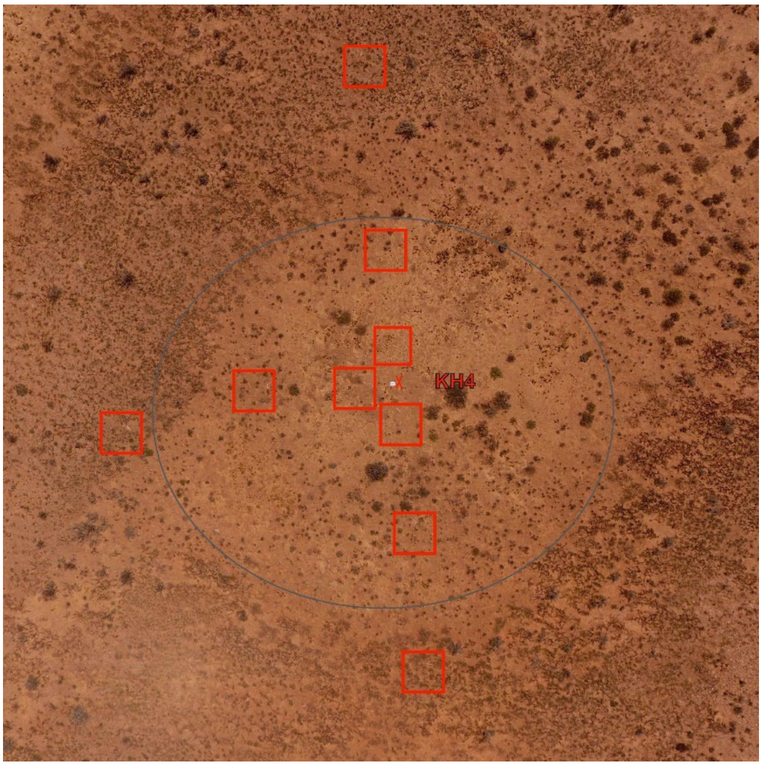


Figure KKKH5 (Kommagas (Succulent Karoo) Heuweltjie 5)

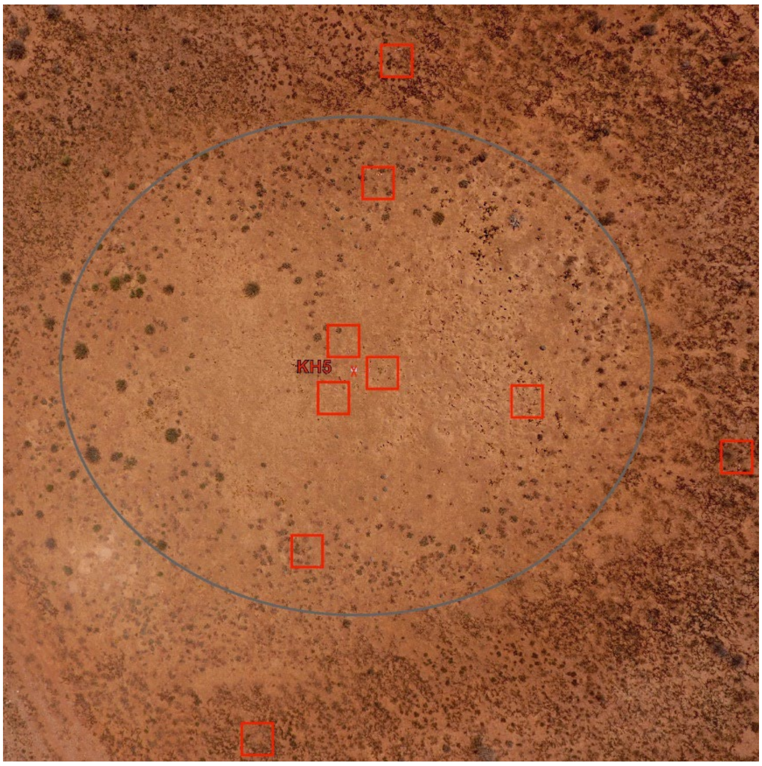


Figure LL KH6 (Kommagas (Succulent Karoo) Heuweltjie 6)

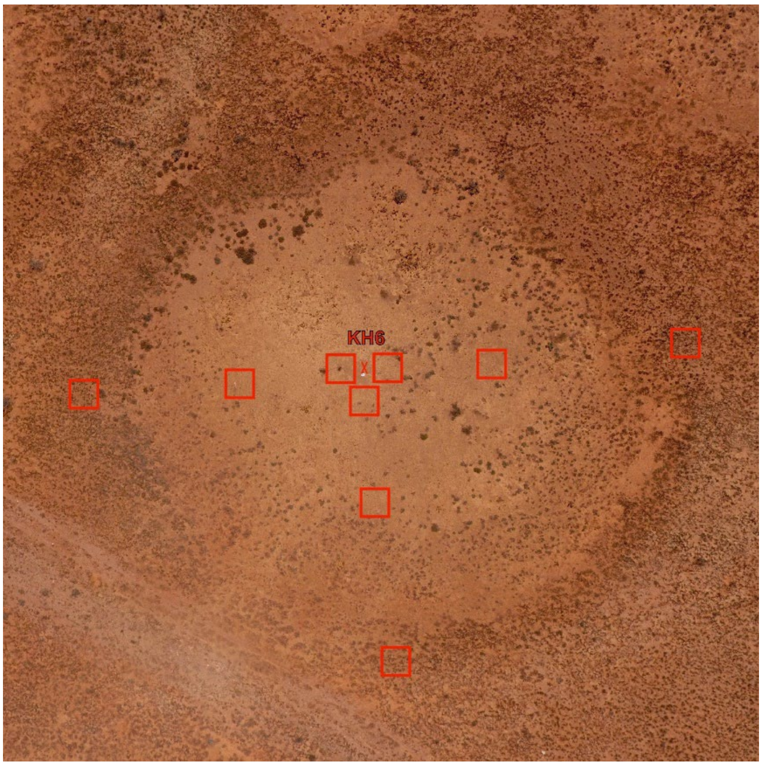
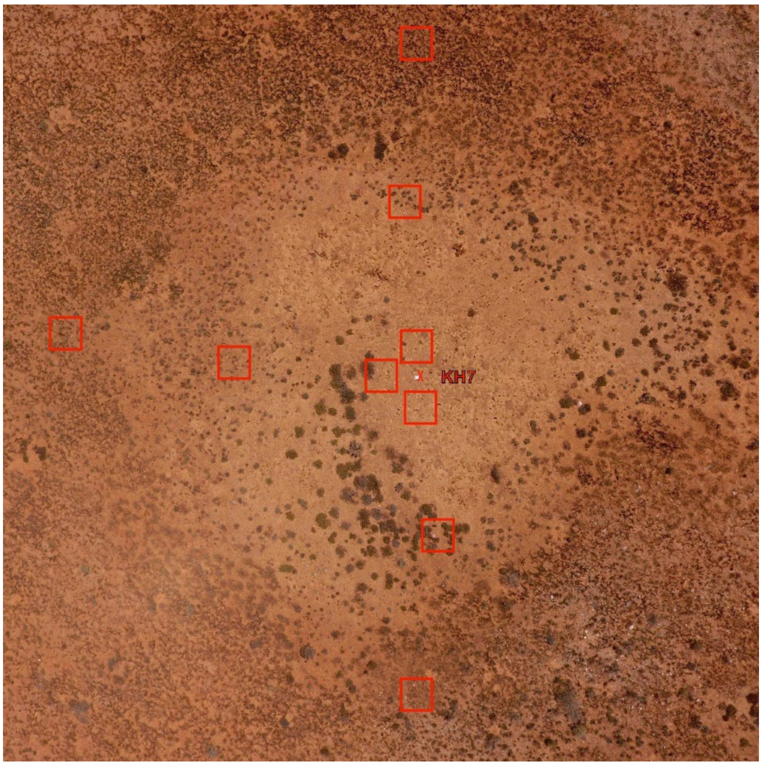


Figure MM KH7 (Kommagas (Succulent Karoo) Heuweltjie 7)



Succulent Karoo More Disturbed

Figure NNBH1 (Buffelsrivier (Succulent Karoo) Heuweltjie 1)

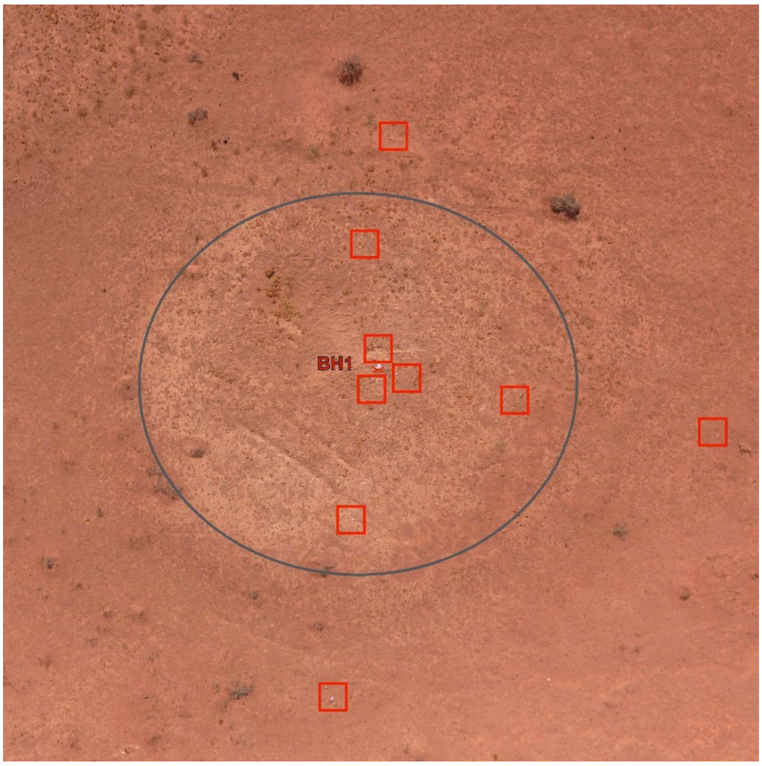


Figure OO BH2 (Buffelsrivier (Succulent Karoo) Heuweltjie 2)

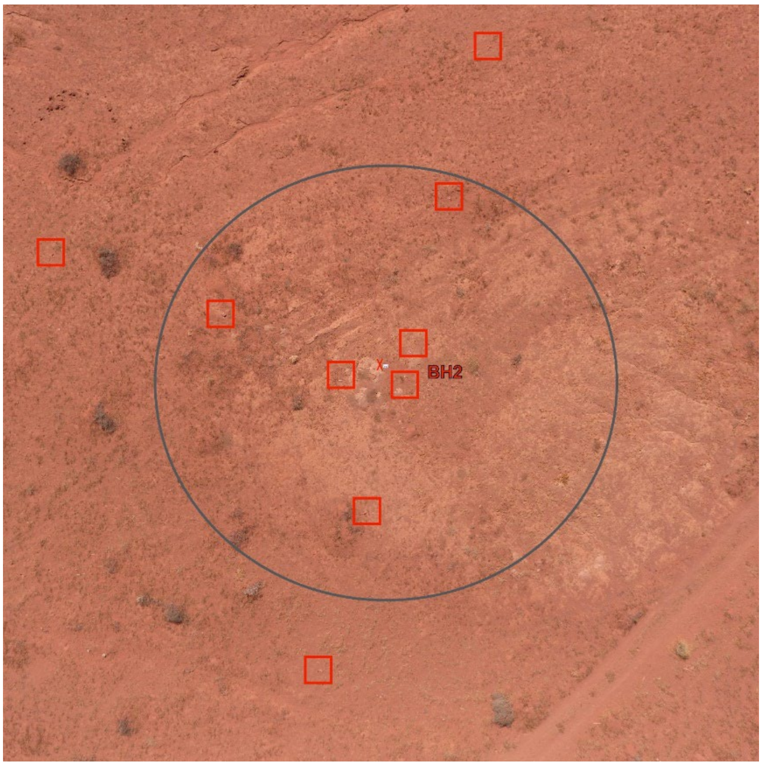


Figure PP *BH3 (Buffelsrivier (Succulent Karoo) Heuweltjie 3)*

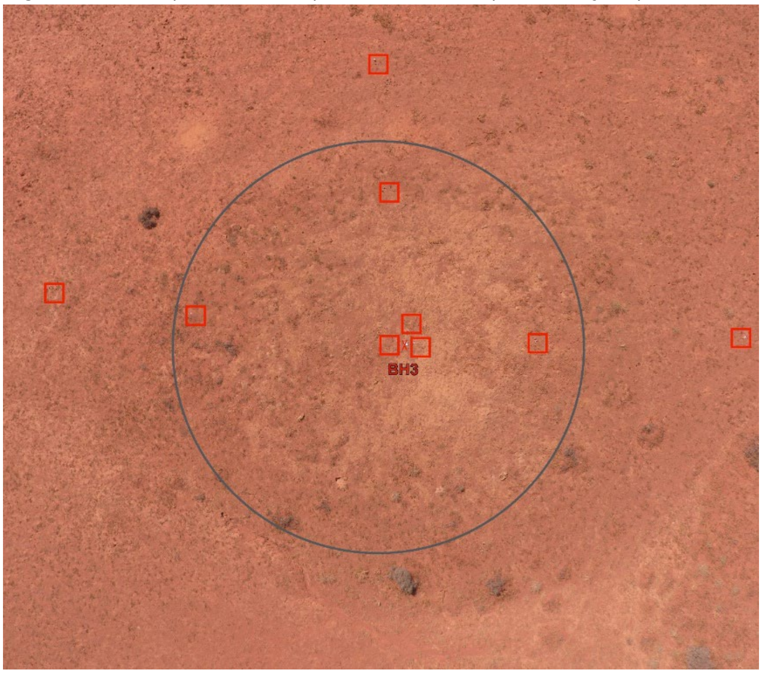
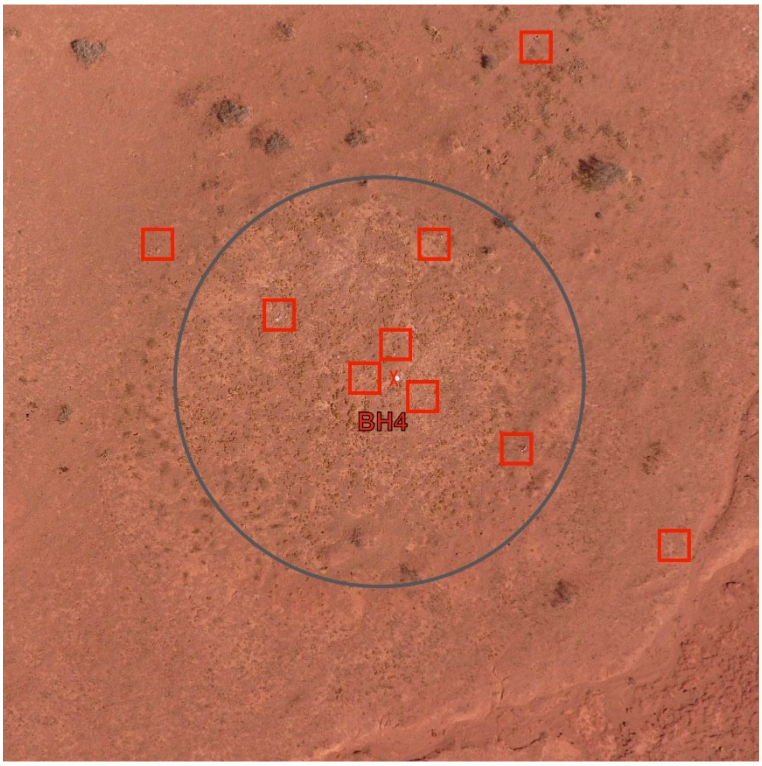


Figure QQ *BH4 (Buffelsrivier (Succulent Karoo) Heuweltjie 4)*



Appendix 7: Plant species list

Table C: Plant Species list

Succulent Karoo

Species Code	Family	*Genus*	*Species*	Growth form
1	Asteraceae	*Faveolina*	*dichotoma*	**Herb**
4=57	Oxalidaceae	*Oxalis*	*annae*	**Herb**
5	Aizoaceae	*Mesembryanthemum*	*barklyi*	**Succulent Herb**
8	Aizoaceae	*Cheirodopsis*	*denticulata*	**Succulent Shrub**
10	Aizoaceae	*Tetragonia*	*microptera*	**Succulent Herb**
12	Brassicaceae	*Heliophilia*	*carnosa*	**Herb**
13=51	Asteraceae	*Leysera*		**Herb**
15	Aizoaceae	*Ruschia*	*leucosperma*	**Succulent Shrub**
16	Aizoaceae	*Jordaaniella*	*cuprea*	**Succulent Shrub**
17	Amaryllidaceae	*Gethyllis*	*ciliaris*	**Geophytic Herb**
19	Asparagaceae	*Ornithogalum*	*xanthochlorum*	**Geophytic Herb**
21=42	Aizoaceae	*Galenia*	*sarophylla*	**Herb**
23	Aizoaceae	*Drosanthemum*	*eburneum*	**Succulent Shrub**
25	Fabaceae	*Lotononis*	*falcata*	**Herb**
29	Asteraceae	*Gazania*	*rigida*	**Herb**
32	Poaceae	*Bromus*		**Graminoid**
33	Poaceae	*Schismus*	*schismoides*	**Graminoid**
34	Asteraceae	*Onocosiphon*	*suffruticosum*	**Herb**
36	Zygophyllaceae	*Roepera*	*cordifolia*	**Succulent Shrub**
38	Oxalidaceae	*Oxalis*	*obtusa*	**Herb**
39	Geraniaceae	*Monsonia*	*spinosa*	**Succulent Shrub**
40	Aizoaceae	*Ruschia*	*centrocapsula?*	**Succulent Shrub**
41	Asteraceae	*Didelta*	*carnosa*	**Succulent Herb**
43	Asparagaceae	*Albuca*		**Succulent Herb**
44	Asteraceae	*Gazania*	*difusa*	**Herb**
45	Aizoaceae	*Ruschia*		**Succulent Shrub**
46	Aizoaceae	*Mesembryanthemum*	*deciduum*	**Succulent Shrub**
47	Asparagaceae	*Ornithogalum*	*pruinosum*	**Geophytic Herb**
48	Aizoaceae			**Succulent Shrub**
49	Aizoaceae	*Tetragonia*	*sarcophylla*	**Succulent Herb**
50	Aizoaceae	*Ruschia*	*goodiae*	**Succulent Shrub**
52				**Herb**
53	Aizoaceae	*Cheirodopsis*		**Succulent Shrub**
54	*Euphorbiaceae*	*Euphorbia*	*hamata*	**Succulent Shrub**
55	*Euphorbiaceae*	*Euphorbia*	*decussata*	**Succulent Shrub**
56	*Euphorbiaceae*	*Euphorbia*	*damarana?*	**Succulent Shrub**
58	Aizoaceae			**Succulent Shrub**
59				**Herb**
60				**Herb**
61	Asteraceae			**Succulent Herb**
62	Aizoaceae			**Succulent Shrub**
63	Aizoaceae	*Conophytum*		**Succulent Shrub**
64	Crassulaceae	*Crassula*	*columnaris*	**Succulent Shrub**
65	Crassulaceae	*Crassula*	*barklyi*	**Succulent Shrub**

66	Asteraceae	*Felicia*	*merxmuellri?*	**Herb**
67				**Succulent Shrub**
68	Amaranthaceae	*Salsola*	*stocksli*	**Low Shrub**
69	Apocynaceae	*Hoodia?*		**Succulent shrub**
70	Aizoaceae			**Succulent shrub**
71	Aizoaceae			**Low Shrub**
72				**Succulent Herb**
73				**Low Shrub**
74	Aizoaceae			**Succulent Shrub**
75				**Low Shrub**
76	Apocynaceae	*Ceropegia*	*pulvinata*	**Succulent Herb**
77				**Succulent Shrub**
78	Aizoaceae	*or Crassulaceae*		**Succulent Shrub**
79	Aizoaceae	*Ruschia*	*cradockensis subsp triticiformis*	**Succulent Shrub**
80	Aizoaceae	*Mesembryanthemum*	*junceum*	**Succulent Herb**
81=7	Aizoaceae	*Mesembryanthemum*	*quartziticola*	**Succulent Shrub**
82	Aizoaceae	*Mesembryanthemum*	*noctiflorum*	**Succulent Shrub**
83	Aizoaceae			**Succulent Shrub**
84	Asteraceae	*Ursina?*		**Herb**
84=2	Asteraceae	*Osteospermum*	*pinnatum*	**Herb**
85	Iridaceae	*Lapeirousia*	*silenoides*	**Geophytic Herb**
86	Amaryllidaceae	*Gethyllis*	*cilliaris*	**Geophytic Herb**
87				**Succulent Shrub**
88				**Low Shrub**
89	Aizoaceae	*Antimima*	*compacta*	**Succulent Shrub**
90	Crassulaceae	*Crassula*	*brevifolia*	**Succulent Shrub**
92	Aizoaceae			**Succulent Shrub**
93	Crassulaceae	*Crassula*	*muscosa*	**Succulent Shrub**
94=91	Asparagaceae	*Albuca?*		**Geophytic Herb**
95	Asteraceae			**Herb**
96	Aizoaceae			**Succulent Shrub**
97	Aizoaceae	*Stoeberia*	*frutescens*	**Succulent Shrub**
98				**Herb**
99	Aizoaceae	*Mesembryanthemum*	*hypertrophicum*	**Succulent Herb**
100				**Low Shrub**
101	Aizoaceae	*Mesembryanthemum*	*guerichianum*	**Succulent Herb**
102	Geraniaceae	*Pelargoium*		**Herb**
103	Zygophyllaceae	*Roepera*		**Succulent Shrub**
104	Caryophyllaceae	*Spergularia*	*media*	**Succulent Herb**
105				**Herb**

Fynbos

Species Code	Family	Genus	Species	Growth form
AA=Q	Asteraceae	*Arctotheca*	*calendula*	**Herb**
AB	Oxalidaceae	*Oxalis*		**Herb**
AC				**Herb**
AD	Poaceae	*Tribolium*	*hispidum?*	**Graminoid**
AE				**Herb**
AF	Iridaceae	*Moraea*	*tripetala*	**Geophytic Herb**
AG				**Succulent Herb**
AH	Convolvulaceae	*Convolvulus*	*capensis*	**Climber**
AI	Iridaceae	*Babiana*	*mucronata*	**Herb**
AJ	Oxalidaceae	*Oxalis*		**Herb**
AK				**Herb**
AL	Poaceae	*Tribolium*		**Graminoid**
AM				**Graminoid**
AN	Poaceae			**Graminoid**
AO	Polygalaceae	*Muraltia*	*spinosa*	**Low Shrub**
AP	Hyacinthaceae	*Lanchenalia*	*unifolia*	**Geophytic Herb**
AQ	Poaceae	*Hordeum*	*capense*	**Graminoid**
AR	Euphorbiaceae	*Euphorbia*	*burmanni*	**Succulent Shrub**
AS				**Herb**
AT				**Climber**
AU				**Herb**
AV	Scrophulariaceae	*Manulea?*		**Herb**
AW	Aizoaceae			**Succulent shrub**
AX				**Herb**
AY	Apocynaceae	*Microloma*	*sagittatum*	**Climber**
AZ				**Herb**
B	Poaceae	*Cladoraphis*	*spinosa*	**Graminoid**
BA				**Herb**
BB				**Geophytic Herb**
BC				**Herb**
BD	Poaceae	*Bromus*	*diandrus*	**Graminoid**
BE	Fabaceae	*Wiborgia?*		**Small tree**
BJ	Asteraceae	*Oncosiphon*	*grandiflorum*	**Herb**
BK	Asteraceae	*Gazania*	*krebsiana*	**Herb**
BL	Asteraceae	*Osteospermum*	*clandestinum*	**Herb**
BM	Geraniaceae	*Erodium*	*cicutarium*	**Herb**
BN				**Low Shrub**
BO	Iridaceae	*Moraea*		**Geophytic Herb**
BP				**Herb**
BQ				**Low Shrub**
BR				**Herb**
BS	Fabaceae	*Wiborgia*		**Low Shrub**
BT				**Herb**
BU	Scrophulariaceae	*Manulea?*		**Herb**
BV				**Herb**
BW				**Geophytic Herb**
BX	Poaceae	*Ehrharta*	*calycina*	**Graminoid**
BY				**Geophytic Herb**
BZ	Hyacinthaceae	*Lanchenalia*	*mutabilis*	**Geophytic herb**
C=K=DG	Asteraceae	*Helichrysum*	*moeserianum*	**Herb**
CA				**Herb**
CC				**Herb**
CD				**Geophytic Herb**
CE				**Low shrub**
CF	Asteraceae	*Eriocephalus*	*africanus*	**Low shrub**
CG				**Low Shrub**
CH	Anacardiaceae	*Searsia*	*undulata*	**Small tree**
CI				**Herb**
CJ				**Herb**
CK =DT	Asparagaceae	*Asparagus*	*rubicundus*	**Low shrub**

CL =EO	Aizoaceae	*Ruschia*	*lunulata*	**Succulent Shrub**
CM=EX				**Small tree**
CN				**Geophytic Herb**
CO	Oxalidaceae	*Oxalis*	*flava*	**Herb**
CP (m)= CT (f)	Restionaceae	*Thamnochortus*	*insignis*	**Graminoid-Restio**
CQ				**Low Shrub**
CR	Anacardiaceae	*Searsia*	*dissecta*	**Low Shrub**
CS	Asteraceae	*Metalasia*	*muricata*	**Low Shrub**
CU	Asteraceae	*Senecio*	*littoreus*	**Herb**
CW				**Herb**
CX				**Climber**
CY	Scrophulariaceae	*Lyperia*	*tristis*	**Herb**
CZ				**Herb**
D	Campanulaceae	*Prismatocarpus?*		**Herb**
DA	Oxalidaceae	*Oxalis*	*levis?*	**Graminoid**
DB				**Low Shrub**
DC				**Succulent Herb**
DD	Campanulaceae	*Wahlenbergia?*		**Herb**
DE	Poaceae	*Ehrharta*	*calycina*	**Graminoid**
DF				**Low Shrub**
DH				**Herb**
DI	Rubiaceae	*Anthospermum*	*spathulatum*	**Low Shrub**
DJ=DP	Asteraceae	*Pteronia*	*incana*	**Low Shrub**
DK				**Low Shrub**
DL				**Low shrub**
DM				**Herb**
DN	Geraniaceae	*Pelargonium*	*triste*	**Herb**
DO	Rutaceae	*Macrostylis*	*hirta*	**Low shrub**
DQ				**Low Shrub**
DR =EJ				**Herb**
DS				**Low Shrub**
DU				**Graminoid**
DV	Asparagaceae	*Lanchenalia*		**Geophytic Herb**
DW	Euphorbiaceae	*Euphorbia*	*hamata*	**Succulent Shrub**
DX	Restionaceae	*Willdenowia*	*incurvata*	**Graminoid-Restio**
DY				**Low Shrub**
E	Asteraceae	*Cotula*	*pruinosa*	**Herb**
EA				**Geophytic herb**
EB				**Herb**
EC				**Herb**
ED				**Herb**
EE				**Herb**
EF				**Low Shrub**
EG				**Geophytic Herb**
EH				**Graminoid**
EI				**Geophytic Herb**
EK	Asteraceae	*Osteospermum*	*moniliferum*	**Low shrub**
EL				**Herb**
EM				**Low Shrub**
EN	Fabaceae			**Low Shrub**
EP =DZ	Restionaceae			**Graminoid-Restio**
EQ				**Herb**
ER	Geraniaceae	*Pelagonium*		**Herb**
ES	Restionaceae	*Willdenowia ?*		**Graminoid-Restio**
ET				**X**
EU				**Low Shrub**
EV				**Low Shrub**
EW				**Low Shrub**
EY	Restionaceae	*Platycaulos*	*major*	**Graminoid-Restio**

EZ				**Herb**
FA	Oxalidaceae	*Oxalis*	*purpurea*	**Herb**
FB	Agavaceae	*Chlorophytum*	*triflorum*	**Geophytic Herb**
FC	Scrophulariaceae	*Diascia*	*longicornis*	**Herb**
FD	Oxalidaceae	*Oxalis*		**Herb**
FE	Aizoaceae	*Ruschia*	*suaveolens*	**Succulent Shrub**
FF	Oxalidaceae	*Oxalis*	*pes capre*	**Herb**
FG	Aizoaceae	*Cleretum*		**Succulent Herb**
FH	Fabaceae	*Wiborgia*	*sericea*	**Low Shrub**
FI	Thymelaeaceae	*Passerina*	*truncta ssp. Truncata*	**Low Shrub**
FJ	Restionaceae	*Elegia*	*mucronata*	**Graminoid-Restio**
FK	Aizoaceae	*Aizoon*	*africanum*	**Low shrub**
FL	Agavaceae	*Chlorophytum*	*undulatum*	**Geophytic Herb**
FM	Asteraceae	*Euryops*	*speciosissimus*	**Low shrub**
FN	Campanulaceae	*Wahlanergia*	*cupensis*	**Herb**
G	Polygalaceae	*Muraltia*		**Herb**
H				**Herb**
I	Fabaceae	*Aspalathus*	*acuminata ssp. acuminata*	**Low shrub**
J	Iridaceae	*Lapeirousis*	*fabicii*	**Geophytic Herb**
L=CB				**Herb**
M	Tecophilaeaceae	*Cyanella*	*hyacinthoides*	**Geophytic Herb**
N				**Geophytic Herb**
O				**Herb**
P				**Herb**
R	Hyacinthaceae	*Albuca*	*cooperi*	**Geophytic Herb**
S	Aizoaceae	*Cleretum*		**Succulent Herb**
T				**Geophytic Herb**
U				**Herb**
V				**Herb**
W	Poaceae			**Graminoid**
X	Oxalidaceae	*Oxalis*		**Herb**
Y				**Graminoid**
Z	Asparagaceae	*Lanchenalia*		**Geophytic Herb**

Appendix 8: Collected plant species list and photos

Table D: Collected plant species

Site	Zone collected	Species code	Species name	Functional type
Fynbos -LD	Centre	CH	Searsia undulata	Small tree
		FE	Ruschia suaveolens	Succulent Shrub
	Off	CS	Metalasia muricata	Low Shrub
		CT	Thamnochortus insignis	Graminoid-Restio
	Slope	Cl	Ruschia lunulata	Succulent Shrub
		FB	Chlorophytum triflorum	Geophytic Herb
Fynbos -MD	Centre	FB	Chlorophytum triflorum	Geophytic Herb
		FE	Ruschia suaveolens	Succulent Shrub
	Off	FE	Ruschia suaveolens	Succulent Shrub
		FI	Passerina truncta ssp. Truncata	Low Shrub
	Slope	FE	Ruschia suaveolens	Succulent Shrub
		FH	Wiborgia sericea	Low Shrub
Succulent Karoo -LD	Centre	Sp10	Tetragonia microptera	Succulent Herb
		Sp5	Mesembryanthemum barklyi	Succulent Herb*
	Off	Sp15	Ruschia leucosperma	Succulent Shrub
		Sp16	Jordaaniella cuprea	Succulent Shrub
	Slope	Sp40	Ruschia centrocapsula?	Succulent Shrub
		Sp8	Cheirodopsis denticulata	Succulent Shrub
Succulent Karoo - MD	Centre	Sp10	Tetragonia microptera	Succulent Herb
		Sp99	Mesembryanthemum hypertrophicum	Succulent Herb*
	Off	Sp10	Tetragonia microptera	Succulent Herb
		Sp25	Lotononis falcata	Herb
	Slope	Sp10	Tetragonia microptera	Succulent Herb
		Sp25	Lotononis falcata	Herb

Photos: Succulent Karoo plants

Figure RR: Sp40- Ruschia centrocapsula?

Figure SS: Sp5- Mesembryanthemum barklyi (soutslaai)

Figure TT: Sp8 - Cheirodopsis denticulata

Figure UU: Sp10- Tetragonia microptera

Figure VV: Sp15- Ruschia leucosperma

Figure WW:Sp16- Jordaaniella cuprea

Figure XX:Sp25- Lotononis falcata

Figure YY: Sp99-Mesembryanthemum hypertrophicum

Fynbos plants

Figure ZZ: FE- Ruschia suaveolens

Figure AAA:FB- Chlorophytum triflorum

Figure BBB: FI- Passerina truncta ssp. Truncata

Figure CCC: FH- Wiborgia sericea

Figure DDD:CH- Searsia undulata

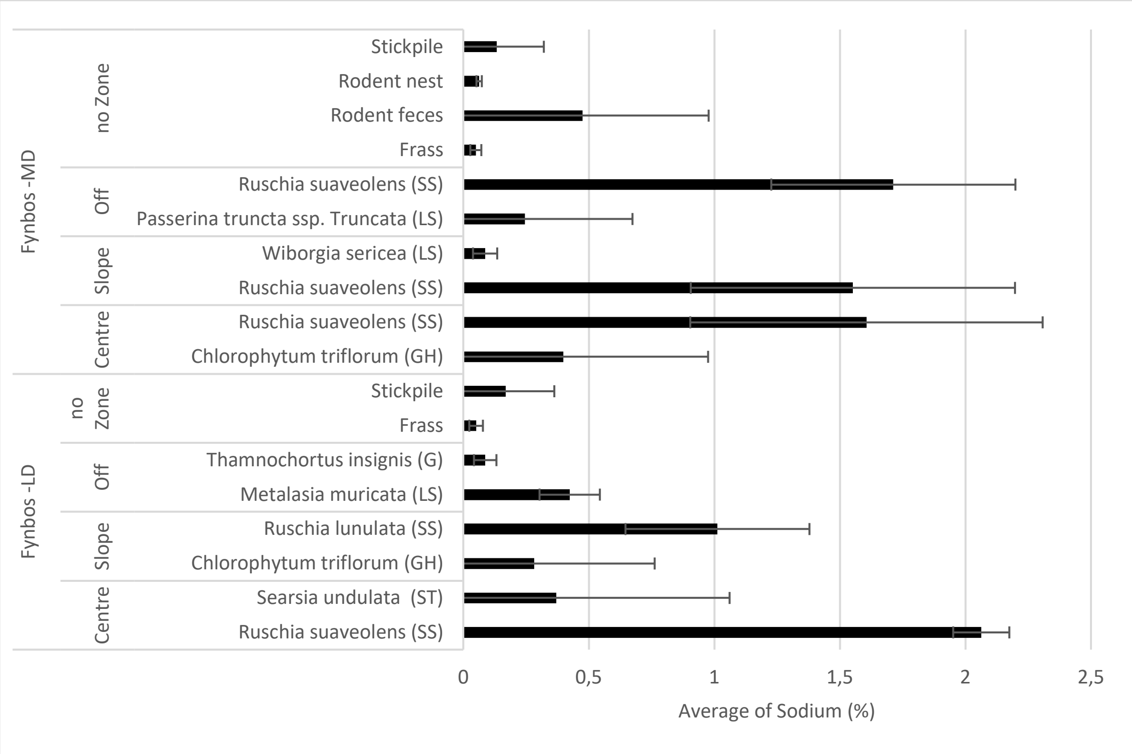
Figure EEE:CS- Metalasia muricata

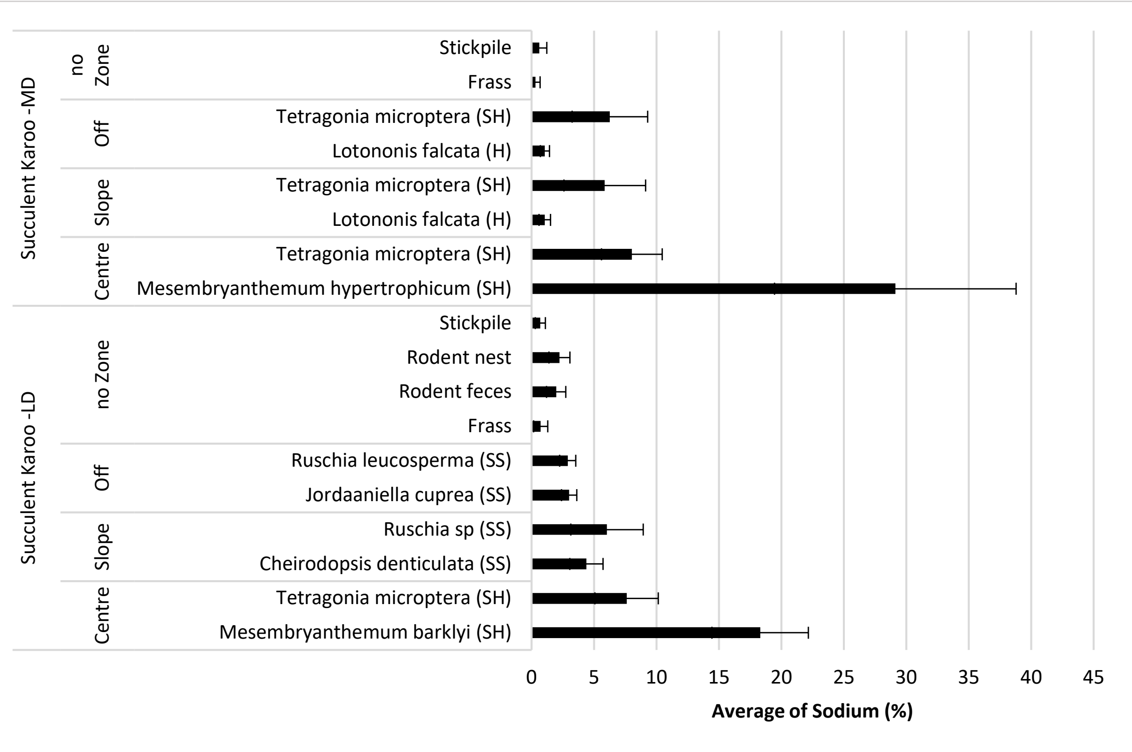
Figure FFF:CL- Ruschia lunulata

Figure GGG:CT= Thamnochortus insignis

Appendix 9: Bar graphs of each ion showing the averages concentrations of each type of plant sample collected and r-code of statistical tests run.

Sodium



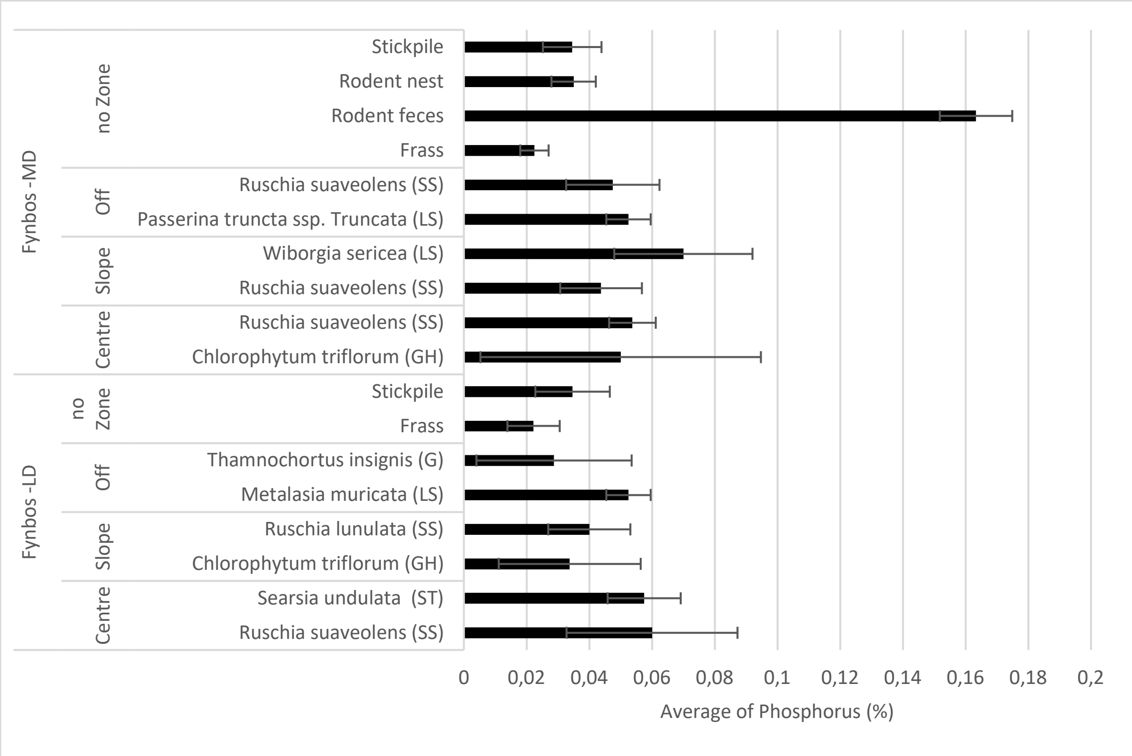


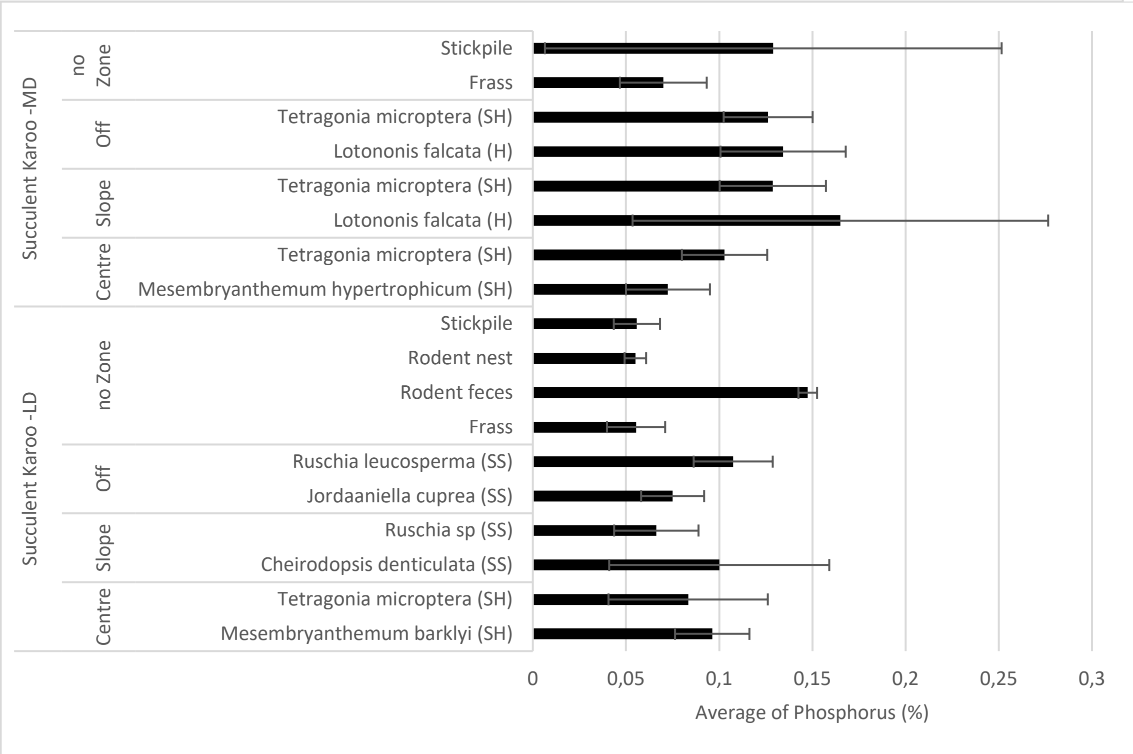

```
> #Na
> #Biome
> wilcox.test(TSA$Na_pc~TSA$Biome)

        Wilcoxon rank sum test with continuity correction

data:  TSA$Na_pc by TSA$Biome
W = 3002, p-value <2e-16
alternative hypothesis: true location shift is not equal to 0

> #Subsite
```

```
> wilcox.test(Fyndata$Na_pc~Fyndata$Site)

         Wilcoxon rank sum test with continuity correction

data:  Fyndata$Na_pc by Fyndata$Site
W = 2773, p-value = 0.89
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$Na_pc~SKdata$Site)

         Wilcoxon rank sum test with continuity correction

data:  SKdata$Na_pc by SKdata$Site
W = 2620, p-value = 0.71
alternative hypothesis: true location shift is not equal to 0

> #Year
> wilcox.test(Fyndata$Na_pc~Fyndata$Year)

         Wilcoxon rank sum test with continuity correction

data:  Fyndata$Na_pc by Fyndata$Year
W = 2444, p-value = 0.26
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$Na_pc~SKdata$Year)

         Wilcoxon rank sum test with continuity correction

data:  SKdata$Na_pc by SKdata$Year
W = 2202, p-value = 0.18
alternative hypothesis: true location shift is not equal to 0

> #Sample type
> kruskal.test(Fyndata$Na_pc~Fyndata$Name)

         Kruskal-Wallis rank sum test

data:  Fyndata$Na_pc by Fyndata$Name
Kruskal-Wallis chi-squared = 105, df = 11, p-value <2e-16

> pairwise.wilcox.test(Fyndata$Na_pc,Fyndata$Name)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndata$Na_pc and Fyndata$Name

             CH    Cl    CS    CT    FB    FE    FH    FI    Frass Rodent feces Rodent nest
Cl           0.415 -     -     -     -     -     -     -     -     -            -
CS           1.000 0.415 -     -     -     -     -     -     -     -            -
CT           0.009 0.205 0.009 -     -     -     -     -     -     -            -
FB           0.908 0.794 0.712 0.238 -     -     -     -     -     -            -
FE           0.009 0.052 0.009 0.002 1e-04 -     -     -     -     -            -
FH           0.009 0.205 0.009 1.000 0.359 0.002 -     -     -     -            -
FI           0.415 0.986 0.415 1.000 0.712 0.004 1.000 -     -     -            -
Frass        3e-05 0.002 3e-05 0.470 3e-06 7e-08 0.976 0.607 -     -            -
Rodent feces 1.000 1.000 1.000 1.000 1.000 0.415 1.000 1.000 0.178 -            -
Rodent nest  1.000 1.000 1.000 1.000 1.000 0.676 1.000 1.000 1.000 1.000        -
Stickpile    0.014 0.011 0.009 1.000 0.607 5e-08 1.000 1.000 0.082 1.000        1.000

P value adjustment method: holm
There were 13 warnings (use warnings() to see them)
> kruskal.test(SKdata$Na_pc~SKdata$Name)

         Kruskal-Wallis rank sum test

data:  SKdata$Na_pc by SKdata$Name
Kruskal-Wallis chi-squared = 122, df = 11, p-value <2e-16

> pairwise.wilcox.test(SKdata$Na_pc,SKdata$Name)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdata$Na_pc and SKdata$Name

             Frass Rodent feces Rodent nest Sp10  Sp15  Sp16  Sp25  Sp40  Sp5   Sp8   Sp99
Rodent feces 0.109 -            -           -     -     -     -     -     -     -     -
Rodent nest  0.022 1.000        -           -     -     -     -     -     -     -     -
Sp10         6e-11 0.028        0.028       -     -     -     -     -     -     -     -
Sp15         3e-05 0.873        0.921       0.002 -     -     -     -     -     -     -
Sp16         3e-05 0.727        0.921       0.003 1.000 -     -     -     -     -     -
Sp25         0.111 0.314        0.101       4e-08 2e-04 2e-04 -     -     -     -     -
Sp40         6e-04 0.533        0.727       1.000 0.314 0.366 0.025 -     -     -     -
Sp5          3e-05 0.109        0.109       3e-06 0.007 0.007 2e-04 0.007 -     -     -
Sp8          3e-05 0.155        0.275       0.216 0.314 0.366 2e-04 0.913 0.007 -     -
Sp99         3e-05 0.109        0.109       3e-06 0.007 0.007 2e-04 0.007 0.109 0.007 -
Stickpile    1.000 0.070        0.028       6e-12 2e-05 2e-05 0.155 3e-04 2e-05 2e-05 2e-05

P value adjustment method: holm
```

```
> #Na
> kruskal.test(Na_pc~Targeted,data=SKdataplants)

         Kruskal-Wallis rank sum test

data:  Na_pc by Targeted
Kruskal-Wallis chi-squared = 40.72, df = 2, p-value = 1.438e-09

> pairwise.wilcox.test(SKdataplants$Na_pc,SKdataplants$Targeted)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$Na_pc and SKdataplants$Targeted

             Maybe   Not Targeted
Not Targeted 6.7e-09 -
Targeted     0.14    3.0e-09

P value adjustment method: holm
>
> kruskal.test(Na_pc~Targeted,data=Fyndataplants)

         Kruskal-Wallis rank sum test

data:  Na_pc by Targeted
Kruskal-Wallis chi-squared = 50.018, df = 2, p-value = 1.376e-11

> pairwise.wilcox.test(Fyndataplants$Na_pc,Fyndataplants$Targeted)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$Na_pc and Fyndataplants$Targeted

             Maybe   Not Targeted
Not Targeted 3.1e-09 -
Targeted     0.18    4.1e-09

P value adjustment method: holm
> #Na
> kruskal.test(Na_pc~Zone,data=SKdataplants)

         Kruskal-Wallis rank sum test

data:  Na_pc by Zone
Kruskal-Wallis chi-squared = 47.113, df = 2, p-value = 5.882e-11

> pairwise.wilcox.test(SKdataplants$Na_pc,SKdataplants$Zone)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$Na_pc and SKdataplants$Zone

      Centre  Off
Off   2.3e-12 -
Slope 9.1e-10 0.23

P value adjustment method: holm
>
> kruskal.test(Na_pc~Zone,data=Fyndataplants)

         Kruskal-Wallis rank sum test

data:  Na_pc by Zone
Kruskal-Wallis chi-squared = 9.1591, df = 2, p-value = 0.01026

> pairwise.wilcox.test(Fyndataplants$Na_pc,Fyndataplants$Zone)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$Na_pc and Fyndataplants$Zone

      Centre Off
Off   0.02   -
Slope 0.03   0.60

P value adjustment method: holm
```

Phosphorous






```
> #P
> #Biome
> wilcox.test(TSA$P_pc~TSA$Biome)

        Wilcoxon rank sum test with continuity correction

data:  TSA$P_pc by TSA$Biome
W = 2769, p-value <2e-16
alternative hypothesis: true location shift is not equal to 0

> #Subsite
> wilcox.test(Fyndata$P_pc~Fyndata$Site)
```

```
         Wilcoxon rank sum test with continuity correction

data:  Fyndata$P_pc by Fyndata$Site
W = 2318, p-value = 0.1
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$P_pc~SKdata$Site)

         Wilcoxon rank sum test with continuity correction

data:  SKdata$P_pc by SKdata$Site
W = 1380, p-value = 2.9e-06
alternative hypothesis: true location shift is not equal to 0

> #Year
> wilcox.test(Fyndata$P_pc~Fyndata$Year)

         Wilcoxon rank sum test with continuity correction

data:  Fyndata$P_pc by Fyndata$Year
W = 3264, p-value = 0.04
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$P_pc~SKdata$Year)

         Wilcoxon rank sum test with continuity correction

data:  SKdata$P_pc by SKdata$Year
W = 2424, p-value = 0.65
alternative hypothesis: true location shift is not equal to 0

> #Sample type
> kruskal.test(Fyndata$P_pc~Fyndata$Name)

         Kruskal-Wallis rank sum test

data:  Fyndata$P_pc by Fyndata$Name
Kruskal-Wallis chi-squared = 82, df = 11, p-value = 6.1e-13

> pairwise.wilcox.test(Fyndata$P_pc,Fyndata$Name)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndata$P_pc and Fyndata$Name

             CH    Cl    CS    CT    FB    FE    FH    FI    Frass Rodent feces Rodent nest
Cl           0.666 -     -     -     -     -     -     -     -     -            -
CS           1.000 1.000 -     -     -     -     -     -     -     -            -
CT           0.407 0.759 0.407 -     -     -     -     -     -     -            -
FB           0.666 1.000 0.829 1.000 -     -     -     -     -     -            -
FE           1.000 1.000 1.000 0.070 0.351 -     -     -     -     -            -
FH           1.000 0.204 0.829 0.316 0.489 0.308 -     -     -     -            -
FI           1.000 1.000 1.000 0.407 0.829 1.000 0.829 -     -     -            -
Frass        8e-04 0.032 8e-04 1.000 0.586 5e-07 7e-04 8e-04 -     -            -
Rodent feces 0.586 0.606 0.586 0.308 0.407 0.205 0.586 0.586 0.096 -            -
Rodent nest  1.000 1.000 1.000 1.000 1.000 1.000 1.000 1.000 0.829 1.000        -
Stickpile    0.014 1.000 0.027 0.463 1.000 0.002 0.003 0.027 0.003 0.227        1.000

P value adjustment method: holm
> kruskal.test(SKdata$P_pc~SKdata$Name)

         Kruskal-Wallis rank sum test

data:  SKdata$P_pc by SKdata$Name
Kruskal-Wallis chi-squared = 67.5, df = 11, p-value = 3.6e-10

> pairwise.wilcox.test(SKdata$P_pc,SKdata$Name)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdata$P_pc and SKdata$Name

             Frass Rodent feces Rodent nest Sp10  Sp15  Sp16  Sp25  Sp40  Sp5   Sp8   Sp99
Rodent feces 0.095 -            -           -     -     -     -     -     -     -     -
Rodent nest  1.000 0.912        -           -     -     -     -     -     -     -     -
Sp10         1e-04 0.990        0.213       -     -     -     -     -     -     -     -
Sp15         0.008 0.876        0.359       1.000 -     -     -     -     -     -     -
Sp16         1.000 0.359        1.000       0.162 0.273 -     -     -     -     -     -
Sp25         8e-05 1.000        0.165       1.000 1.000 0.030 -     -     -     -     -
Sp40         1.000 0.359        1.000       0.075 0.187 1.000 0.020 -     -     -     -
Sp5          0.024 0.359        0.359       1.000 1.000 1.000 0.538 0.767 -     -     -
Sp8          1.000 1.000        1.000       1.000 1.000 1.000 1.000 1.000 1.000 -     -
Sp99         1.000 0.359        1.000       0.186 0.359 1.000 0.035 1.000 1.000 1.000 -
Stickpile    1.000 0.359        1.000       0.014 0.244 1.000 0.004 1.000 1.000 1.000 1.000

P value adjustment method: holm

>
```

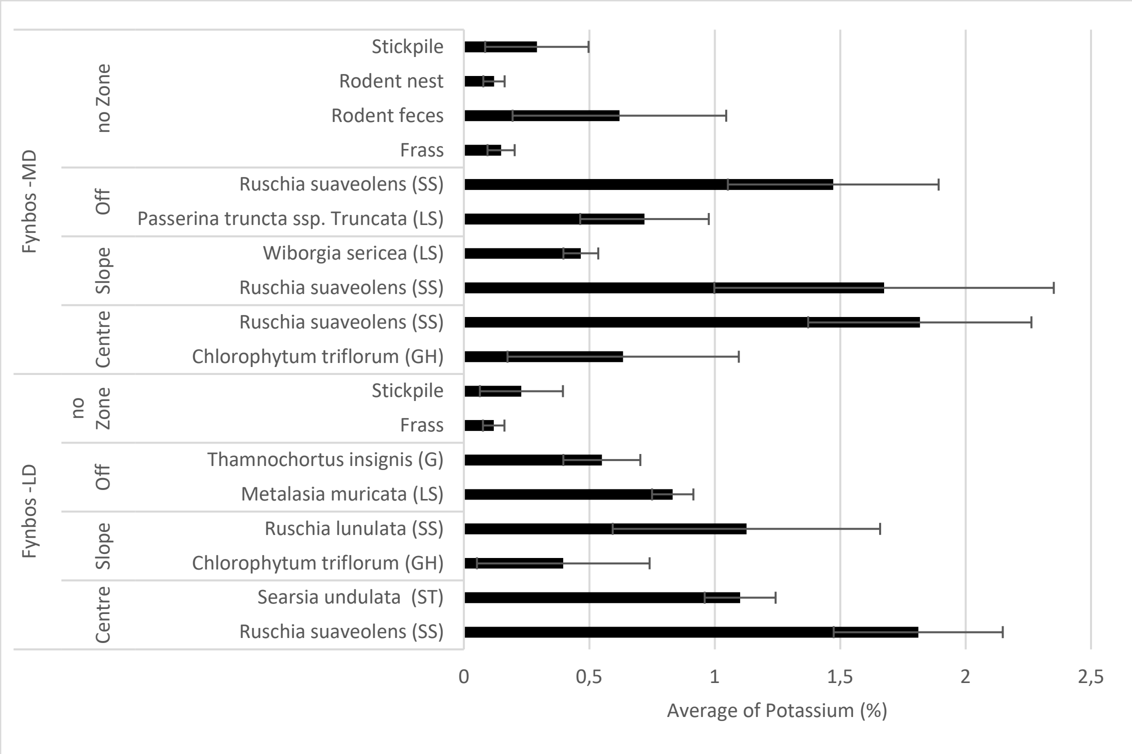
Figure HHH

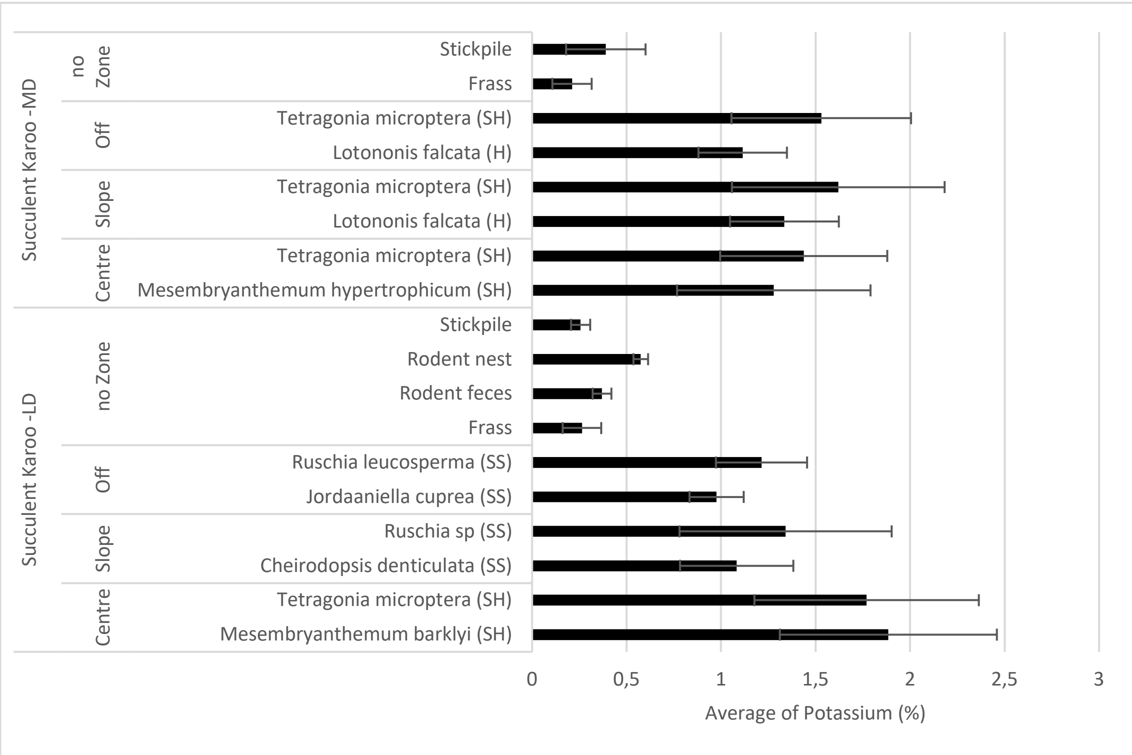
Figure III

Figure JJJ:

```
>
> #P
> kruskal.test(P_pc~Targeted,data=SKdataplants)

        Kruskal-Wallis rank sum test

data:  P_pc by Targeted
Kruskal-Wallis chi-squared = 15.023, df = 2, p-value = 0.0005467

> pairwise.wilcox.test(SKdataplants$P_pc,SKdataplants$Targeted)

        Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$P_pc and SKdataplants$Targeted

             Maybe  Not Targeted
Not Targeted 0.3140 -
Targeted     0.0076 0.0076

P value adjustment method: holm
>
> kruskal.test(P_pc~Targeted,data=Fyndataplants)

        Kruskal-Wallis rank sum test

data:  P_pc by Targeted
Kruskal-Wallis chi-squared = 1.164, df = 2, p-value = 0.5588

> pairwise.wilcox.test(Fyndataplants$P_pc,Fyndataplants$Targeted)

        Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$P_pc and Fyndataplants$Targeted

             Maybe Not Targeted
Not Targeted 1.00  -
Targeted     1.00  0.82

P value adjustment method: holm
>
> #P
> kruskal.test(P_pc~Zone,data=SKdataplants)

        Kruskal-Wallis rank sum test

data:  P_pc by Zone
Kruskal-Wallis chi-squared = 5.4088, df = 2, p-value = 0.06691

> pairwise.wilcox.test(SKdataplants$P_pc,SKdataplants$Zone)

        Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$P_pc and SKdataplants$Zone

      Centre Off
Off   0.044  -
Slope 0.290  0.664

P value adjustment method: holm
>
> kruskal.test(P_pc~Zone,data=Fyndataplants)

        Kruskal-Wallis rank sum test

data:  P_pc by Zone
Kruskal-Wallis chi-squared = 3.1567, df = 2, p-value = 0.2063

> pairwise.wilcox.test(Fyndataplants$P_pc,Fyndataplants$Zone)

        Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$P_pc and Fyndataplants$Zone

      Centre Off
Off   0.29   -
Slope 0.33   0.92

P value adjustment method: holm
>
```

Potassium



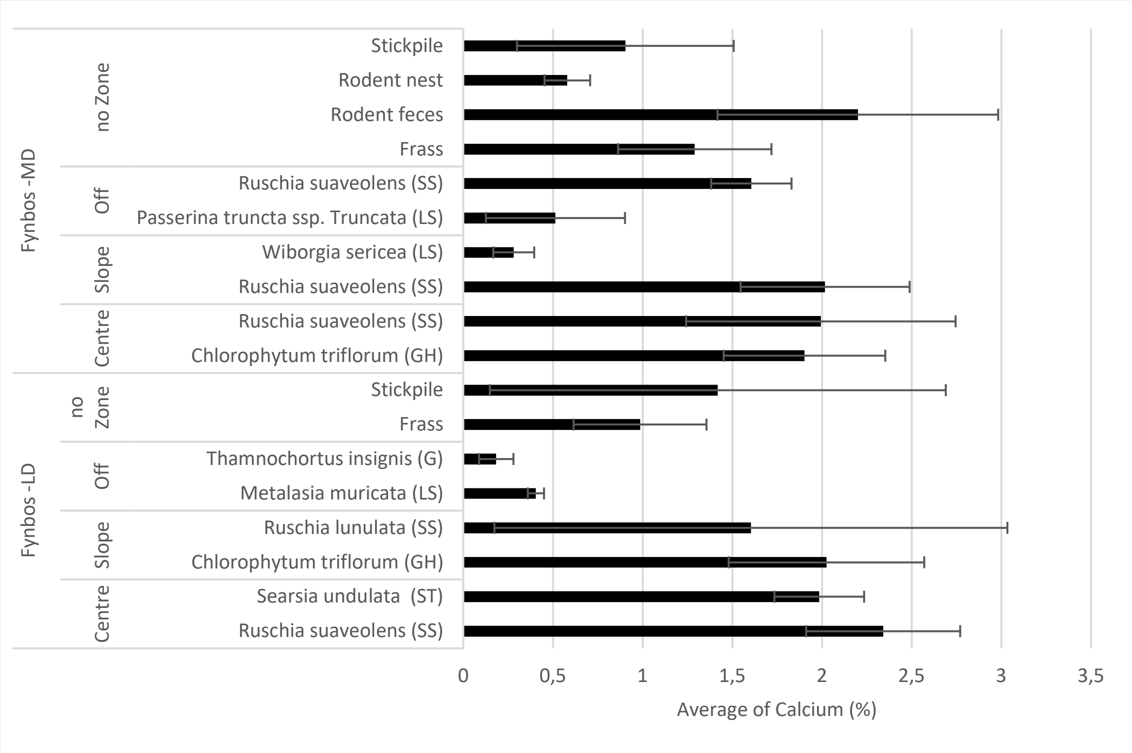


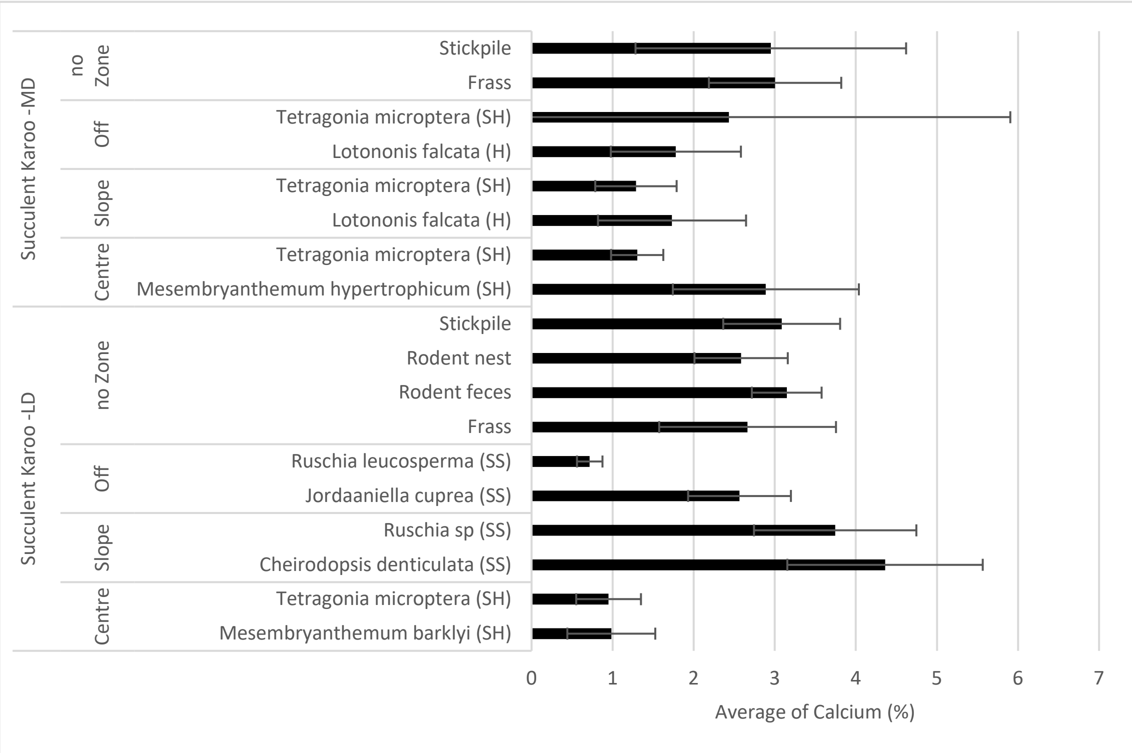

```
> #K
> options(digits = 5)
> #Biome
> wilcox.test(TSA$K_pc~TSA$Biome)

        Wilcoxon rank sum test with continuity correction

data:  TSA$K_pc by TSA$Biome
W = 7988, p-value = 3e-04
alternative hypothesis: true location shift is not equal to 0

> #Subsite
> wilcox.test(Fyndata$K_pc~Fyndata$Site)
```

```
         Wilcoxon rank sum test with continuity correction

data:  Fyndata$K_pc by Fyndata$Site
W = 2569, p-value = 0.52
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$K_pc~SKdata$Site)

         Wilcoxon rank sum test with continuity correction

data:  SKdata$K_pc by SKdata$Site
W = 2136, p-value = 0.11
alternative hypothesis: true location shift is not equal to 0

> #Year
> wilcox.test(Fyndata$K_pc~Fyndata$Year)

         Wilcoxon rank sum test with continuity correction

data:  Fyndata$K_pc by Fyndata$Year
W = 2786, p-value = 0.85
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$K_pc~SKdata$Year)

         Wilcoxon rank sum test with continuity correction

data:  SKdata$K_pc by SKdata$Year
W = 2544, p-value = 0.97
alternative hypothesis: true location shift is not equal to 0

> #Sample type
> kruskal.test(Fyndata$K_pc~Fyndata$Name)

         Kruskal-Wallis rank sum test

data:  Fyndata$K_pc by Fyndata$Name
Kruskal-Wallis chi-squared = 114, df = 11, p-value <2e-16

> pairwise.wilcox.test(Fyndata$K_pc,Fyndata$Name)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndata$K_pc and Fyndata$Name

             CH    Cl    CS    CT    FB    FE    FH    FI    Frass Rodent feces Rodent nest
Cl           1.000 -     -     -     -     -     -     -     -     -            -
CS           0.081 0.449 -     -     -     -     -     -     -     -            -
CT           0.008 0.446 0.287 -     -     -     -     -     -     -            -
FB           0.017 0.293 0.312 1.000 -     -     -     -     -     -            -
FE           0.061 0.400 0.006 0.003 5e-05 -     -     -     -     -            -
FH           0.008 0.446 0.042 1.000 1.000 0.003 -     -     -     -            -
FI           0.446 1.000 1.000 1.000 1.000 0.006 0.094 -     -     -            -
Frass        0.003 0.005 0.003 0.003 0.001 1e-07 0.003 0.003 -     -            -
Rodent feces 1.000 1.000 1.000 1.000 1.000 0.446 1.000 1.000 0.388 -            -
Rodent nest  1.000 1.000 1.000 1.000 1.000 0.570 1.000 1.000 1.000 1.000        -
Stickpile    0.002 0.009 0.002 0.066 0.370 3e-08 0.106 0.015 0.487 1.000        1.000

P value adjustment method: holm
> kruskal.test(SKdata$K_pc~SKdata$Name)

         Kruskal-Wallis rank sum test

data:  SKdata$K_pc by SKdata$Name
Kruskal-Wallis chi-squared = 108, df = 11, p-value <2e-16

> pairwise.wilcox.test(SKdata$K_pc,SKdata$Name)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdata$K_pc and SKdata$Name

             Frass Rodent feces Rodent nest Sp10  Sp15  Sp16  Sp25  Sp40  Sp5   Sp8   Sp99
Rodent feces 1.000 -            -           -     -     -     -     -     -     -     -
Rodent nest  0.097 0.767        -           -     -     -     -     -     -     -     -
Sp10         1e-07 0.074        0.074       -     -     -     -     -     -     -     -
Sp15         0.003 0.174        0.174       1.000 -     -     -     -     -     -     -
Sp16         0.003 0.174        0.174       0.107 0.853 -     -     -     -     -     -
Sp25         3e-05 0.142        0.142       0.767 1.000 0.321 -     -     -     -     -
Sp40         0.006 0.755        1.000       1.000 1.000 1.000 1.000 -     -     -     -
Sp5          0.003 0.174        0.174       1.000 0.599 0.031 0.218 1.000 -     -     -
Sp8          0.003 0.284        1.000       0.494 1.000 1.000 1.000 1.000 0.321 -     -
Sp99         0.003 0.174        0.283       1.000 1.000 1.000 1.000 1.000 1.000 1.000 -
Stickpile    0.767 1.000        0.437       9e-08 0.002 0.003 3e-05 0.006 0.002 0.004 0.003

P value adjustment method: holm
> #K
> kruskal.test(K_pc~Targeted,data=SKdataplants)

         Kruskal-Wallis rank sum test
```

```
data:  K_pc by Targeted
Kruskal-Wallis chi-squared = 3.0419, df = 2, p-value = 0.2185

> pairwise.wilcox.test(SKdataplants$K_pc,SKdataplants$Targeted)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$K_pc and SKdataplants$Targeted

             Maybe Not Targeted
Not Targeted 0.22  -
Targeted     0.42  0.41

P value adjustment method: holm
>
> kruskal.test(K_pc~Targeted,data=Fyndataplants)

         Kruskal-Wallis rank sum test

data:  K_pc by Targeted
Kruskal-Wallis chi-squared = 46.168, df = 2, p-value = 9.437e-11

> pairwise.wilcox.test(Fyndataplants$K_pc,Fyndataplants$Targeted)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$K_pc and Fyndataplants$Targeted

             Maybe   Not Targeted
Not Targeted 2.0e-08 -
Targeted     0.13    1.3e-08

P value adjustment method: holm
#K
> kruskal.test(K_pc~Zone,data=SKdataplants)

         Kruskal-Wallis rank sum test

data:  K_pc by Zone
Kruskal-Wallis chi-squared = 7.8194, df = 2, p-value = 0.02005

> pairwise.wilcox.test(SKdataplants$K_pc,SKdataplants$Zone)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$K_pc and SKdataplants$Zone

      Centre Off
Off   0.025  -
Slope 0.177  0.177

P value adjustment method: holm
>
> kruskal.test(K_pc~Zone,data=Fyndataplants)

         Kruskal-Wallis rank sum test

data:  K_pc by Zone
Kruskal-Wallis chi-squared = 10.441, df = 2, p-value = 0.005404

> pairwise.wilcox.test(Fyndataplants$K_pc,Fyndataplants$Zone)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$K_pc and Fyndataplants$Zone

      Centre Off
Off   0.0096 -
Slope 0.0191 0.5019

P value adjustment method: holm
```

Calcium



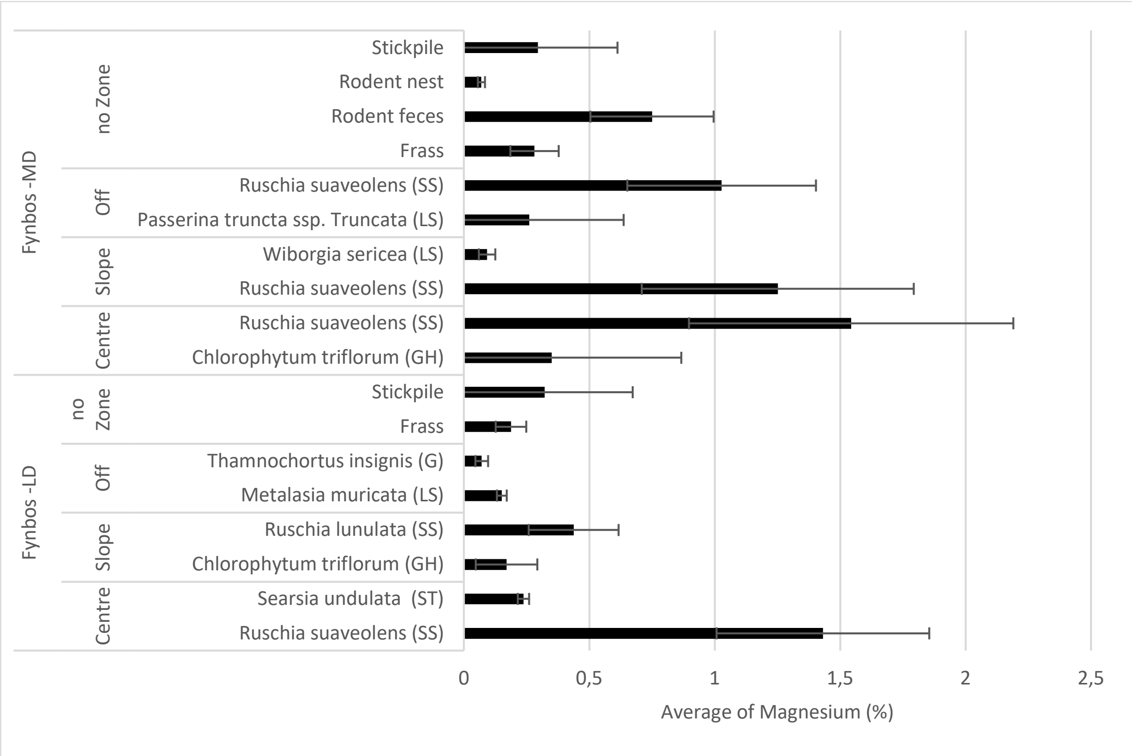


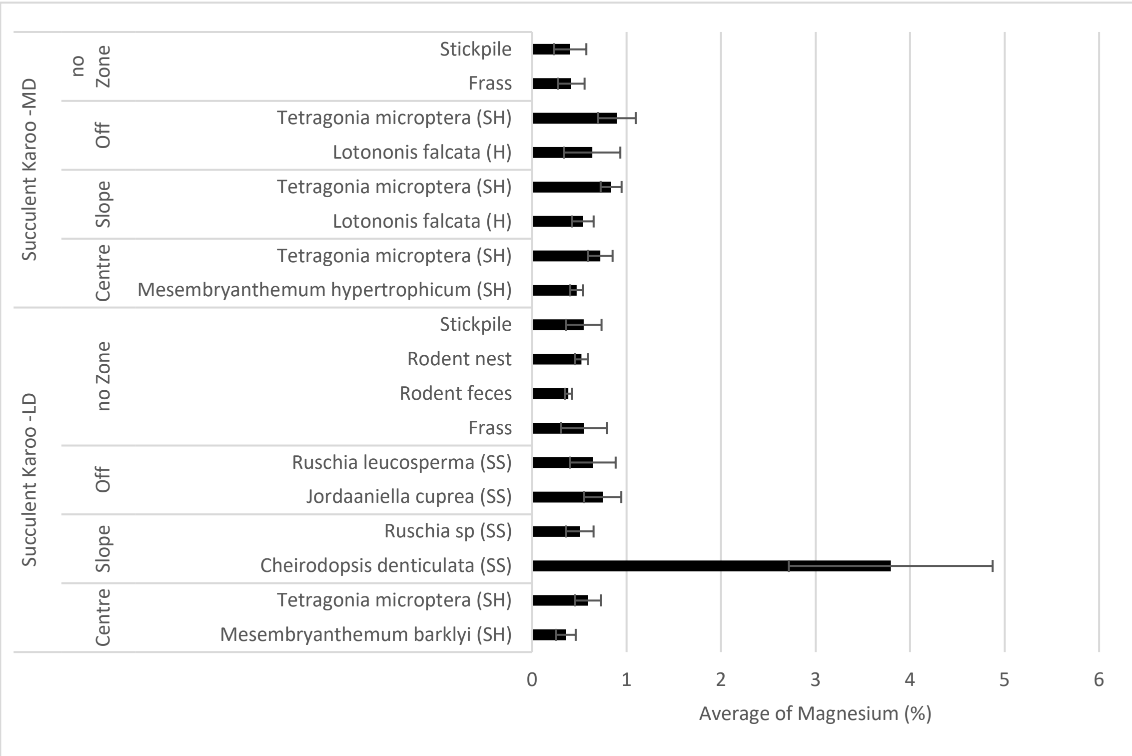

```
> #Ca
> options(digits = 5)
> #Biome
> wilcox.test(TSA$Ca_pc~TSA$Biome)

        Wilcoxon rank sum test with continuity correction

data:  TSA$Ca_pc by TSA$Biome
W = 5720, p-value = 1.2e-11
alternative hypothesis: true location shift is not equal to 0

> #Subsite
> wilcox.test(Fyndata$Ca_pc~Fyndata$Site)
```

```
        Wilcoxon rank sum test with continuity correction

data:  Fyndata$Ca_pc by Fyndata$Site
W = 2704, p-value = 0.9
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$Ca_pc~SKdata$Site)

        Wilcoxon rank sum test with continuity correction

data:  SKdata$Ca_pc by SKdata$Site
W = 2948, p-value = 0.089
alternative hypothesis: true location shift is not equal to 0

> #Year
> wilcox.test(Fyndata$Ca_pc~Fyndata$Year)

        Wilcoxon rank sum test with continuity correction

data:  Fyndata$Ca_pc by Fyndata$Year
W = 2773, p-value = 0.89
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$Ca_pc~SKdata$Year)

        Wilcoxon rank sum test with continuity correction

data:  SKdata$Ca_pc by SKdata$Year
W = 2536, p-value = 1
alternative hypothesis: true location shift is not equal to 0

> #Sample type
> kruskal.test(Fyndata$Ca_pc~Fyndata$Name)

        Kruskal-Wallis rank sum test

data:  Fyndata$Ca_pc by Fyndata$Name
Kruskal-Wallis chi-squared = 94.6, df = 11, p-value = 2e-15

> pairwise.wilcox.test(Fyndata$Ca_pc,Fyndata$Name)

        Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndata$Ca_pc and Fyndata$Name

             CH    Cl    CS    CT    FB    FE    FH    FI    Frass Rodent feces Rodent nest
Cl           0.541 -     -     -     -     -     -     -     -     -            -
CS           0.042 0.042 -     -     -     -     -     -     -     -            -
CT           0.042 0.008 0.205 -     -     -     -     -     -     -            -
FB           1.000 0.306 0.005 2e-04 -     -     -     -     -     -            -
FE           1.000 0.286 0.003 0.001 1.000 -     -     -     -     -            -
FH           0.042 0.008 0.734 0.734 2e-04 0.001 -     -     -     -            -
FI           0.042 0.280 1.000 0.280 0.008 0.003 0.779 -     -     -            -
Frass        0.014 1.000 0.004 0.003 0.002 7e-05 0.003 0.067 -     -            -
Rodent feces 1.000 1.000 0.541 0.400 1.000 1.000 0.400 0.740 0.734 -            -
Rodent nest  0.940 0.933 1.000 0.933 0.405 0.740 1.000 1.000 1.000 1.000        -
Stickpile    0.191 1.000 0.306 0.003 0.035 0.003 0.038 0.677 1.000 1.000        1.000

P value adjustment method: holm
> kruskal.test(SKdata$Ca_pc~SKdata$Name)

        Kruskal-Wallis rank sum test

data:  SKdata$Ca_pc by SKdata$Name
Kruskal-Wallis chi-squared = 89, df = 11, p-value = 2.7e-14

> pairwise.wilcox.test(SKdata$Ca_pc,SKdata$Name)

        Pairwise comparisons using Wilcoxon rank sum test

data:  SKdata$Ca_pc and SKdata$Name

             Frass Rodent feces Rodent nest Sp10  Sp15  Sp16  Sp25  Sp40  Sp5   Sp8   Sp99
Rodent feces 1.000 -            -           -     -     -     -     -     -     -     -
Rodent nest  1.000 1.000        -           -     -     -     -     -     -     -     -
Sp10         1e-04 0.136        0.166       -     -     -     -     -     -     -     -
Sp15         0.004 0.162        0.162       0.037 -     -     -     -     -     -     -
Sp16         1.000 1.000        1.000       0.006 0.044 -     -     -     -     -     -
Sp25         0.162 0.580        1.000       0.636 0.045 0.699 -     -     -     -     -
Sp40         0.777 1.000        1.000       0.008 0.009 0.886 0.042 -     -     -     -
Sp5          0.012 0.162        0.275       1.000 1.000 0.085 0.377 0.016 -     -     -
Sp8          0.203 1.000        0.275       0.005 0.009 0.162 0.011 1.000 0.009 -     -
Sp99         1.000 1.000        1.000       0.037 0.009 1.000 0.635 1.000 0.049 0.886 -
Stickpile    1.000 1.000        1.000       8e-06 0.003 1.000 0.037 0.607 0.005 0.176 1.000

P value adjustment method: holm

>
> #Ca
```

```
> kruskal.test(Ca_pc~Targeted,data=SKdataplants)

         Kruskal-Wallis rank sum test

data:  Ca_pc by Targeted
Kruskal-Wallis chi-squared = 30.636, df = 2, p-value = 2.226e-07

> pairwise.wilcox.test(SKdataplants$Ca_pc,SKdataplants$Targeted)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$Ca_pc and SKdataplants$Targeted

             Maybe   Not Targeted
Not Targeted 0.00044 -
Targeted     9.6e-08 0.50758

P value adjustment method: holm
>
> kruskal.test(Ca_pc~Targeted,data=Fyndataplants)

         Kruskal-Wallis rank sum test

data:  Ca_pc by Targeted
Kruskal-Wallis chi-squared = 21.122, df = 2, p-value = 2.591e-05

> pairwise.wilcox.test(Fyndataplants$Ca_pc,Fyndataplants$Targeted)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$Ca_pc and Fyndataplants$Targeted

             Maybe   Not Targeted
Not Targeted 5.2e-05 -
Targeted     0.80379 0.00064

P value adjustment method: holm
#Ca
> kruskal.test(Ca_pc~Zone,data=SKdataplants)

         Kruskal-Wallis rank sum test

data:  Ca_pc by Zone
Kruskal-Wallis chi-squared = 13.009, df = 2, p-value = 0.001497

> pairwise.wilcox.test(SKdataplants$Ca_pc,SKdataplants$Zone)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$Ca_pc and SKdataplants$Zone

      Centre Off
Off   0.5994 -
Slope 0.0042 0.0064

P value adjustment method: holm
>
> kruskal.test(Ca_pc~Zone,data=Fyndataplants)

         Kruskal-Wallis rank sum test

data:  Ca_pc by Zone
Kruskal-Wallis chi-squared = 38.386, df = 2, p-value = 4.619e-09

> pairwise.wilcox.test(Fyndataplants$Ca_pc,Fyndataplants$Zone)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$Ca_pc and Fyndataplants$Zone

      Centre  Off
Off   3.4e-09 -
Slope 0.0024  0.0024

P value adjustment method: holm
>
>
```

Magnesium



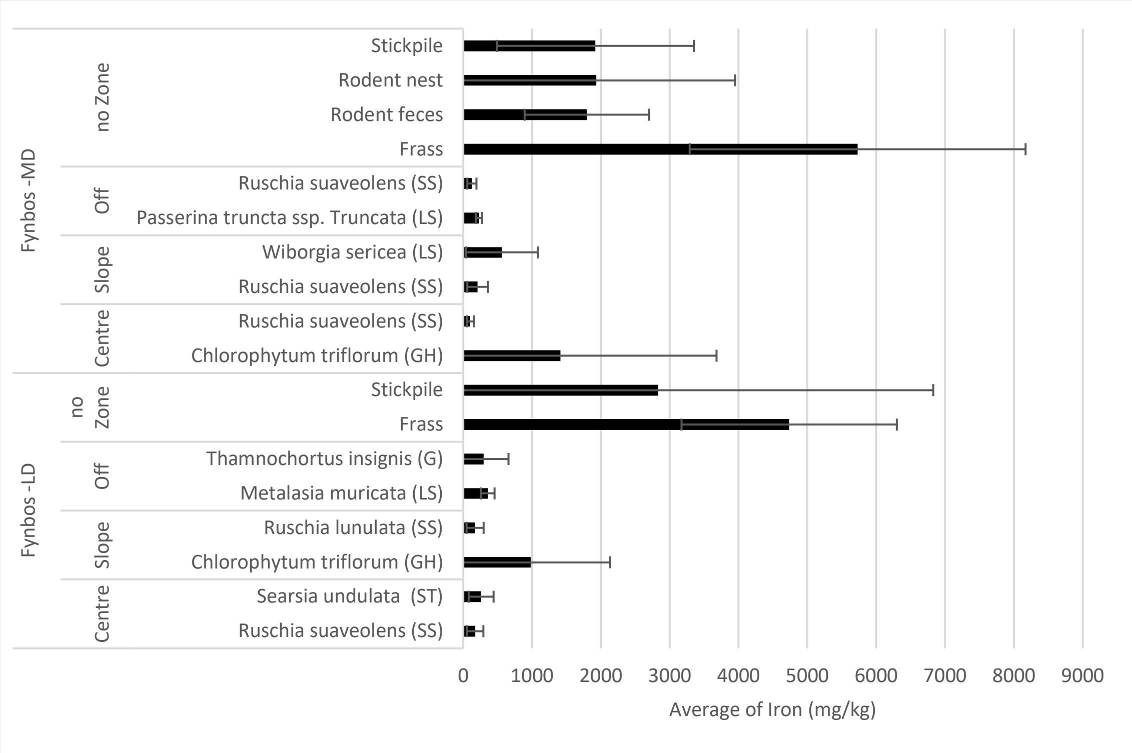


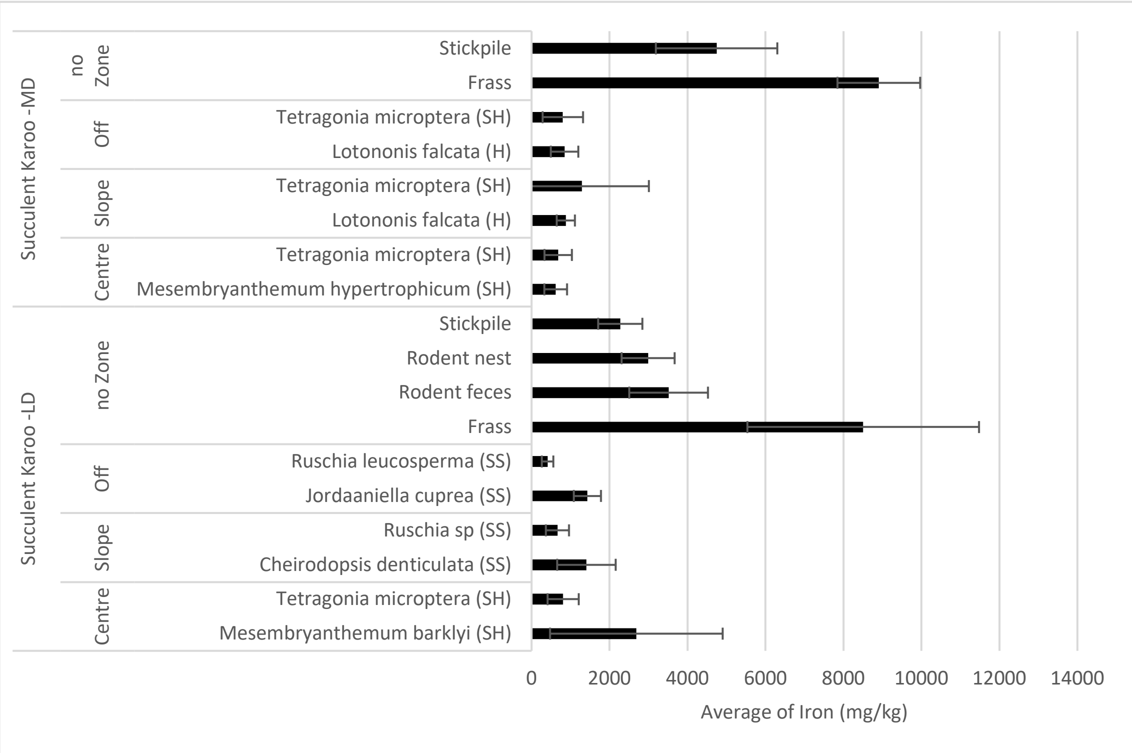

```
> #Mg
> options(digits = 5)
> #Biome
> wilcox.test(TSA$Mg_pc~TSA$Biome)

        Wilcoxon rank sum test with continuity correction

data:  TSA$Mg_pc by TSA$Biome
W = 5664, p-value = 7.2e-12
alternative hypothesis: true location shift is not equal to 0

> #Subsite
> wilcox.test(Fyndata$Mg_pc~Fyndata$Site)
```

```
         Wilcoxon rank sum test with continuity correction

data:  Fyndata$Mg_pc by Fyndata$Site
W = 2222, p-value = 0.049
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$Mg_pc~SKdata$Site)

         Wilcoxon rank sum test with continuity correction

data:  SKdata$Mg_pc by SKdata$Site
W = 2490, p-value = 0.88
alternative hypothesis: true location shift is not equal to 0

> #Year
> wilcox.test(Fyndata$Mg_pc~Fyndata$Year)

         Wilcoxon rank sum test with continuity correction

data:  Fyndata$Mg_pc by Fyndata$Year
W = 2470, p-value = 0.31
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$Mg_pc~SKdata$Year)

         Wilcoxon rank sum test with continuity correction

data:  SKdata$Mg_pc by SKdata$Year
W = 2194, p-value = 0.17
alternative hypothesis: true location shift is not equal to 0

> #Sample type
> kruskal.test(Fyndata$Mg_pc~Fyndata$Name)

         Kruskal-Wallis rank sum test

data:  Fyndata$Mg_pc by Fyndata$Name
Kruskal-Wallis chi-squared = 91.5, df = 11, p-value = 8.4e-15

> pairwise.wilcox.test(Fyndata$Mg_pc,Fyndata$Name)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndata$Mg_pc and Fyndata$Name

             CH    Cl    CS    CT    FB    FE    FH    FI    Frass Rodent feces Rodent nest
Cl           0.533 -     -     -     -     -     -     -     -     -            -
CS           0.047 0.533 -     -     -     -     -     -     -     -            -
CT           0.047 0.053 0.047 -     -     -     -     -     -     -            -
FB           0.854 0.490 1.000 0.036 -     -     -     -     -     -            -
FE           0.009 0.014 0.004 0.001 1e-04 -     -     -     -     -            -
FH           0.047 0.077 0.240 1.000 0.346 0.001 -     -     -     -            -
FI           0.533 1.000 1.000 0.105 1.000 0.008 0.633 -     -     -            -
Frass        1.000 0.171 0.326 0.004 1.000 3e-06 0.022 0.573 -     -            -
Rodent feces 0.633 1.000 0.633 0.633 0.655 1.000 0.633 1.000 0.299 -            -
Rodent nest  1.000 1.000 1.000 1.000 1.000 0.655 1.000 1.000 0.917 1.000        -
Stickpile    1.000 1.000 1.000 0.321 1.000 9e-07 0.917 1.000 1.000 1.000        1.000

P value adjustment method: holm
> kruskal.test(SKdata$Mg_pc~SKdata$Name)

         Kruskal-Wallis rank sum test

data:  SKdata$Mg_pc by SKdata$Name
Kruskal-Wallis chi-squared = 72.7, df = 11, p-value = 3.8e-11

> pairwise.wilcox.test(SKdata$Mg_pc,SKdata$Name)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdata$Mg_pc and SKdata$Name

             Frass Rodent feces Rodent nest Sp10  Sp15  Sp16  Sp25  Sp40  Sp5   Sp8   Sp99
Rodent feces 1.000 -            -           -     -     -     -     -     -     -     -
Rodent nest  1.000 1.000        -           -     -     -     -     -     -     -     -
Sp10         0.004 0.079        0.327       -     -     -     -     -     -     -     -
Sp15         1.000 1.000        1.000       1.000 -     -     -     -     -     -     -
Sp16         0.261 0.351        1.000       1.000 1.000 -     -     -     -     -     -
Sp25         1.000 0.197        1.000       0.013 1.000 0.351 -     -     -     -     -
Sp40         1.000 1.000        1.000       0.065 1.000 0.676 1.000 -     -     -     -
Sp5          1.000 1.000        0.350       0.002 0.676 0.049 0.022 1.000 -     -     -
Sp8          0.003 0.351        0.351       0.001 0.009 0.009 0.007 0.049 0.049 -     -
Sp99         1.000 1.000        1.000       0.005 1.000 0.056 1.000 1.000 1.000 0.009 -
Stickpile    1.000 1.000        1.000       3e-04 1.000 0.116 1.000 1.000 1.000 0.003 1.000

P value adjustment method: holm

>

> #Mg
> kruskal.test(Mg_pc~Targeted,data=SKdataplants)
```

```
        Kruskal-Wallis rank sum test

data:  Mg_pc by Targeted
Kruskal-Wallis chi-squared = 27.722, df = 2, p-value = 9.556e-07

> pairwise.wilcox.test(SKdataplants$Mg_pc,SKdataplants$Targeted)

        Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$Mg_pc and SKdataplants$Targeted

             Maybe   Not Targeted
Not Targeted 0.00063 -
Targeted     0.11400 9.9e-07

P value adjustment method: holm
>
> kruskal.test(Mg_pc~Targeted,data=Fyndataplants)

        Kruskal-Wallis rank sum test

data:  Mg_pc by Targeted
Kruskal-Wallis chi-squared = 51.858, df = 2, p-value = 5.486e-12

> pairwise.wilcox.test(Fyndataplants$Mg_pc,Fyndataplants$Targeted)

        Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$Mg_pc and Fyndataplants$Targeted

             Maybe   Not Targeted
Not Targeted 7.8e-10 -
Targeted     0.93    2.1e-09

P value adjustment method: holm
> #Mg
> kruskal.test(Mg_pc~Zone,data=SKdataplants)

        Kruskal-Wallis rank sum test

data:  Mg_pc by Zone
Kruskal-Wallis chi-squared = 14.759, df = 2, p-value = 0.0006239

> pairwise.wilcox.test(SKdataplants$Mg_pc,SKdataplants$Zone)

        Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$Mg_pc and SKdataplants$Zone

      Centre Off
Off   0.0022 -
Slope 0.0022 0.6157

P value adjustment method: holm
>
> kruskal.test(Mg_pc~Zone,data=Fyndataplants)

        Kruskal-Wallis rank sum test

data:  Mg_pc by Zone
Kruskal-Wallis chi-squared = 15.61, df = 2, p-value = 0.0004078

> pairwise.wilcox.test(Fyndataplants$Mg_pc,Fyndataplants$Zone)

        Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$Mg_pc and Fyndataplants$Zone

      Centre  Off
Off   0.00019 -
Slope 0.02021 0.39718

P value adjustment method: holm
```

Iron



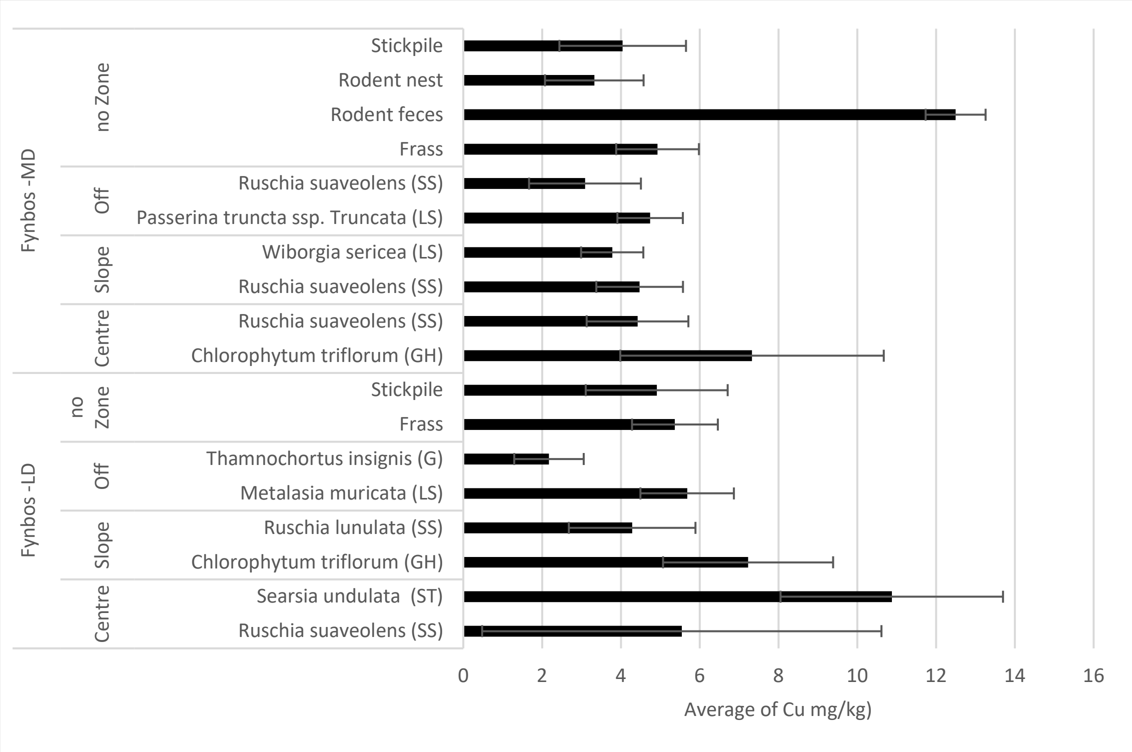


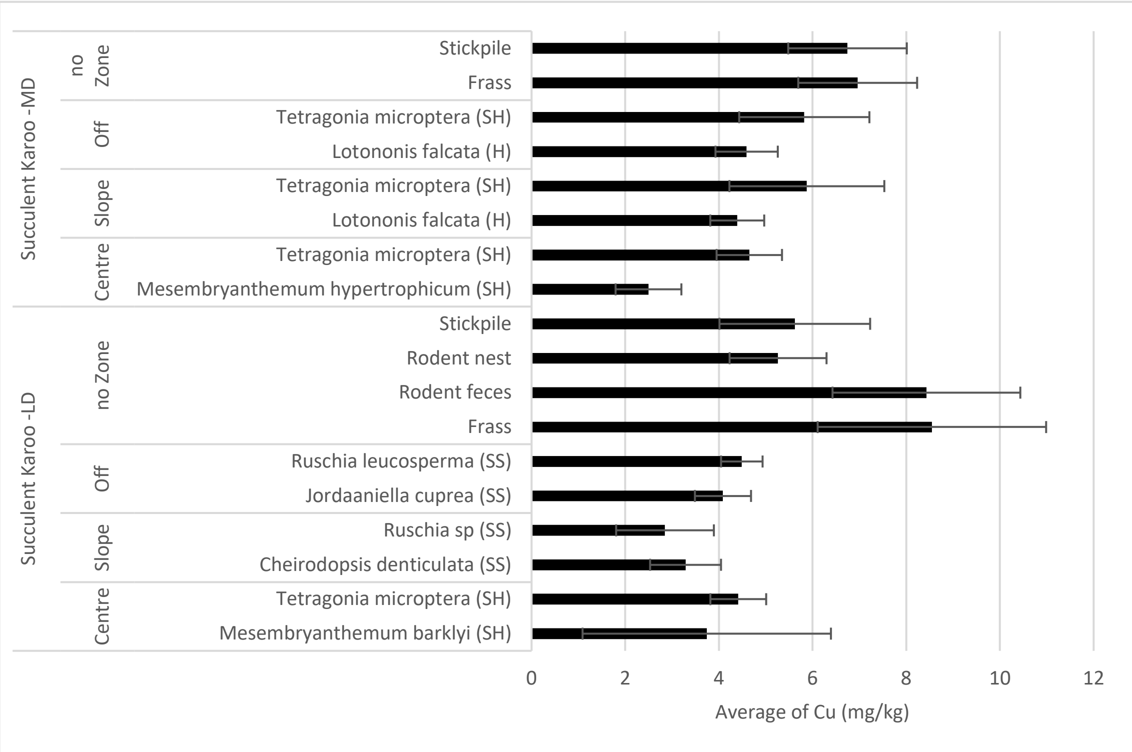

```
> #Fe
> #Biome
> wilcox.test(TSA$Fe_mgkg~TSA$Biome)

         Wilcoxon rank sum test with continuity correction

data:  TSA$Fe_mgkg by TSA$Biome
W = 6032, p-value = 2.3e-10
alternative hypothesis: true location shift is not equal to 0

> #Subsite
> wilcox.test(Fyndata$Fe_mgkg~Fyndata$Site)
```

```
	Wilcoxon rank sum test with continuity correction

data:  Fyndata$Fe_mgkg by Fyndata$Site
W = 2814, p-value = 0.77
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$Fe_mgkg~SKdata$Site)

	Wilcoxon rank sum test with continuity correction

data:  SKdata$Fe_mgkg by SKdata$Site
W = 2986, p-value = 0.063
alternative hypothesis: true location shift is not equal to 0

> #Year
> wilcox.test(Fyndata$Fe_mgkg~Fyndata$Year)

	Wilcoxon rank sum test with continuity correction

data:  Fyndata$Fe_mgkg by Fyndata$Year
W = 2664, p-value = 0.78
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$Fe_mgkg~SKdata$Year)

	Wilcoxon rank sum test with continuity correction

data:  SKdata$Fe_mgkg by SKdata$Year
W = 2251, p-value = 0.25
alternative hypothesis: true location shift is not equal to 0

> #Sample type
> kruskal.test(Fyndata$Fe_mgkg~Fyndata$Name)

	Kruskal-Wallis rank sum test

data:  Fyndata$Fe_mgkg by Fyndata$Name
Kruskal-Wallis chi-squared = 110, df = 11, p-value <2e-16

> pairwise.wilcox.test(Fyndata$Fe_mgkg,Fyndata$Name)

	Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndata$Fe_mgkg and Fyndata$Name

             CH    Cl    CS    CT    FB    FE    FH    FI    Frass Rodent feces Rodent nest
Cl           1.000 -     -     -     -     -     -     -     -     -            -
CS           0.872 0.531 -     -     -     -     -     -     -     -            -
CT           1.000 1.000 0.703 -     -     -     -     -     -     -            -
FB           1.000 0.557 1.000 1.000 -     -     -     -     -     -            -
FE           0.498 1.000 0.016 1.000 0.001 -     -     -     -     -            -
FH           1.000 0.872 1.000 1.000 1.000 0.011 -     -     -     -            -
FI           1.000 1.000 0.301 1.000 0.940 0.325 0.703 -     -     -            -
Frass        3e-05 3e-05 3e-05 3e-05 6e-04 4e-13 3e-05 3e-05 -     -            -
Rodent feces 0.497 0.497 0.497 0.776 1.000 0.015 1.000 0.497 0.174 -            -
Rodent nest  1.000 1.000 1.000 1.000 1.000 0.160 1.000 1.000 1.000 1.000        -
Stickpile    1e-05 7e-06 1e-05 1e-04 0.119 6e-15 0.009 7e-06 7e-06 1.000        1.000

P value adjustment method: holm
> kruskal.test(SKdata$Fe_mgkg~SKdata$Name)

	Kruskal-Wallis rank sum test

data:  SKdata$Fe_mgkg by SKdata$Name
Kruskal-Wallis chi-squared = 108, df = 11, p-value <2e-16

> pairwise.wilcox.test(SKdata$Fe_mgkg,SKdata$Name)

	Pairwise comparisons using Wilcoxon rank sum test

data:  SKdata$Fe_mgkg and SKdata$Name

             Frass Rodent feces Rodent nest Sp10  Sp15  Sp16  Sp25  Sp40  Sp5   Sp8   Sp99
Rodent feces 0.072 -            -           -     -     -     -     -     -     -     -
Rodent nest  0.072 1.000        -           -     -     -     -     -     -     -     -
Sp10         2e-11 0.025        0.025       -     -     -     -     -     -     -     -
Sp15         3e-05 0.145        0.145       0.516 -     -     -     -     -     -     -
Sp16         1e-04 0.145        0.145       0.052 0.008 -     -     -     -     -     -
Sp25         2e-08 0.023        0.023       1.000 0.004 0.055 -     -     -     -     -
Sp40         3e-05 0.145        0.145       1.000 1.000 0.015 1.000 -     -     -     -
Sp5          7e-04 1.000        1.000       0.389 0.008 1.000 1.000 0.189 -     -     -
Sp8          1e-04 0.145        0.388       0.547 0.015 1.000 0.761 0.369 1.000 -     -
Sp99         3e-05 0.145        0.145       1.000 1.000 0.008 1.000 1.000 0.563 0.271 -
Stickpile    2e-06 1.000        1.000       3e-09 2e-05 0.002 1e-08 2e-05 1.000 0.020 2e-05

P value adjustment method: holm

>

> #Fe
> kruskal.test(Fe_mgkg~Targeted,data=SKdataplants)
```

```
         Kruskal-Wallis rank sum test

data:  Fe_mgkg by Targeted
Kruskal-Wallis chi-squared = 1.2985, df = 2, p-value = 0.5224

> pairwise.wilcox.test(SKdataplants$Fe_mgkg,SKdataplants$Targeted)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$Fe_mgkg and SKdataplants$Targeted

             Maybe Not Targeted
Not Targeted 1     -
Targeted     1     1

P value adjustment method: holm
>
> kruskal.test(Fe_mgkg~Targeted,data=Fyndataplants)

         Kruskal-Wallis rank sum test

data:  Fe_mgkg by Targeted
Kruskal-Wallis chi-squared = 20.57, df = 2, p-value = 3.414e-05

> pairwise.wilcox.test(Fyndataplants$Fe_mgkg,Fyndataplants$Targeted)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$Fe_mgkg and Fyndataplants$Targeted

             Maybe  Not Targeted
Not Targeted 0.0002 -
Targeted     0.7238 0.0002

P value adjustment method: holm
>
> #Fe
> kruskal.test(Fe_mgkg~Zone,data=SKdataplants)

         Kruskal-Wallis rank sum test

data:  Fe_mgkg by Zone
Kruskal-Wallis chi-squared = 0.44314, df = 2, p-value = 0.8013

> pairwise.wilcox.test(SKdataplants$Fe_mgkg,SKdataplants$Zone)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$Fe_mgkg and SKdataplants$Zone

      Centre Off
Off   1      -
Slope 1      1

P value adjustment method: holm
>
> kruskal.test(Fe_mgkg~Zone,data=Fyndataplants)

         Kruskal-Wallis rank sum test

data:  Fe_mgkg by Zone
Kruskal-Wallis chi-squared = 4.7366, df = 2, p-value = 0.09364

> pairwise.wilcox.test(Fyndataplants$Fe_mgkg,Fyndataplants$Zone)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$Fe_mgkg and Fyndataplants$Zone

      Centre Off
Off   0.39   -
Slope 0.15   0.39

P value adjustment method: holm
```

Copper



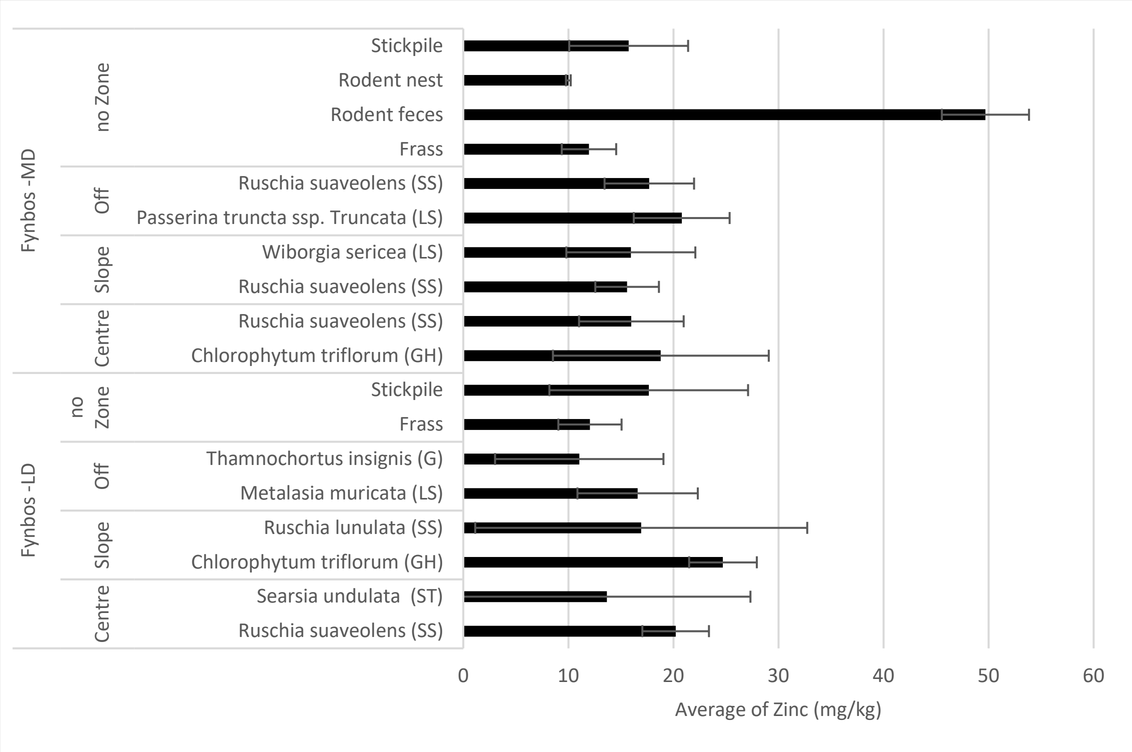


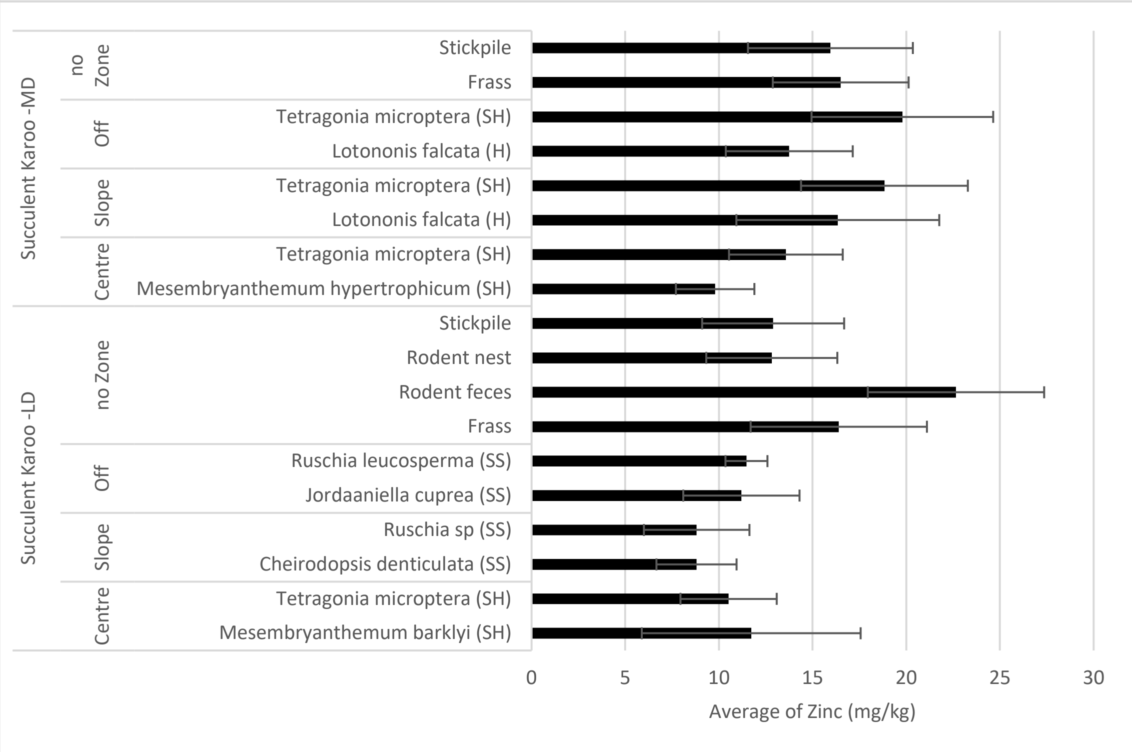

```
> #Cu
> #Biome
> wilcox.test(TSA$Cu_mgkg~TSA$Biome)

        Wilcoxon rank sum test with continuity correction

data:  TSA$Cu_mgkg by TSA$Biome
W = 9806, p-value = 0.28
alternative hypothesis: true location shift is not equal to 0

> #Subsite
> wilcox.test(Fyndata$Cu_mgkg~Fyndata$Site)
```

```
         Wilcoxon rank sum test with continuity correction

data:  Fyndata$Cu_mgkg by Fyndata$Site
W = 3166, p-value = 0.099
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$Cu_mgkg~SKdata$Site)

         Wilcoxon rank sum test with continuity correction

data:  SKdata$Cu_mgkg by SKdata$Site
W = 2258, p-value = 0.27
alternative hypothesis: true location shift is not equal to 0

> #Year
> wilcox.test(Fyndata$Cu_mgkg~Fyndata$Year)

         Wilcoxon rank sum test with continuity correction

data:  Fyndata$Cu_mgkg by Fyndata$Year
W = 2782, p-value = 0.86
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$Cu_mgkg~SKdata$Year)

         Wilcoxon rank sum test with continuity correction

data:  SKdata$Cu_mgkg by SKdata$Year
W = 1978, p-value = 0.024
alternative hypothesis: true location shift is not equal to 0

> #Sample type
> kruskal.test(Fyndata$Cu_mgkg~Fyndata$Name)

         Kruskal-Wallis rank sum test

data:  Fyndata$Cu_mgkg by Fyndata$Name
Kruskal-Wallis chi-squared = 71.5, df = 11, p-value = 6.2e-11

> pairwise.wilcox.test(Fyndata$Cu_mgkg,Fyndata$Name)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndata$Cu_mgkg and Fyndata$Name

             CH    Cl    CS    CT    FB    FE    FH    FI    Frass Rodent feces Rodent nest
Cl           0.009 -     -     -     -     -     -     -     -     -            -
CS           0.009 1.000 -     -     -     -     -     -     -     -            -
CT           0.009 0.205 0.032 -     -     -     -     -     -     -            -
FB           0.060 0.287 1.000 0.001 -     -     -     -     -     -            -
FE           0.003 1.000 0.259 0.015 0.002 -     -     -     -     -            -
FH           0.009 1.000 0.054 0.287 0.003 1.000 -     -     -     -            -
FI           0.009 1.000 1.000 0.054 0.448 1.000 1.000 -     -     -            -
Frass        3e-05 1.000 1.000 2e-04 0.165 0.075 0.287 1.000 -     -            -
Rodent feces 1.000 0.448 0.448 0.448 0.448 0.332 0.448 0.448 0.050 -            -
Rodent nest  1.000 1.000 1.000 1.000 1.000 1.000 1.000 1.000 1.000 1.000        -
Stickpile    0.002 1.000 1.000 0.081 0.020 1.000 1.000 1.000 1.000 0.250        1.000

P value adjustment method: holm
> kruskal.test(SKdata$Cu_mgkg~SKdata$Name)

         Kruskal-Wallis rank sum test

data:  SKdata$Cu_mgkg by SKdata$Name
Kruskal-Wallis chi-squared = 91.6, df = 11, p-value = 8e-15

> pairwise.wilcox.test(SKdata$Cu_mgkg,SKdata$Name)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdata$Cu_mgkg and SKdata$Name

             Frass Rodent feces Rodent nest Sp10  Sp15  Sp16  Sp25  Sp40  Sp5   Sp8   Sp99
Rodent feces 1.000 -            -           -     -     -     -     -     -     -     -
Rodent nest  0.463 1.000        -           -     -     -     -     -     -     -     -
Sp10         1e-04 0.171        1.000       -     -     -     -     -     -     -     -
Sp15         0.003 0.162        1.000       1.000 -     -     -     -     -     -     -
Sp16         0.003 0.162        0.735       0.162 1.000 -     -     -     -     -     -
Sp25         4e-05 0.025        1.000       0.735 1.000 1.000 -     -     -     -     -
Sp40         0.003 0.162        0.242       0.004 0.029 0.217 0.014 -     -     -     -
Sp5          0.014 0.162        1.000       1.000 1.000 1.000 1.000 1.000 -     -     -
Sp8          0.003 0.162        0.463       0.008 0.049 1.000 0.136 1.000 1.000 -     -
Sp99         0.003 0.162        0.162       0.002 0.016 0.082 0.003 1.000 1.000 0.872 -
Stickpile    0.151 0.872        1.000       0.653 0.130 0.027 0.020 2e-04 0.738 1e-03 2e-04

P value adjustment method: holm

>

> #Cu
>
```

```
> kruskal.test(Cu_mgkg~Targeted,data=SKdataplants)

	Kruskal-Wallis rank sum test

data:  Cu_mgkg by Targeted
Kruskal-Wallis chi-squared = 27.523, df = 2, p-value = 1.055e-06

> pairwise.wilcox.test(SKdataplants$Cu_mgkg,SKdataplants$Targeted)

	Pairwise comparisons using Wilcoxon rank sum test 

data:  SKdataplants$Cu_mgkg and SKdataplants$Targeted 

             Maybe   Not Targeted
Not Targeted 0.616   -           
Targeted     6.2e-07 0.011       

P value adjustment method: holm 
> 
> kruskal.test(Cu_mgkg~Targeted,data=Fyndataplants)

	Kruskal-Wallis rank sum test

data:  Cu_mgkg by Targeted
Kruskal-Wallis chi-squared = 8.384, df = 2, p-value = 0.01512

> pairwise.wilcox.test(Fyndataplants$Cu_mgkg,Fyndataplants$Targeted)

	Pairwise comparisons using Wilcoxon rank sum test 

data:  Fyndataplants$Cu_mgkg and Fyndataplants$Targeted 

             Maybe Not Targeted
Not Targeted 0.092 -           
Targeted     0.104 0.038       

P value adjustment method: holm 
> 
> #Cu
> 
> kruskal.test(Cu_mgkg~Zone,data=SKdataplants)

	Kruskal-Wallis rank sum test

data:  Cu_mgkg by Zone
Kruskal-Wallis chi-squared = 4.9277, df = 2, p-value = 0.08511

> pairwise.wilcox.test(SKdataplants$Cu_mgkg,SKdataplants$Zone)

	Pairwise comparisons using Wilcoxon rank sum test 

data:  SKdataplants$Cu_mgkg and SKdataplants$Zone 

      Centre Off 
Off   0.15   -   
Slope 0.82   0.13

P value adjustment method: holm 
> 
> kruskal.test(Cu_mgkg~Zone,data=Fyndataplants)

	Kruskal-Wallis rank sum test

data:  Cu_mgkg by Zone
Kruskal-Wallis chi-squared = 13.743, df = 2, p-value = 0.001037

> pairwise.wilcox.test(Fyndataplants$Cu_mgkg,Fyndataplants$Zone)

	Pairwise comparisons using Wilcoxon rank sum test 

data:  Fyndataplants$Cu_mgkg and Fyndataplants$Zone 

      Centre Off   
Off   0.0013 -     
Slope 0.0644 0.0644

P value adjustment method: holm 
```

Zinc



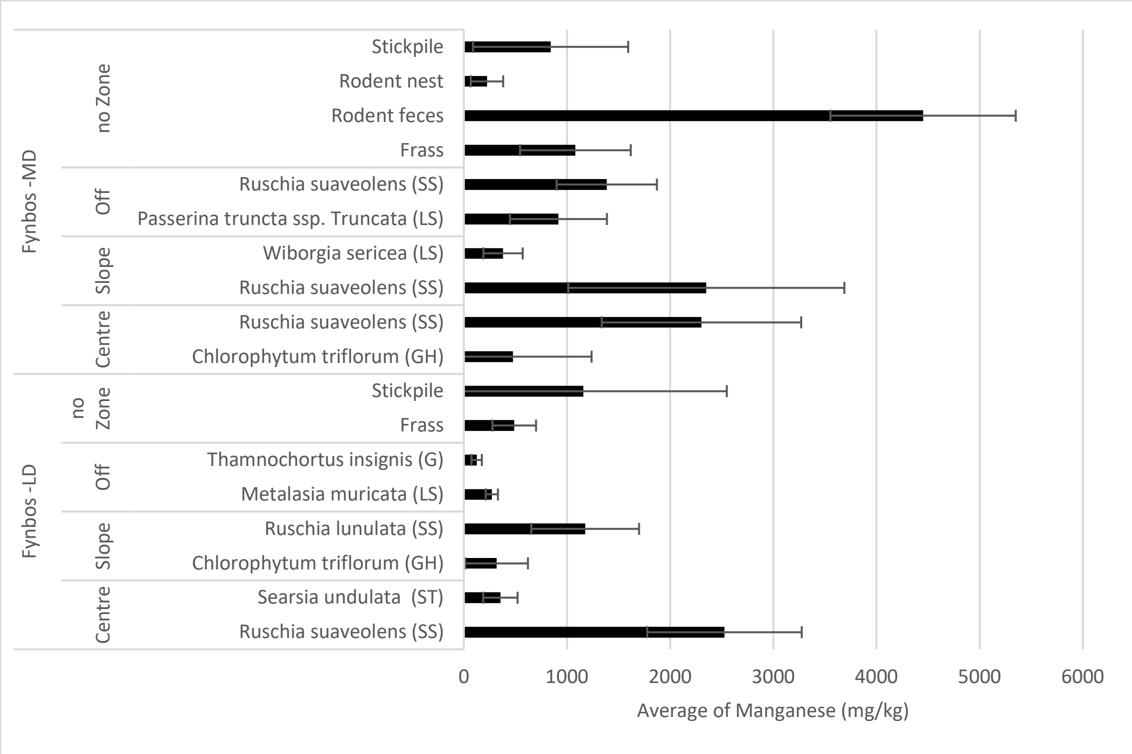


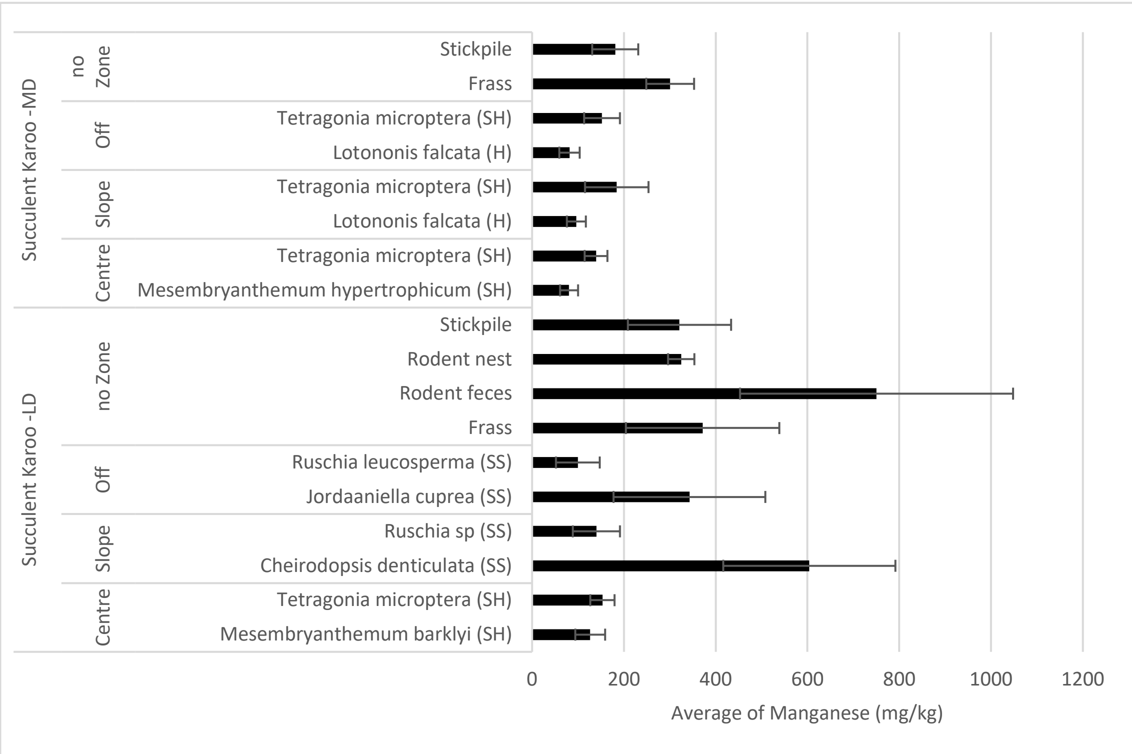

```
> #Zn
> #Biome
> wilcox.test(TSA$Zn_mgkg~TSA$Biome)

        Wilcoxon rank sum test with continuity correction

data:  TSA$Zn_mgkg by TSA$Biome
W = 12692, p-value = 0.0033
alternative hypothesis: true location shift is not equal to 0

> #Subsite
> wilcox.test(Fyndata$Zn_mgkg~Fyndata$Site)

        Wilcoxon rank sum test with continuity correction
```

```
data:  Fyndata$Zn_mgkg by Fyndata$Site
W = 2406, p-value = 0.21
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$Zn_mgkg~SKdata$Site)

         Wilcoxon rank sum test with continuity correction

data:  SKdata$Zn_mgkg by SKdata$Site
W = 1580, p-value = 0.00012
alternative hypothesis: true location shift is not equal to 0

> #Year
> wilcox.test(Fyndata$Zn_mgkg~Fyndata$Year)

         Wilcoxon rank sum test with continuity correction

data:  Fyndata$Zn_mgkg by Fyndata$Year
W = 2552, p-value = 0.48
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$Zn_mgkg~SKdata$Year)

         Wilcoxon rank sum test with continuity correction

data:  SKdata$Zn_mgkg by SKdata$Year
W = 1338, p-value = 1.2e-06
alternative hypothesis: true location shift is not equal to 0

> #Sample type
> kruskal.test(Fyndata$Zn_mgkg~Fyndata$Name)

         Kruskal-Wallis rank sum test

data:  Fyndata$Zn_mgkg by Fyndata$Name
Kruskal-Wallis chi-squared = 49.4, df = 11, p-value = 8.2e-07

> pairwise.wilcox.test(Fyndata$Zn_mgkg,Fyndata$Name)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndata$Zn_mgkg and Fyndata$Name

             CH    Cl    CS    CT    FB    FE    FH    FI    Frass Rodent feces Rodent nest
Cl           0.578 -     -     -     -     -     -     -     -     -            -
CS           1.000 1.000 -     -     -     -     -     -     -     -            -
CT           0.551 0.551 0.551 -     -     -     -     -     -     -            -
FB           0.986 1.000 1.000 0.059 -     -     -     -     -     -            -
FE           0.892 1.000 1.000 0.030 1.000 -     -     -     -     -            -
FH           1.000 1.000 1.000 0.551 1.000 1.000 -     -     -     -            -
FI           0.171 1.000 1.000 0.551 1.000 0.426 0.986 -     -     -            -
Frass        1.000 0.075 0.551 0.976 0.060 0.006 1.000 0.007 -     -            -
Rodent feces 0.570 0.570 0.570 0.570 0.578 0.073 0.570 0.570 0.381 -            -
Rodent nest  1.000 1.000 1.000 1.000 1.000 0.203 1.000 1.000 1.000 1.000        -
Stickpile    1.000 1.000 1.000 0.491 1.000 1.000 1.000 1.000 0.570 0.034        1.000

P value adjustment method: holm
> kruskal.test(SKdata$Zn_mgkg~SKdata$Name)

         Kruskal-Wallis rank sum test

data:  SKdata$Zn_mgkg by SKdata$Name
Kruskal-Wallis chi-squared = 53.2, df = 11, p-value = 1.7e-07

> pairwise.wilcox.test(SKdata$Zn_mgkg,SKdata$Name)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdata$Zn_mgkg and SKdata$Name

             Frass Rodent feces Rodent nest Sp10  Sp15  Sp16  Sp25  Sp40  Sp5   Sp8   Sp99
Rodent feces 0.803 -            -           -     -     -     -     -     -     -     -
Rodent nest  1.000 1.000        -           -     -     -     -     -     -     -     -
Sp10         1.000 0.634        1.000       -     -     -     -     -     -     -     -
Sp15         0.056 0.210        1.000       0.375 -     -     -     -     -     -     -
Sp16         0.084 0.210        1.000       0.728 1.000 -     -     -     -     -     -
Sp25         1.000 0.804        1.000       1.000 0.916 1.000 -     -     -     -     -
Sp40         0.001 0.210        1.000       0.046 1.000 1.000 0.046 -     -     -     -
Sp5          1.000 0.210        1.000       1.000 1.000 1.000 1.000 1.000 -     -     -
Sp8          9e-04 0.210        1.000       0.009 0.827 1.000 0.046 1.000 1.000 -     -
Sp99         0.008 0.210        1.000       0.028 1.000 1.000 0.102 1.000 1.000 1.000 -
Stickpile    1.000 0.128        1.000       1.000 1.000 1.000 1.000 0.062 1.000 0.035 0.210

P value adjustment method: holm

>

> #Zn
> kruskal.test(Zn_mgkg~Targeted,data=SKdataplants)
```

```
         Kruskal-Wallis rank sum test

data:  Zn_mgkg by Targeted
Kruskal-Wallis chi-squared = 22.613, df = 2, p-value = 1.229e-05

> pairwise.wilcox.test(SKdataplants$Zn_mgkg,SKdataplants$Targeted)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$Zn_mgkg and SKdataplants$Targeted

             Maybe   Not Targeted
Not Targeted 0.142   -
Targeted     2.1e-05 0.028

P value adjustment method: holm
>
> kruskal.test(Zn_mgkg~Targeted,data=Fyndataplants)

         Kruskal-Wallis rank sum test

data:  Zn_mgkg by Targeted
Kruskal-Wallis chi-squared = 10.947, df = 2, p-value = 0.004197

> pairwise.wilcox.test(Fyndataplants$Zn_mgkg,Fyndataplants$Targeted)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$Zn_mgkg and Fyndataplants$Targeted

             Maybe Not Targeted
Not Targeted 0.178 -
Targeted     0.006 0.043

P value adjustment method: holm
> #Zn
> kruskal.test(Zn_mgkg~Zone,data=SKdataplants)

         Kruskal-Wallis rank sum test

data:  Zn_mgkg by Zone
Kruskal-Wallis chi-squared = 3.4837, df = 2, p-value = 0.1752

> pairwise.wilcox.test(SKdataplants$Zn_mgkg,SKdataplants$Zone)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$Zn_mgkg and SKdataplants$Zone

      Centre Off
Off   0.16   -
Slope 0.65   0.65

P value adjustment method: holm
>
> kruskal.test(Zn_mgkg~Zone,data=Fyndataplants)

         Kruskal-Wallis rank sum test

data:  Zn_mgkg by Zone
Kruskal-Wallis chi-squared = 1.226, df = 2, p-value = 0.5417

> pairwise.wilcox.test(Fyndataplants$Zn_mgkg,Fyndataplants$Zone)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$Zn_mgkg and Fyndataplants$Zone

      Centre Off
Off   1.0    -
Slope 0.7    1.0

P value adjustment method: holm
```

Manganese



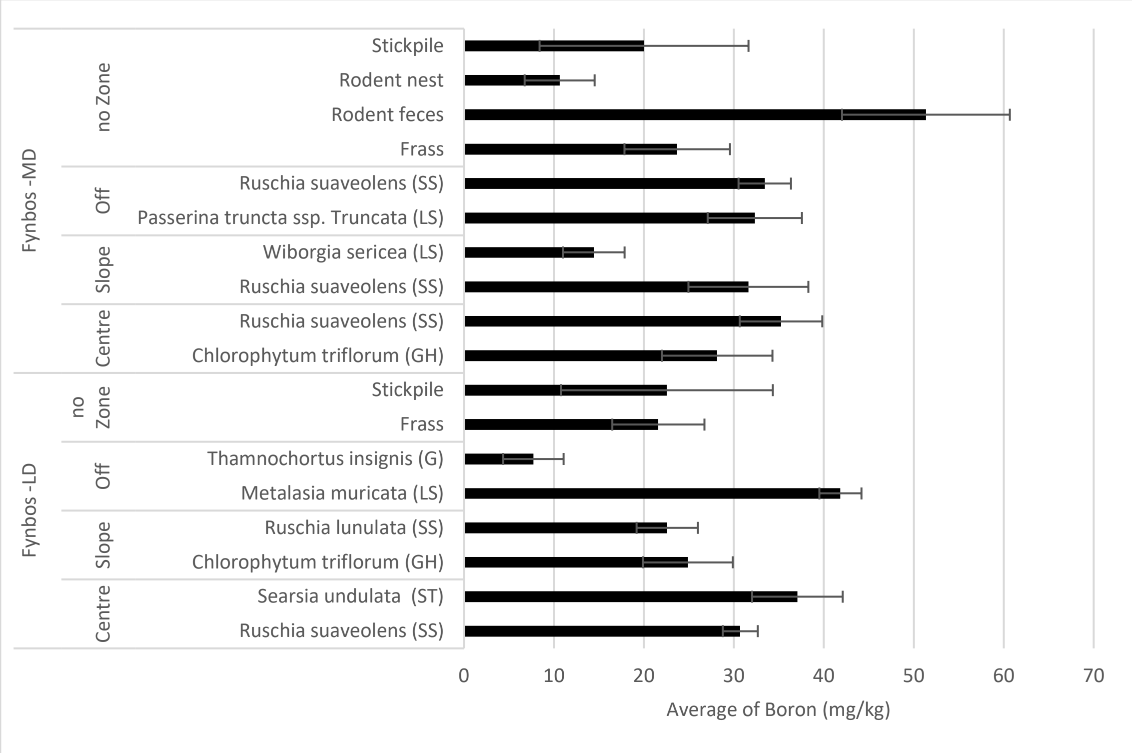


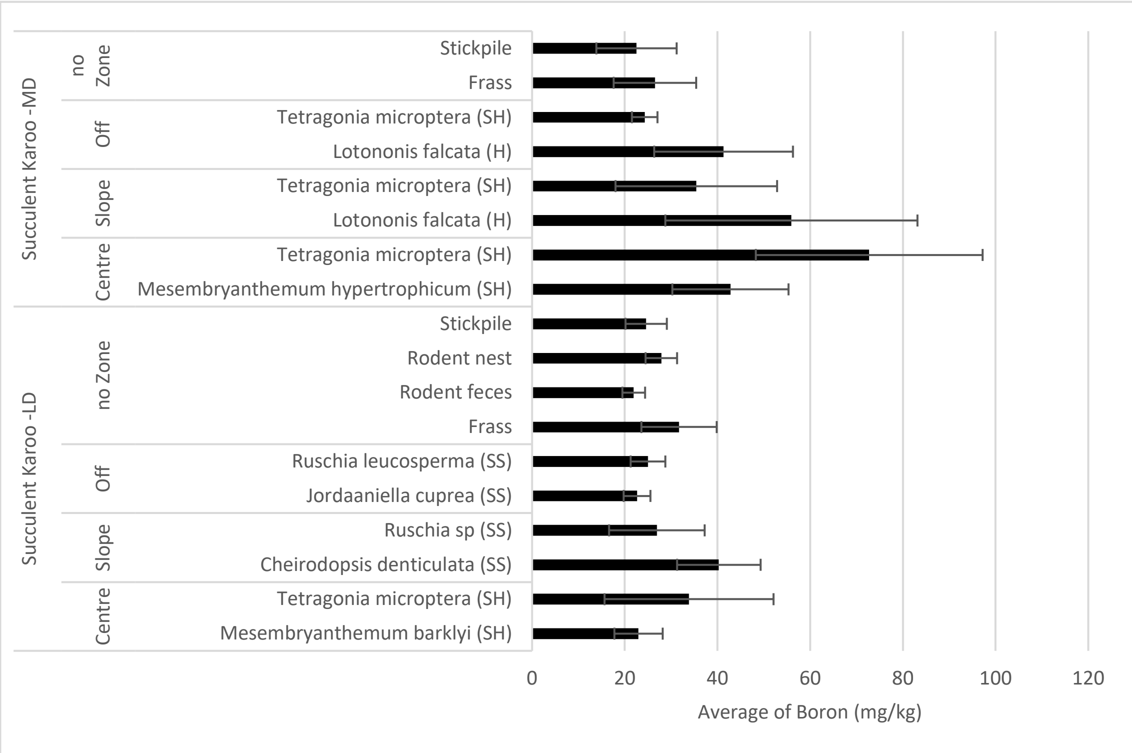

```
> #Mn
> #Biome
> wilcox.test(TSA$Mn_mgkg~TSA$Biome)

        Wilcoxon rank sum test with continuity correction

data:  TSA$Mn_mgkg by TSA$Biome
W = 16981, p-value <2e-16
alternative hypothesis: true location shift is not equal to 0

> #Subsite
> wilcox.test(Fyndata$Mn_mgkg~Fyndata$Site)
```

```
         Wilcoxon rank sum test with continuity correction

data:  Fyndata$Mn_mgkg by Fyndata$Site
W = 1854, p-value = 0.00073
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$Mn_mgkg~SKdata$Site)

         Wilcoxon rank sum test with continuity correction

data:  SKdata$Mn_mgkg by SKdata$Site
W = 3772, p-value = 4.4e-07
alternative hypothesis: true location shift is not equal to 0

> #Year
> wilcox.test(Fyndata$Mn_mgkg~Fyndata$Year)

         Wilcoxon rank sum test with continuity correction

data:  Fyndata$Mn_mgkg by Fyndata$Year
W = 2533, p-value = 0.44
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$Mn_mgkg~SKdata$Year)

         Wilcoxon rank sum test with continuity correction

data:  SKdata$Mn_mgkg by SKdata$Year
W = 2307, p-value = 0.36
alternative hypothesis: true location shift is not equal to 0

> #Sample type
> kruskal.test(Fyndata$Mn_mgkg~Fyndata$Name)

         Kruskal-Wallis rank sum test

data:  Fyndata$Mn_mgkg by Fyndata$Name
Kruskal-Wallis chi-squared = 85.2, df = 11, p-value = 1.4e-13

> pairwise.wilcox.test(Fyndata$Mn_mgkg,Fyndata$Name)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndata$Mn_mgkg and Fyndata$Name

             CH    Cl    CS    CT    FB    FE    FH    FI    Frass Rodent feces Rodent nest
Cl           0.364 -     -     -     -     -     -     -     -     -            -
CS           1.000 0.266 -     -     -     -     -     -     -     -            -
CT           0.017 0.017 0.017 -     -     -     -     -     -     -            -
FB           1.000 0.164 1.000 1.000 -     -     -     -     -     -            -
FE           5e-05 0.211 3e-05 2e-06 8e-07 -     -     -     -     -            -
FH           1.000 0.413 1.000 0.017 1.000 5e-05 -     -     -     -            -
FI           0.088 1.000 0.017 0.009 0.097 0.016 0.186 -     -     -            -
Frass        0.099 1.000 0.005 0.004 0.017 7e-06 0.352 1.000 -     -            -
Rodent feces 0.412 0.412 0.412 0.412 0.095 0.141 0.412 0.412 0.047 -            -
Rodent nest  1.000 1.000 1.000 1.000 1.000 0.266 1.000 1.000 0.822 1.000        -
Stickpile    1.000 1.000 1.000 0.022 0.644 0.002 1.000 1.000 1.000 0.097        1.000

P value adjustment method: holm
> kruskal.test(SKdata$Mn_mgkg~SKdata$Name)

         Kruskal-Wallis rank sum test

data:  SKdata$Mn_mgkg by SKdata$Name
Kruskal-Wallis chi-squared = 111, df = 11, p-value <2e-16

> pairwise.wilcox.test(SKdata$Mn_mgkg,SKdata$Name)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdata$Mn_mgkg and SKdata$Name

             Frass Rodent feces Rodent nest Sp10  Sp15  Sp16  Sp25  Sp40  Sp5   Sp8   Sp99
Rodent feces 0.133 -            -           -     -     -     -     -     -     -     -
Rodent nest  1.000 0.478        -           -     -     -     -     -     -     -     -
Sp10         4e-09 0.002        0.008       -     -     -     -     -     -     -     -
Sp15         3e-05 0.133        0.133       0.136 -     -     -     -     -     -     -
Sp16         1.000 0.291        1.000       0.003 0.023 -     -     -     -     -     -
Sp25         2e-08 0.020        0.020       1e-06 1.000 2e-04 -     -     -     -     -
Sp40         2e-04 0.133        0.133       1.000 1.000 0.023 0.186 -     -     -     -
Sp5          3e-05 0.133        0.133       1.000 1.000 0.008 0.247 1.000 -     -     -
Sp8          0.053 1.000        0.727       6e-06 0.008 0.478 2e-04 0.008 0.008 -     -
Sp99         3e-05 0.133        0.133       2e-05 1.000 0.008 1.000 0.212 0.161 0.008 -
Stickpile    0.212 0.011        1.000       0.008 0.002 1.000 5e-08 0.085 0.009 0.001 2e-05

P value adjustment method: holm

>

> #Mn
> kruskal.test(Mn_mgkg~Targeted,data=SKdataplants)
```

```
         Kruskal-Wallis rank sum test

data:  Mn_mgkg by Targeted
Kruskal-Wallis chi-squared = 16.275, df = 2, p-value = 0.0002924

> pairwise.wilcox.test(SKdataplants$Mn_mgkg,SKdataplants$Targeted)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$Mn_mgkg and SKdataplants$Targeted

             Maybe   Not Targeted
Not Targeted 0.00069 -
Targeted     0.00691 0.01495

P value adjustment method: holm
>
> kruskal.test(Mn_mgkg~Targeted,data=Fyndataplants)

         Kruskal-Wallis rank sum test

data:  Mn_mgkg by Targeted
Kruskal-Wallis chi-squared = 55.838, df = 2, p-value = 7.496e-13

> pairwise.wilcox.test(Fyndataplants$Mn_mgkg,Fyndataplants$Targeted)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$Mn_mgkg and Fyndataplants$Targeted

             Maybe   Not Targeted
Not Targeted 5.4e-10 -
Targeted     0.00056 2.2e-13

P value adjustment method: holm
>
#Mn
> kruskal.test(Mn_mgkg~Zone,data=SKdataplants)

         Kruskal-Wallis rank sum test

data:  Mn_mgkg by Zone
Kruskal-Wallis chi-squared = 5.0563, df = 2, p-value = 0.07981

> pairwise.wilcox.test(SKdataplants$Mn_mgkg,SKdataplants$Zone)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$Mn_mgkg and SKdataplants$Zone

      Centre Off
Off   0.391  -
Slope 0.086  0.328

P value adjustment method: holm
>
> kruskal.test(Mn_mgkg~Zone,data=Fyndataplants)

         Kruskal-Wallis rank sum test

data:  Mn_mgkg by Zone
Kruskal-Wallis chi-squared = 5.6885, df = 2, p-value = 0.05818

> pairwise.wilcox.test(Fyndataplants$Mn_mgkg,Fyndataplants$Zone)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$Mn_mgkg and Fyndataplants$Zone

      Centre Off
Off   0.053  -
Slope 0.452  0.452

P value adjustment method: holm
```

Boron



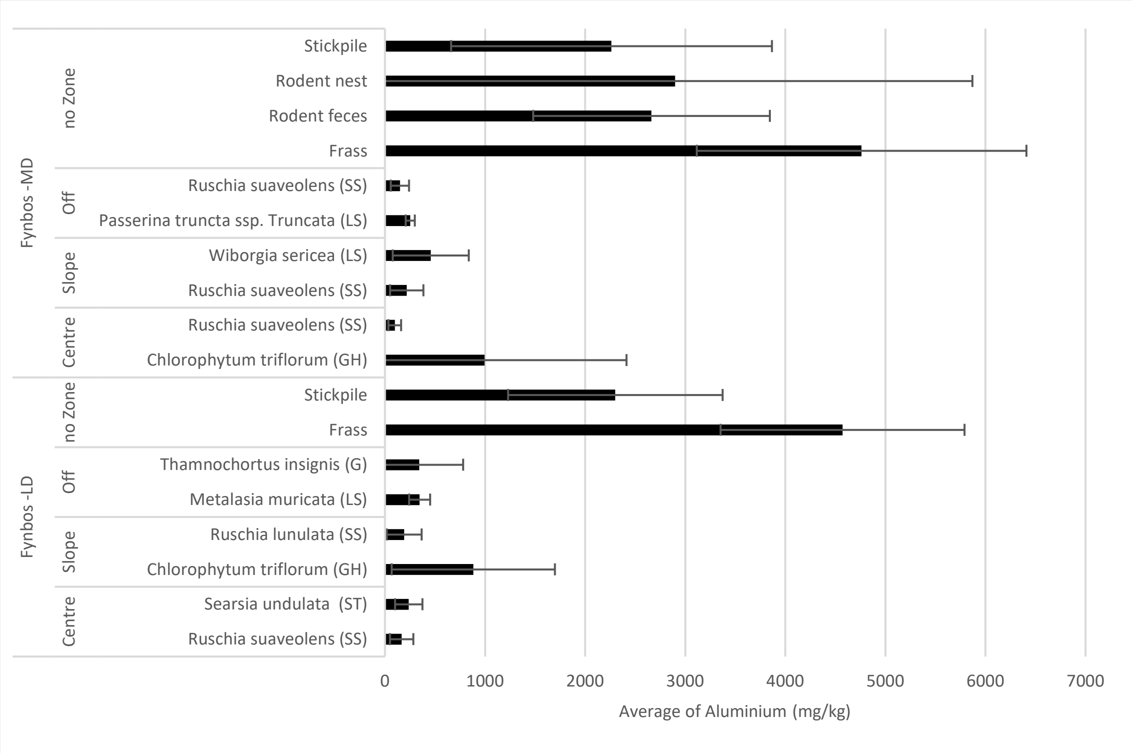


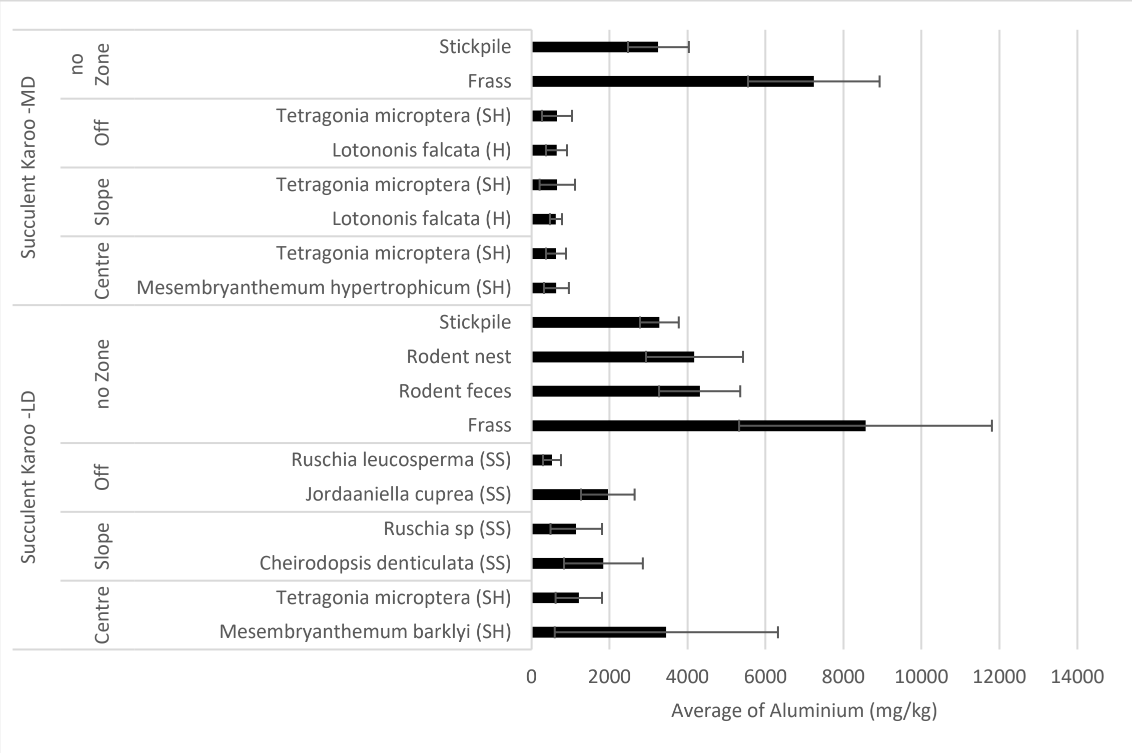

```
> #B
> #Biome
> wilcox.test(TSA$B_mgkg~TSA$Biome)

        Wilcoxon rank sum test with continuity correction

data:  TSA$B_mgkg by TSA$Biome
W = 8960, p-value = 0.024
alternative hypothesis: true location shift is not equal to 0

> #Subsite
> wilcox.test(Fyndata$B_mgkg~Fyndata$Site)
```

```
        Wilcoxon rank sum test with continuity correction

data:  Fyndata$B_mgkg by Fyndata$Site
W = 2504, p-value = 0.38
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$B_mgkg~SKdata$Site)

        Wilcoxon rank sum test with continuity correction

data:  SKdata$B_mgkg by SKdata$Site
W = 1771, p-value = 0.0021
alternative hypothesis: true location shift is not equal to 0

> #Year
> wilcox.test(Fyndata$B_mgkg~Fyndata$Year)

        Wilcoxon rank sum test with continuity correction

data:  Fyndata$B_mgkg by Fyndata$Year
W = 2680, p-value = 0.83
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$B_mgkg~SKdata$Year)

        Wilcoxon rank sum test with continuity correction

data:  SKdata$B_mgkg by SKdata$Year
W = 1764, p-value = 0.0018
alternative hypothesis: true location shift is not equal to 0

> #Sample type
> kruskal.test(Fyndata$B_mgkg~Fyndata$Name)

        Kruskal-Wallis rank sum test

data:  Fyndata$B_mgkg by Fyndata$Name
Kruskal-Wallis chi-squared = 96.1, df = 11, p-value = 1e-15

> pairwise.wilcox.test(Fyndata$B_mgkg,Fyndata$Name)

        Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndata$B_mgkg and Fyndata$Name

             CH    Cl    CS    CT    FB    FE    FH    FI    Frass Rodent feces Rodent nest
Cl           0.037 -     -     -     -     -     -     -     -     -            -
CS           0.525 0.008 -     -     -     -     -     -     -     -            -
CT           0.037 0.008 0.008 -     -     -     -     -     -     -            -
FB           0.028 1.000 2e-04 3e-04 -     -     -     -     -     -            -
FE           0.632 0.003 0.002 1e-03 0.022 -     -     -     -     -            -
FH           0.037 0.027 0.008 0.065 0.002 0.001 -     -     -     -            -
FI           1.000 0.039 0.027 0.008 0.575 1.000 0.008 -     -     -            -
Frass        0.004 1.000 3e-05 1e-04 1.000 5e-06 0.032 0.017 -     -            -
Rodent feces 0.632 0.352 1.000 0.352 0.070 0.159 0.352 0.352 0.037 -            -
Rodent nest  0.756 0.756 0.756 1.000 0.352 0.486 1.000 0.756 0.632 1.000        -
Stickpile    0.094 1.000 0.014 0.009 0.428 0.003 1.000 0.312 1.000 0.123        1.000

P value adjustment method: holm
> kruskal.test(SKdata$B_mgkg~SKdata$Name)

        Kruskal-Wallis rank sum test

data:  SKdata$B_mgkg by SKdata$Name
Kruskal-Wallis chi-squared = 57.6, df = 11, p-value = 2.6e-08

> pairwise.wilcox.test(SKdata$B_mgkg,SKdata$Name)

        Pairwise comparisons using Wilcoxon rank sum test

data:  SKdata$B_mgkg and SKdata$Name

             Frass Rodent feces Rodent nest Sp10  Sp15  Sp16  Sp25  Sp40  Sp5   Sp8   Sp99
Rodent feces 1.000 -            -           -     -     -     -     -     -     -     -
Rodent nest  1.000 1.000        -           -     -     -     -     -     -     -     -
Sp10         1.000 0.702        1.000       -     -     -     -     -     -     -     -
Sp15         1.000 1.000        1.000       1.000 -     -     -     -     -     -     -
Sp16         1.000 1.000        1.000       0.564 1.000 -     -     -     -     -     -
Sp25         0.202 0.029        0.105       0.936 5e-04 3e-04 -     -     -     -     -
Sp40         1.000 1.000        1.000       1.000 1.000 1.000 0.057 -     -     -     -
Sp5          1.000 1.000        1.000       0.822 1.000 1.000 5e-04 1.000 -     -     -
Sp8          0.641 0.202        0.202       1.000 0.018 0.009 1.000 0.329 0.009 -     -
Sp99         0.447 0.385        0.385       1.000 0.071 0.052 1.000 0.441 0.052 1.000 -
Stickpile    0.969 1.000        1.000       0.101 1.000 1.000 4e-04 1.000 1.000 0.011 0.008

P value adjustment method: holm

>

> #B
> kruskal.test(B_mgkg~Targeted,data=SKdataplants)
```

```
         Kruskal-Wallis rank sum test

data:  B_mgkg by Targeted
Kruskal-Wallis chi-squared = 0.17019, df = 2, p-value = 0.9184

> pairwise.wilcox.test(SKdataplants$B_mgkg,SKdataplants$Targeted)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$B_mgkg and SKdataplants$Targeted

             Maybe Not Targeted
Not Targeted 1     -
Targeted     1     1

P value adjustment method: holm
>
> kruskal.test(B_mgkg~Targeted,data=Fyndataplants)

         Kruskal-Wallis rank sum test

data:  B_mgkg by Targeted
Kruskal-Wallis chi-squared = 16.628, df = 2, p-value = 0.0002451

> pairwise.wilcox.test(Fyndataplants$B_mgkg,Fyndataplants$Targeted)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$B_mgkg and Fyndataplants$Targeted

             Maybe   Not Targeted
Not Targeted 5.3e-06 -
Targeted     0.074   0.973

P value adjustment method: holm
>
> #B
> kruskal.test(B_mgkg~Zone,data=SKdataplants)

         Kruskal-Wallis rank sum test

data:  B_mgkg by Zone
Kruskal-Wallis chi-squared = 10.119, df = 2, p-value = 0.00635

> pairwise.wilcox.test(SKdataplants$B_mgkg,SKdataplants$Zone)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$B_mgkg and SKdataplants$Zone

      Centre Off
Off   0.016  -
Slope 0.789  0.016

P value adjustment method: holm
>
> kruskal.test(B_mgkg~Zone,data=Fyndataplants)

         Kruskal-Wallis rank sum test

data:  B_mgkg by Zone
Kruskal-Wallis chi-squared = 17.571, df = 2, p-value = 0.000153

> pairwise.wilcox.test(Fyndataplants$B_mgkg,Fyndataplants$Zone)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$B_mgkg and Fyndataplants$Zone

      Centre  Off
Off   0.867   -
Slope 1.5e-05 0.023

P value adjustment method: holm
```

Aluminium






```
> #Al
> #Biome
> wilcox.test(TSA$Al_mgkg~TSA$Biome)

         Wilcoxon rank sum test with continuity correction

data:  TSA$Al_mgkg by TSA$Biome
W = 5996, p-value = 1.7e-10
alternative hypothesis: true location shift is not equal to 0

> #Subsite
> wilcox.test(Fyndata$Al_mgkg~Fyndata$Site)

         Wilcoxon rank sum test with continuity correction
```

```
data:  Fyndata$Al_mgkg by Fyndata$Site
W = 2764, p-value = 0.92
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$Al_mgkg~SKdata$Site)

         Wilcoxon rank sum test with continuity correction

data:  SKdata$Al_mgkg by SKdata$Site
W = 3612, p-value = 1.1e-05
alternative hypothesis: true location shift is not equal to 0

> #Year
> wilcox.test(Fyndata$Al_mgkg~Fyndata$Year)

         Wilcoxon rank sum test with continuity correction

data:  Fyndata$Al_mgkg by Fyndata$Year
W = 2528, p-value = 0.43
alternative hypothesis: true location shift is not equal to 0

> wilcox.test(SKdata$Al_mgkg~SKdata$Year)

         Wilcoxon rank sum test with continuity correction

data:  SKdata$Al_mgkg by SKdata$Year
W = 2188, p-value = 0.16
alternative hypothesis: true location shift is not equal to 0

> #Sample type
> kruskal.test(Fyndata$Al_mgkg~Fyndata$Name)

         Kruskal-Wallis rank sum test

data:  Fyndata$Al_mgkg by Fyndata$Name
Kruskal-Wallis chi-squared = 114, df = 11, p-value <2e-16

> pairwise.wilcox.test(Fyndata$Al_mgkg,Fyndata$Name)

         Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndata$Al_mgkg and Fyndata$Name

             CH    Cl    CS    CT    FB    FE    FH    FI    Frass Rodent feces Rodent nest
Cl           1.000 -     -     -     -     -     -     -     -     -            -
CS           1.000 1.000 -     -     -     -     -     -     -     -            -
CT           1.000 1.000 1.000 -     -     -     -     -     -     -            -
FB           0.655 0.533 1.000 0.655 -     -     -     -     -     -            -
FE           1.000 1.000 0.044 1.000 0.001 -     -     -     -     -            -
FH           1.000 1.000 1.000 1.000 1.000 0.103 -     -     -     -            -
FI           1.000 1.000 1.000 1.000 0.533 0.526 1.000 -     -     -            -
Frass        0.002 0.002 0.002 0.002 6e-05 7e-08 0.002 0.002 -     -            -
Rodent feces 0.661 0.533 0.533 0.661 1.000 0.234 0.533 0.660 1.000 -            -
Rodent nest  1.000 1.000 1.000 1.000 1.000 0.714 1.000 1.000 1.000 1.000        -
Stickpile    0.002 0.002 0.002 0.003 0.031 5e-09 0.002 0.002 2e-04 1.000        1.000

P value adjustment method: holm
> kruskal.test(SKdata$Al_mgkg~SKdata$Name)

         Kruskal-Wallis rank sum test

data:  SKdata$Al_mgkg by SKdata$Name
Kruskal-Wallis chi-squared = 117, df = 11, p-value <2e-16

> pairwise.wilcox.test(SKdata$Al_mgkg,SKdata$Name)

         Pairwise comparisons using Wilcoxon rank sum test

data:  SKdata$Al_mgkg and SKdata$Name

             Frass Rodent feces Rodent nest Sp10  Sp15  Sp16  Sp25  Sp40  Sp5   Sp8   Sp99
Rodent feces 0.149 -            -           -     -     -     -     -     -     -     -
Rodent nest  0.149 1.000        -           -     -     -     -     -     -     -     -
Sp10         1e-07 0.059        0.059       -     -     -     -     -     -     -     -
Sp15         0.003 0.137        0.137       1.000 -     -     -     -     -     -     -
Sp16         0.003 0.210        0.210       0.009 0.008 -     -     -     -     -     -
Sp25         3e-05 0.023        0.023       1.000 1.000 5e-04 -     -     -     -     -
Sp40         0.003 0.137        0.137       1.000 0.758 1.000 0.516 -     -     -     -
Sp5          0.046 1.000        1.000       0.011 0.069 1.000 0.010 0.516 -     -     -
Sp8          0.003 0.388        0.388       0.030 0.008 1.000 1e-03 1.000 1.000 -     -
Sp99         0.003 0.137        0.137       1.000 1.000 0.008 1.000 1.000 0.069 0.046 -
Stickpile    3e-06 1.000        1.000       9e-08 0.002 0.036 2e-05 0.002 1.000 0.062 0.002

P value adjustment method: holm

>
```

```
> #Al
> kruskal.test(Al_mgkg~Targeted,data=SKdataplants)

	Kruskal-Wallis rank sum test

data:  Al_mgkg by Targeted
Kruskal-Wallis chi-squared = 12.526, df = 2, p-value = 0.001905

> pairwise.wilcox.test(SKdataplants$Al_mgkg,SKdataplants$Targeted)

	Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$Al_mgkg and SKdataplants$Targeted

             Maybe  Not Targeted
Not Targeted 0.6108 -
Targeted     0.0036 0.0677

P value adjustment method: holm
>
> kruskal.test(Al_mgkg~Targeted,data=Fyndataplants)

	Kruskal-Wallis rank sum test

data:  Al_mgkg by Targeted
Kruskal-Wallis chi-squared = 21.356, df = 2, p-value = 2.304e-05

> pairwise.wilcox.test(Fyndataplants$Al_mgkg,Fyndataplants$Targeted)

	Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$Al_mgkg and Fyndataplants$Targeted

             Maybe   Not Targeted
Not Targeted 0.00018 -
Targeted     0.61902 0.00016

P value adjustment method: holm

>


> #Al
> kruskal.test(Al_mgkg~Zone,data=SKdataplants)

	Kruskal-Wallis rank sum test

data:  Al_mgkg by Zone
Kruskal-Wallis chi-squared = 1.6427, df = 2, p-value = 0.4398

> pairwise.wilcox.test(SKdataplants$Al_mgkg,SKdataplants$Zone)

	Pairwise comparisons using Wilcoxon rank sum test

data:  SKdataplants$Al_mgkg and SKdataplants$Zone

      Centre Off
Off   0.63   -
Slope 0.82   0.82

P value adjustment method: holm
>
> kruskal.test(Al_mgkg~Zone,data=Fyndataplants)

	Kruskal-Wallis rank sum test

data:  Al_mgkg by Zone
Kruskal-Wallis chi-squared = 4.535, df = 2, p-value = 0.1036

> pairwise.wilcox.test(Fyndataplants$Al_mgkg,Fyndataplants$Zone)

	Pairwise comparisons using Wilcoxon rank sum test

data:  Fyndataplants$Al_mgkg and Fyndataplants$Zone

      Centre Off
Off   0.29   -
Slope 0.17   0.32

P value adjustment method: holm
```

Table E: Average tissue salt concentrations in the different plant species

Tissue salts	Na (%)	P(%)	K (%)	Ca (%)	Mg (%)	Cu (mg/kg)	Zn (mg/kg)	Mn (mg/kg)	B (mg/kg)	Al (mg/kg)	Fe (mg/kg)
Fynbos species											
Chlorophytum triflorum (GH)	0,34	0,04	0,52	1,96	0,26	7,28	21,76	396,40	26,52	938,38	1194,12
Metalasia muricata (LS)	0,42	0,05	0,83	0,41	0,15	5,68	16,59	271,09	41,85	346,25	355,38
Passerina truncta sp, Truncata (LS)	0,25	0,05	0,72	0,51	0,26	4,74	20,79	916,49	32,33	252,50	228,69
Ruschia lunulata (SS)	1,01	0,04	1,13	1,60	0,44	4,29	16,94	1175,85	22,60	191,88	171,23
Ruschia suaveolens (SS)	1,73	0,05	1,69	1,99	1,31	4,38	17,38	2141,21	32,75	157,72	151,70
Searsia undulata (ST)	0,37	0,06	1,10	1,99	0,24	10,88	13,66	354,01	37,07	237,50	258,84
Thamnochortus insignis (G)	0,09	0,03	0,55	0,18	0,07	2,18	11,04	123,92	7,73	343,75	295,05
Wiborgia sericea (LS)	0,09	0,07	0,47	0,28	0,09	3,78	15,95	379,61	14,44	457,50	560,04
Succulent Karoo species											
Cheirodopsis denticulata (SS)	4,39	0,10	1,08	4,36	3,80	3,29	8,80	604,05	40,29	1838,93	1406,93
Jordaaniella cuprea (SS)	3,01	0,08	0,98	2,57	0,75	4,09	11,20	342,81	22,64	1953,75	1431,79
Lotononis falcata (H)	1,06	0,15	1,23	1,75	0,58	4,48	15,14	89,48	49,12	630,67	863,67
Mesembryanthemum barklyi (SH)	18,30	0,10	1,89	0,98	0,36	3,74	11,72	126,64	22,97	3453,75	2687,58
Mesembryanthemum hypertrophicum (SH)	29,12	0,07	1,28	2,89	0,47	2,50	9,80	80,54	42,79	632,50	619,01
Ruschia leucosperma (SS)	2,89	0,11	1,21	0,72	0,64	4,49	11,47	99,66	25,01	525,00	411,01
Ruschia sp (SS)	6,04	0,07	1,34	3,75	0,50	2,85	8,81	140,10	26,92	1146,25	663,65
Tetragonia microptera (SH)	6,86	0,11	1,58	1,54	0,77	5,26	16,11	158,24	41,04	764,49	910,79

Table F: Tissue salts in FE and sp10 samples growing in each zone of the mound

Tissue salts		Na (%)	P (%)	K (%)	Ca (%)	Mg (%)	Cu (mg/kg)	Zn (mg/kg)	Mn (mg/kg)	B (mg/kg)	Al (mg/kg)	Fe (mg/kg)
Ruschia suaveolens (SS)	F-MD											
	Centre	1,61	0,05	1,82	1,99	1,54	4,42	16,00	2303,35	35,24	97,63	101,56
	Slope	1,55	0,04	1,68	2,02	1,25	4,48	15,59	2349,96	31,62	217,50	208,13
	Off	1,71	0,05	1,47	1,61	1,03	3,09	17,71	1385,16	33,44	149,00	124,96
	F-LD											
	Centre	2,06	0,06	1,81	2,34	1,43	5,55	20,21	2526,38	30,71	166,75	172,15
Tetragonia microptera (SH)	S-MD											
	Centre	8,03	0,10	1,44	1,30	0,72	4,65	13,57	139,29	72,74	627,17	682,89
	Slope	5,86	0,13	1,62	1,29	0,84	5,88	18,83	184,47	35,43	661,25	1294,49
	Off	6,27	0,13	1,53	2,43	0,90	5,82	19,79	152,37	24,30	653,75	801,96
	S-LD											
	Centre	7,61	0,08	1,77	0,95	0,59	4,41	10,52	153,23	33,86	1210,00	810,17

Appendix 10: Boxplots and table of plant tissues ions groups by mound location and targeted plants

Table G: Table of plant tissues ions groups by mound location and targeted plants

Biome	Zone	Targeted	Species	Sodium (%)	Calcium (%)	Magnesium (%)	Phosphorus (%)	Potassium (%)	Aluminium (mg/kg)	Boron (mg/kg)	Copper (mg/kg)	Iron (mg/kg)	Manganese (mg/kg)	Zinc (mg/kg)
Fynbos	Centre	Targeted	CH	0.37 ±0.11	1.99 ± 0.25	0.24 ±0.02	0.06 ±0.01	1.10 ±0.14	237.50 ±136.88	37.07 ±5.03	10.88 ±2.82	258.84 ±181.06	354.01 ±166.66	13.66 ±3.17
			FB	0.40 ±0.58	1.90 ±0.45	0.35 ±0.52	0.05 ±0.04	0.64 ±0.46	994.25 ±1419.30	28.15 ±6.14	7.33 ±3.34	1411.23 ±2267.81	475.94 ±761.82	18.81 ±10.27
		Not Targeted	FE	1.83 ±0.71	2.17 ±0.62	1.49 ±0.53	0.06 ±0.02	1.81 ±0.38	132.19 ±97.55	32.98 ±4.13	4.98 ±3.62	136.86 ±96.19	2414.86 ±842.98	18.11 ±10.18
	Slope	Maybe Targeted	Cl	1.01 ±0.48	1.60 ±1.43	0.44 ±0.18	0.04 ±0.01	1.13 ±0.53	191.88 ±174.31	22.60 ±3.41	4.29 ±1.61	171.23 ±123.98	1175.85 ±522.25	16.94 ±3.22
			FB	0.28 ±0.37	2.03 ±0.55	0.17 ±0.12	0.03 ±0.02	0.40 ±0.34	882.50 ±815.56	24.89 ±4.99	7.23 ±2.16	977.01 ±1153.94	316.86 ±304.07	24.72 ±15.80
			FH	0.09 ±0.05	0.28 ±0.11	0.09 ±0.03	0.07 ±0.02	0.47 ±0.07	457.50 ±379.88	14.44 ±3.42	3.78 ±0.79	560.04 ±521.14	379.61 ±190.50	15.95 ±6.14
		Not Targeted	FE	1.55 ±0.65	2.02 ±0.47	1.25 ±0.54	0.04 ±0.01	1.68 ±0.68	217.50 ±166.59	31.62 ±6.67	4.48 ±1.10	208.13 ±149.58	2349.96 ±1338.09	15.59 ±3.03
	Off	Targeted	CS	0.42 ±0.12	0.41 ±0.05	0.15 ±0.02	0.05 ±0.01	0.83 ±0.08	346.25 ±105.01	41.85 ±2.34	5.68 ±1.19	355.38 ±98.72	271.09 ±59.25	16.59 ±5.73
			CT	0.09 ±0.04	0.18 ±0.10	0.07 ±0.03	0.03 ±0.02	0.55 ±0.15	343.75 ±437.29	7.73 ±3.35	2.18 ±0.88	295.05 ±361.60	123.92 ±50.25	11.04 ±8.02
		Maybe Targeted	FI	0.25 ±0.49	0.51 ±0.39	0.26 ±0.38	0.05 ±0.01	0.72 ±0.26	252.50 ±45.59	32.33 ±5.24	4.74 ±0.83	228.69 ±42.00	916.49 ±470.06	20.79 ±4.26
		Not Targeted	FE	1.71 ±0.43	1.61 ±0.22	1.03 ±0.38	0.05 ±0.01	1.47 ±0.42	149.00 ±91.30	33.44 ±2.92	3.09 ±1.42	124.96 ±66.24	1385.16 ±486.00	17.71 ±4.56
S. Karoo	Centre	Targeted	Sp10	7.84 ±2.38	1.14 ±0.39	0.66 ±0.14	0.09 ±0.03	1.59 ±0.52	896.17 ±522.08	54.79 ±29.06	4.54 ±0.64	741.63 ±364.20	145.73 ±25.52	12.16 ±3.14
		Not Targeted	Sp5	18.30 ±3.86	0.98 ±0.54	0.360.10	0.10 ±0.02	1.89 ±0.57	3453.75 ±2862.13	22.97 ±5.21	3.74 ±2.65	2687.58 ±2213.85	126.64 ±32.48	11.72 ±5.84
			Sp99	29.12 ±9.67	2.89 ±1.15	0.47 ±0.07	0.07 ±0.02	1.28 ±0.51	632.50 ±320.21	42.79 ±12.53	2.50 ±0.70	619.01 ±291.02	80.54 ±19.62	9.80 ±2.09
	Slope	Targeted	Sp10	5.86 ±3.27	1.29 ±0.50	0.84 ±0.11	0.13 ±0.03	1.62 ±0.56	661.25 ±456.52	35.43 ±17.43	5.88 ±1.65	1294.49 ±1714.67	184.47 ±69.17	18.83 ±4.45
			Sp25	1.06 ±0.46	1.73 ±0.91	0.54 ±0.11	0.17 ±0.11	1.34 ±0.29	621.08 ±155.44	55.95 ±27.20	4.39 ±0.58	878.36 ±233.51	96.48 ±20.66	16.35 ±5.41
		Maybe Targeted	Sp40	6.04 ±2.90	3.75 ±1.00	0.50 ±0.15	0.07 ±0.02	1.34 ±0.56	1146.25 ±660.41	26.92 ±10.31	2.85 ±1.04	663.65 ±296.63	140.10 ±51.37	8.81 ±2.82
			Sp8	4.39 ±1.33	4.36 ±1.21	3.80 ±1.08	0.10 ±0.06	1.08 ±0.30	1838.93 ±1011.80	40.29 ±9.01	3.29 ±0.76	1406.93 ±750.22	604.05 ±187.51	8.80 ±2.14
	Off	Targeted	Sp10	6.27 ±3.02	2.43 ±3.47	0.90 ±0.20	0.13 ±0.02	1.53 ±0.48	653.75 ±385.37	24.30 ±2.76	5.82 ±1.39	801.96 ±518.45	152.37 ±39.02	19.79 ±4.85
			Sp15	2.89 ±0.64	0.72 ±0.16	0.64 ±0.24	0.11 ±0.02	1.210.24	525.00 ±226.02	25.01 ±3.74	4.49 ±0.44	411.01 ±147.84	99.66 ±47.66	11.47 ±1.12
			Sp16	3.01 ±0.61	2.57 ±0.63	0.75 ±0.20	0.08 ±0.02	0.98 ±0.14	1953.75 ±688.50	22.64 ±2.89	4.09 ±0.60	1431.79 ±348.39	342.81 ±165.38	11.20 ±3.10
			Sp25	1.07 ±0.37	1.78 ±0.80	0.64 ±0.30	0.13 ±0.03	1.11 ±0.23	641.63 ±271.84	41.32 ±14.97	4.59 ±0.67	846.87 ±352.79	81.49 ±22.15	13.75 ±3.39

Sodium

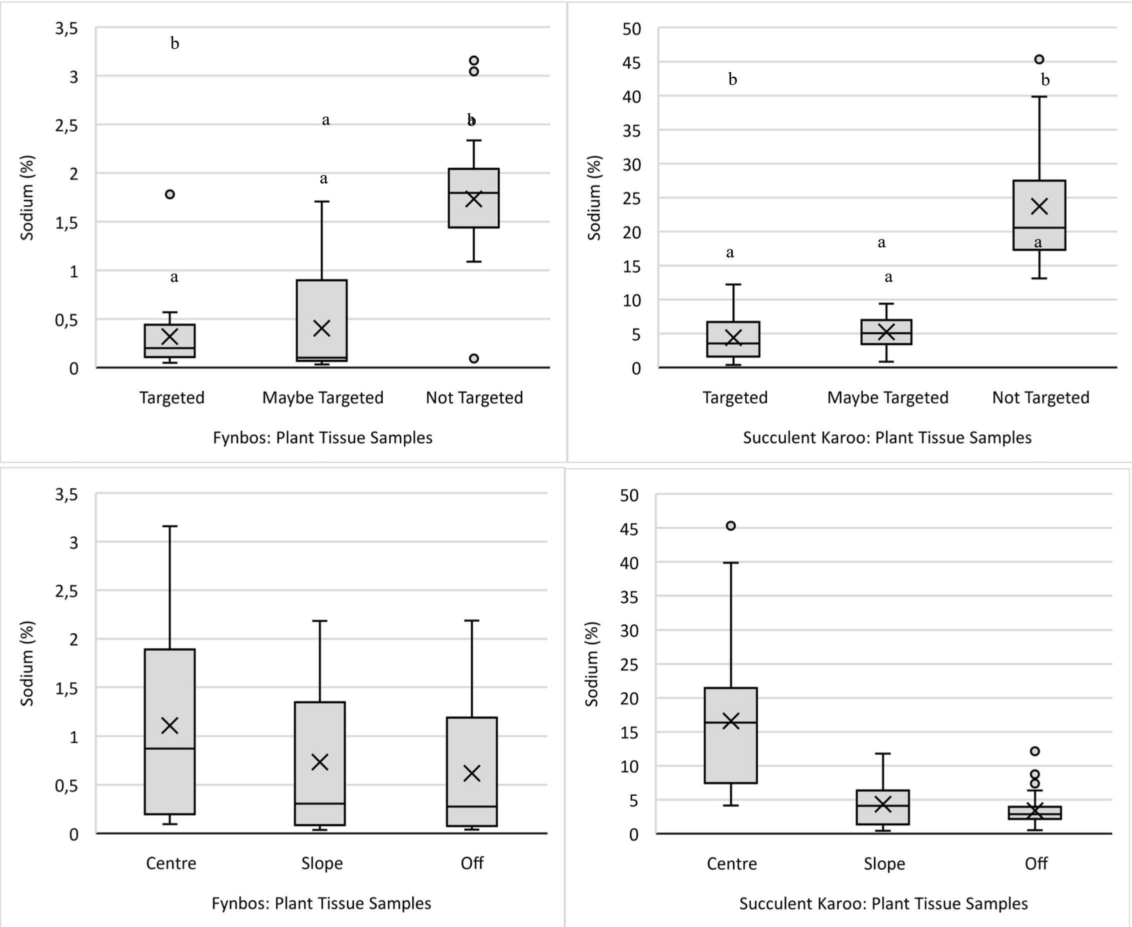
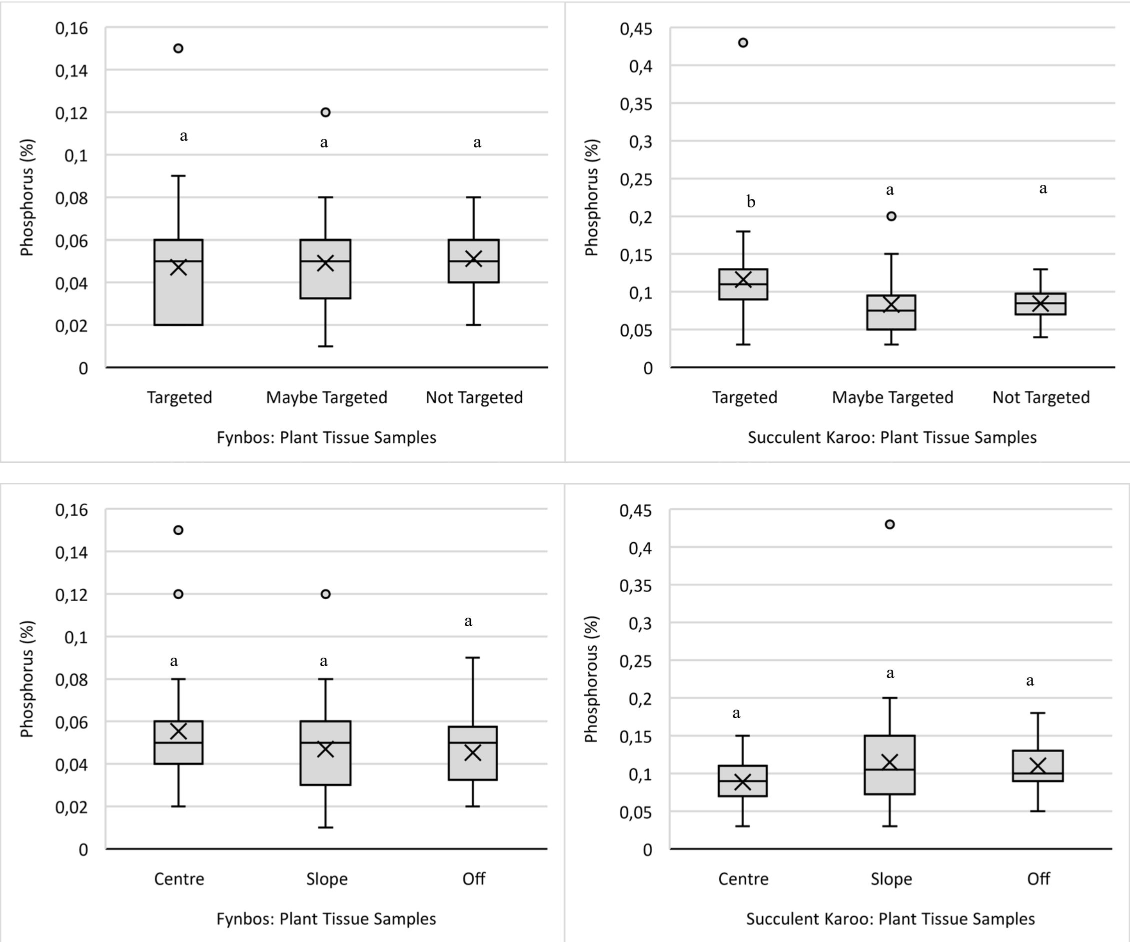


Figure KKK: Boxplots showing the comparison of sodium in dried plant material in 1) plants "Targeted" or "Not Targeted" by termites and rodents and 2) Where the plants can be found growing on the heuweltjie mounds. Groups with the same letter on each graph are not significantly different ($p < 0.05$) from each other, with different letters indication significant differences according to pairwise comparisons run in R using Wilcoxon rank sum tests.

Average of Sodium (%)				
Row Labels	**Centre**	**Slope**	**Off**	**Targeted Total**
Fynbos	**1,11**	**0,73**	**0,62**	**0,82**
Targeted	0,38		0,26	0,32
Maybe Targeted		0,46	0,25	0,41
Not Targeted	1,83	1,55	1,71	1,73
Succulent Karoo	**16,59**	**4,34**	**3,38**	**7,88**
Targeted	7,84	3,46	3,38	4,37
Maybe Targeted		5,22		5,22
Not Targeted	23,71			23,71

Phosphorous



Figure#

Figure LLL: Boxplots showing the comparison of phosphorus in dried plant material in 1) plants "Targeted" or "Not Targeted" by termites and rodents and 2) Where the plants can be found growing on the heuweltjie mounds. Groups with the same letter on each graph are not significantly different ($p < 0.05$) from each other, with different letters indication significant differences according to pairwise comparisons run in R using Wilcoxon rank sum tests.

Average of Phosphorus (%)				
Row Labels	**Centre**	**Slope**	**Off**	**Targeted Total**
Fynbos	**0,06**	**0,05**	**0,05**	**0,05**
Targeted	0,05		0,04	0,05
Maybe Targeted		0,05	0,05	0,05
Not Targeted	0,06	0,04	0,05	0,05
Succulent Karoo	**0,09**	**0,12**	**0,11**	**0,11**
Targeted	0,09	0,15	0,11	0,12
Maybe Targeted		0,08		0,08
Not Targeted	0,08			0,08

Potassium

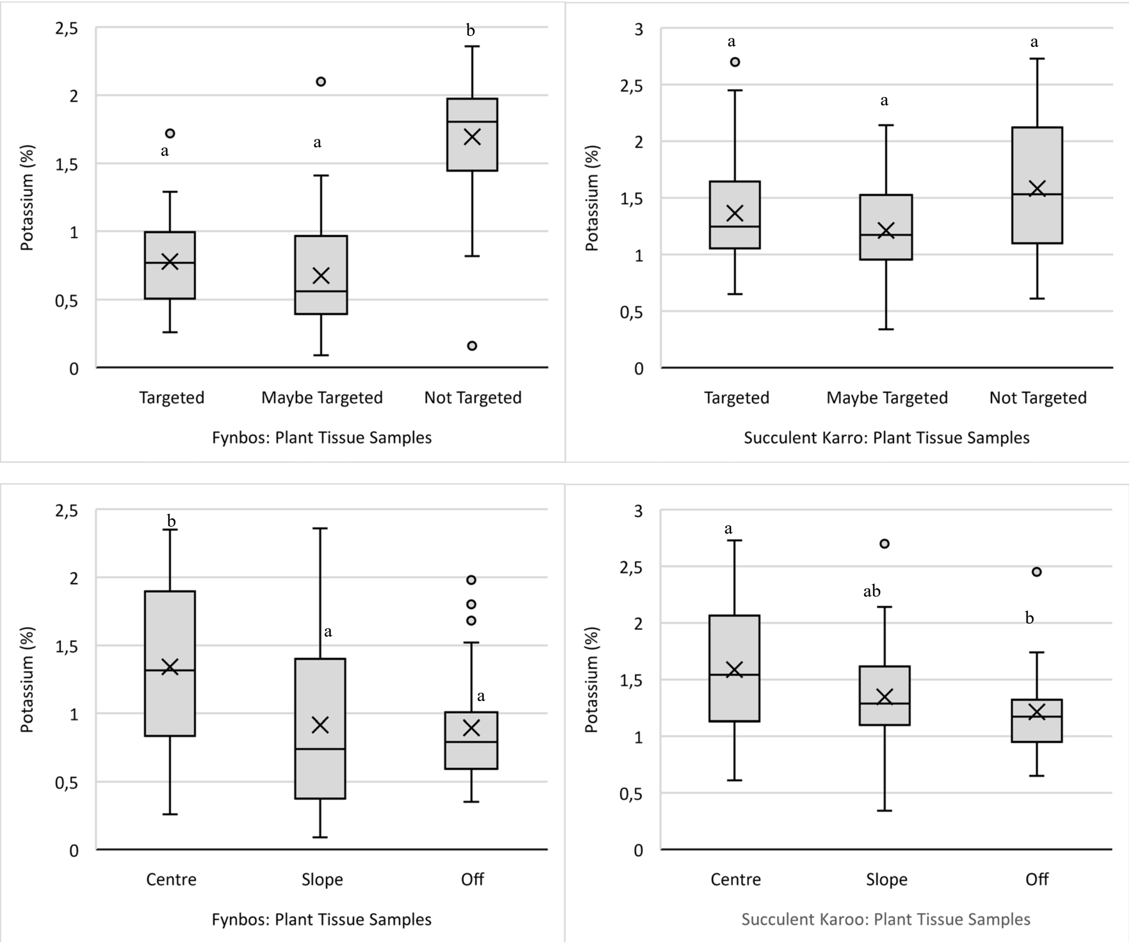


Figure MMM: Boxplots showing the comparison of potassium in dried plant material in 1) plants "Targeted" or "Not Targeted" by termites and rodents and 2) Where the plants can be found growing on the heuweltjie mounds. Groups with the same letter on each graph are not significantly different ($p < 0.05$) from each other, with different letters indication significant differences according to pairwise comparisons run in R using Wilcoxon rank sum tests.

Average of Potassium (%)				
Row Labels	**Centre**	**Slope**	**Off**	**Targeted Total**
Fynbos	**1,34**	**0,92**	**0,89**	**1,05**
Targeted	0,87		0,69	0,78
Maybe Targeted		0,66	0,72	0,68
Not Targeted	1,81	1,68	1,47	1,69
Succulent Karoo	**1,59**	**1,34**	**1,21**	**1,38**
Targeted	1,59	1,48	1,21	1,36
Maybe Targeted		1,21		1,21
Not Targeted	1,58			1,58

Calcium

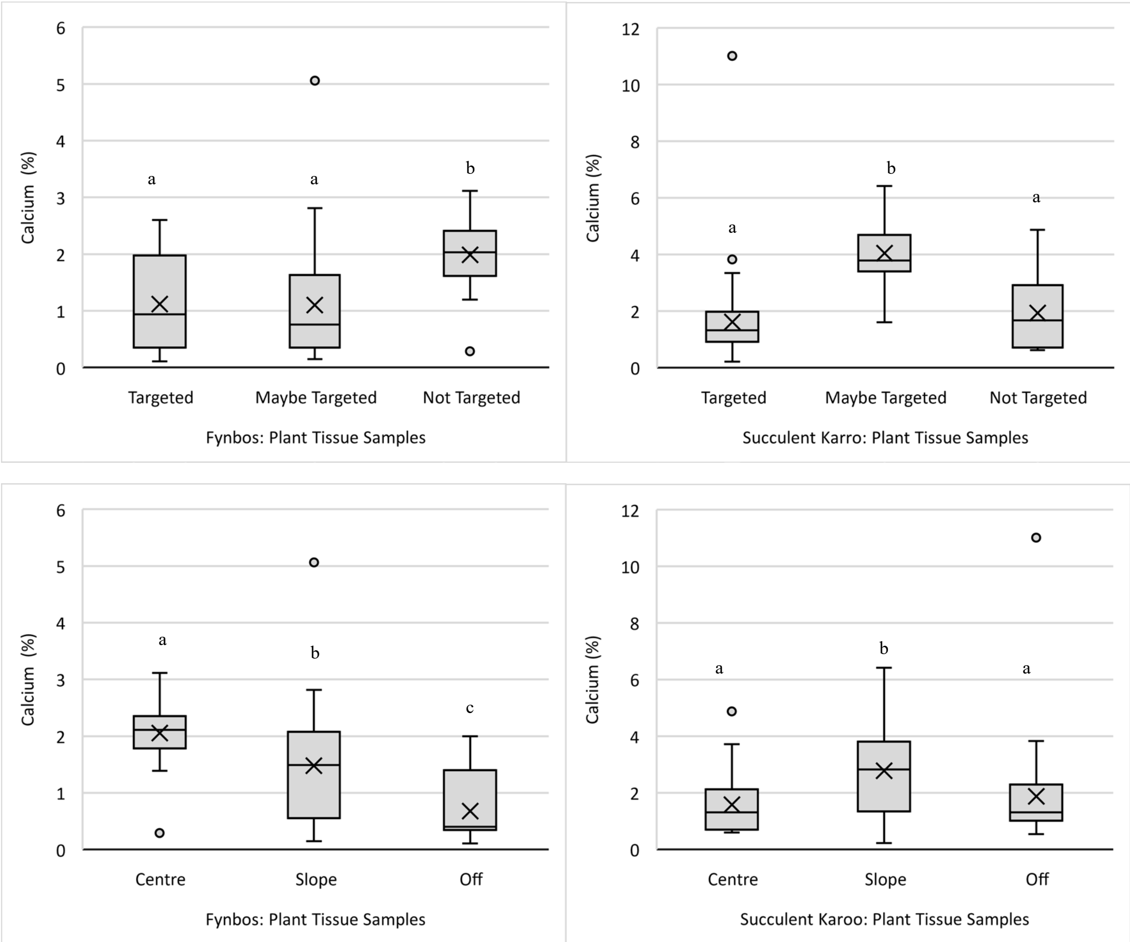


Figure NNN: Boxplots showing the comparison of calcium in dried plant material in 1) plants "Targeted" or "Not Targeted" by termites and rodents and 2) Where the plants can be found growing on the heuweltjie mounds. Groups with the same letter on each graph are not significantly different ($p < 0.05$) from each other, with different letters indication significant differences according to pairwise comparisons run in R using Wilcoxon rank sum tests.

Average of Calcium (%)				
Row Labels	**Centre**	**Slope**	**Off**	**Targeted Total**
Fynbos	**2,06**	**1,48**	**0,68**	**1,40**
Targeted	1,94		0,29	1,12
Maybe Targeted		1,30	0,51	1,11
Not Targeted	2,17	2,02	1,61	1,99
Succulent Karoo	**1,58**	**2,78**	**1,88**	**2,10**
Targeted	1,14	1,51	1,88	1,62
Maybe Targeted		4,05		4,05
Not Targeted	1,94			1,94

Magnesium

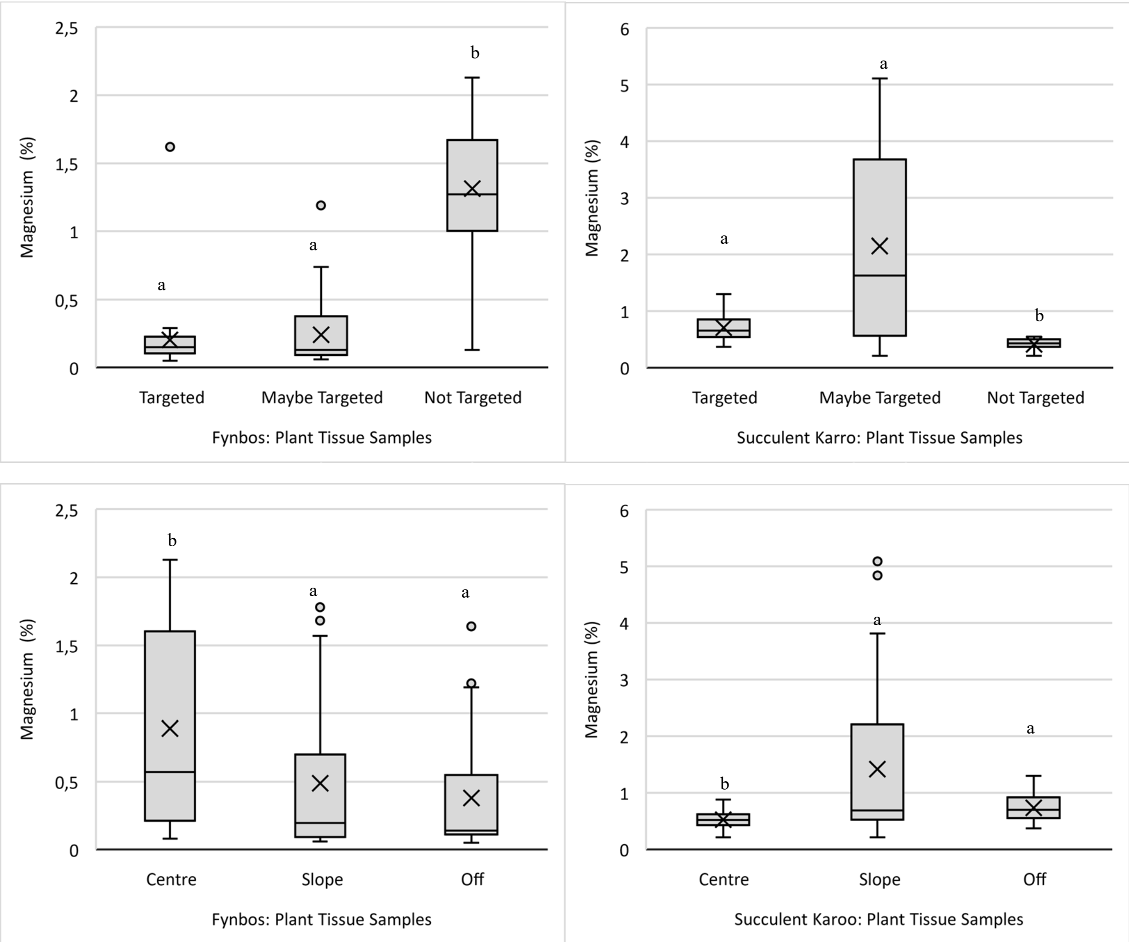


Figure OOO: Boxplots showing the comparison of magnesium in dried plant material in 1) plants "Targeted" or "Not Targeted" by termites and rodents and 2) Where the plants can be found growing on the heuweltjie mounds. Groups with the same letter on each graph are not significantly different ($p < 0.05$) from each other, with different letters indication significant differences according to pairwise comparisons run in R using Wilcoxon rank sum tests.

Average of Magnesium (%)				
Row Labels	**Centre**	**Slope**	**Off**	**Targeted Total**
Fynbos	**0,89**	**0,49**	**0,38**	**0,59**
Targeted	0,29		0,11	0,20
Maybe Targeted		0,23	0,26	0,24
Not Targeted	1,49	1,25	1,03	1,31
Succulent Karoo	**0,53**	**1,42**	**0,73**	**0,91**
Targeted	0,66	0,69	0,73	0,71
Maybe Targeted		2,15		2,15
Not Targeted	0,42			0,42

Copper

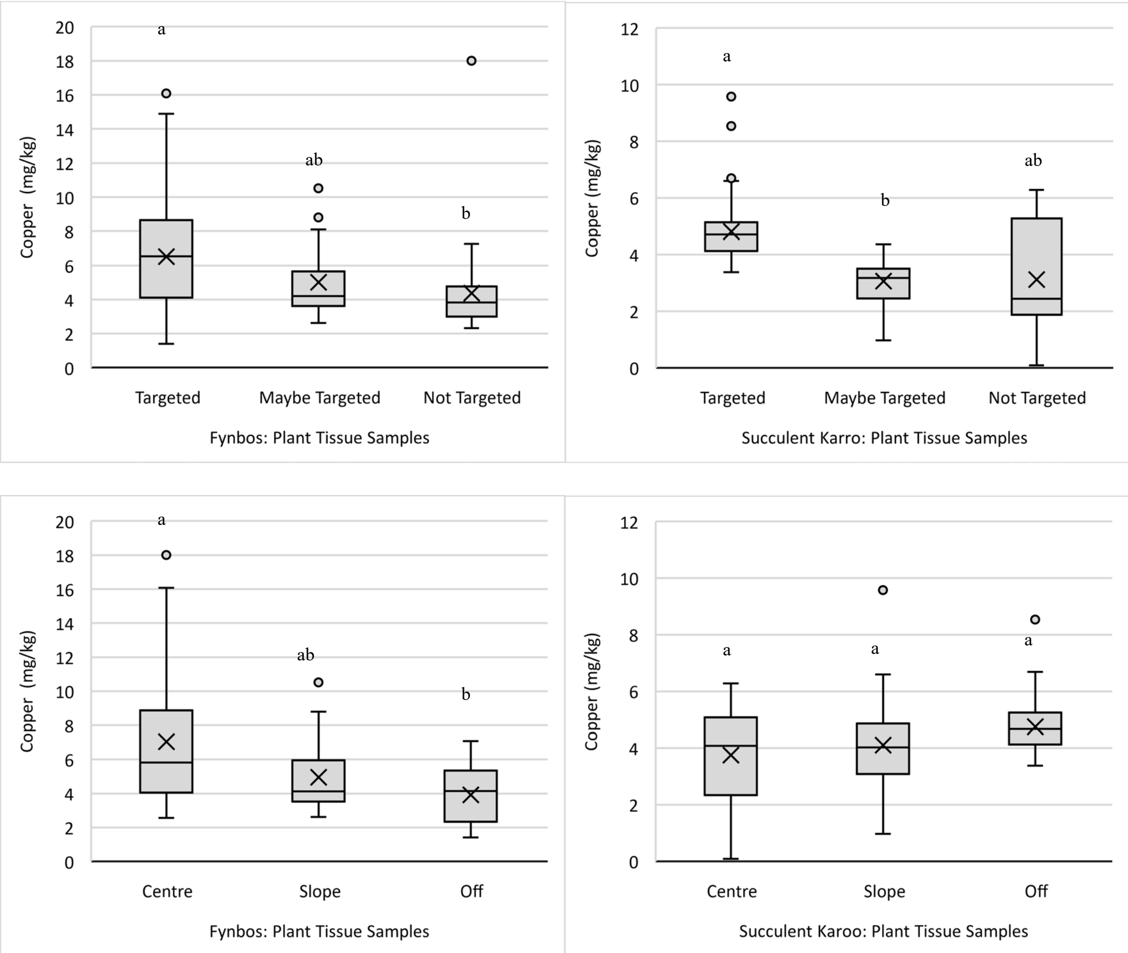


Figure PPP: Boxplots showing the comparison of copper in dried plant material in 1) plants "Targeted" or "Not Targeted" by termites and rodents and 2) Where the plants can be found growing on the heuweltjie mounds. Groups with the same letter on each graph are not significantly different ($p < 0.05$) from each other, with different letters indication significant differences according to pairwise comparisons run in R using Wilcoxon rank sum tests.

Average of Copper (mg/kg)				
Row Labels	**Centre**	**Slope**	**Off**	**Targeted Total**
Fynbos	**7,04**	**4,94**	**3,92**	**5,30**
Targeted	9,10		3,93	6,52
Maybe Targeted		5,10	4,74	5,01
Not Targeted	4,98	4,48	3,09	4,38
Succulent Karoo	**3,76**	**4,10**	**4,75**	**4,21**
Targeted	4,54	5,13	4,75	4,81
Maybe Targeted		3,07		3,07
Not Targeted	3,12			3,12

Zinc

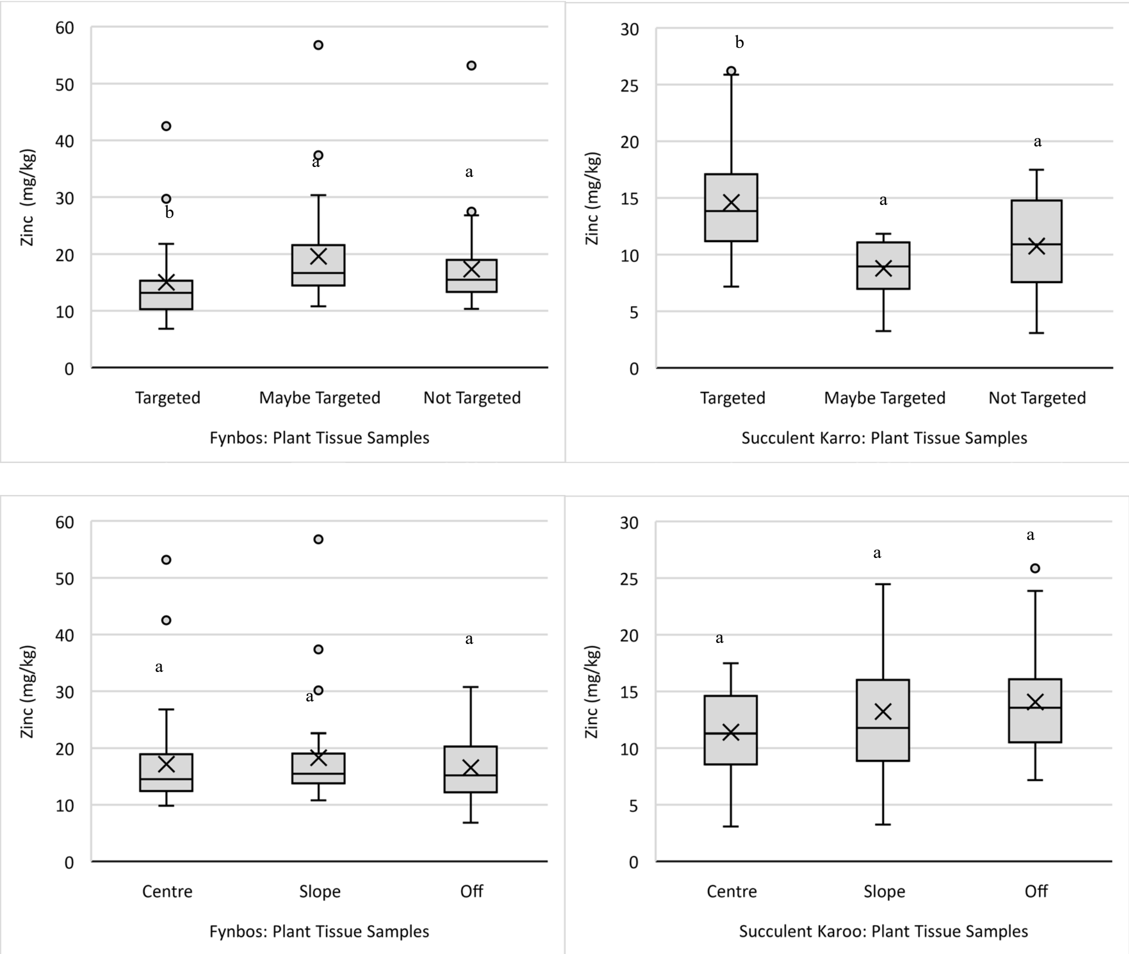


Figure QQQ: Boxplots showing the comparison of zinc in dried plant material in 1) plants "Targeted" or "Not Targeted" by termites and rodents and 2) Where the plants can be found growing on the heuweltjie mounds. Groups with the same letter on each graph are not significantly different ($p < 0.05$) from each other, with different letters indication significant differences according to pairwise comparisons run in R using Wilcoxon rank sum tests.

Average of Zinc (mg/kg)				
Row Labels	**Centre**	**Slope**	**Off**	**Targeted Total**
Fynbos	**17,17**	**18,30**	**16,53**	**17,33**
Targeted	16,23		13,82	15,02
Maybe Targeted		19,20	20,79	19,60
Not Targeted	18,11	15,59	17,71	17,38
Succulent Karoo	**11,39**	**13,20**	**14,06**	**12,92**
Targeted	12,16	17,59	14,06	14,59
Maybe Targeted		8,81		8,81
Not Targeted	10,76			10,76

Manganese

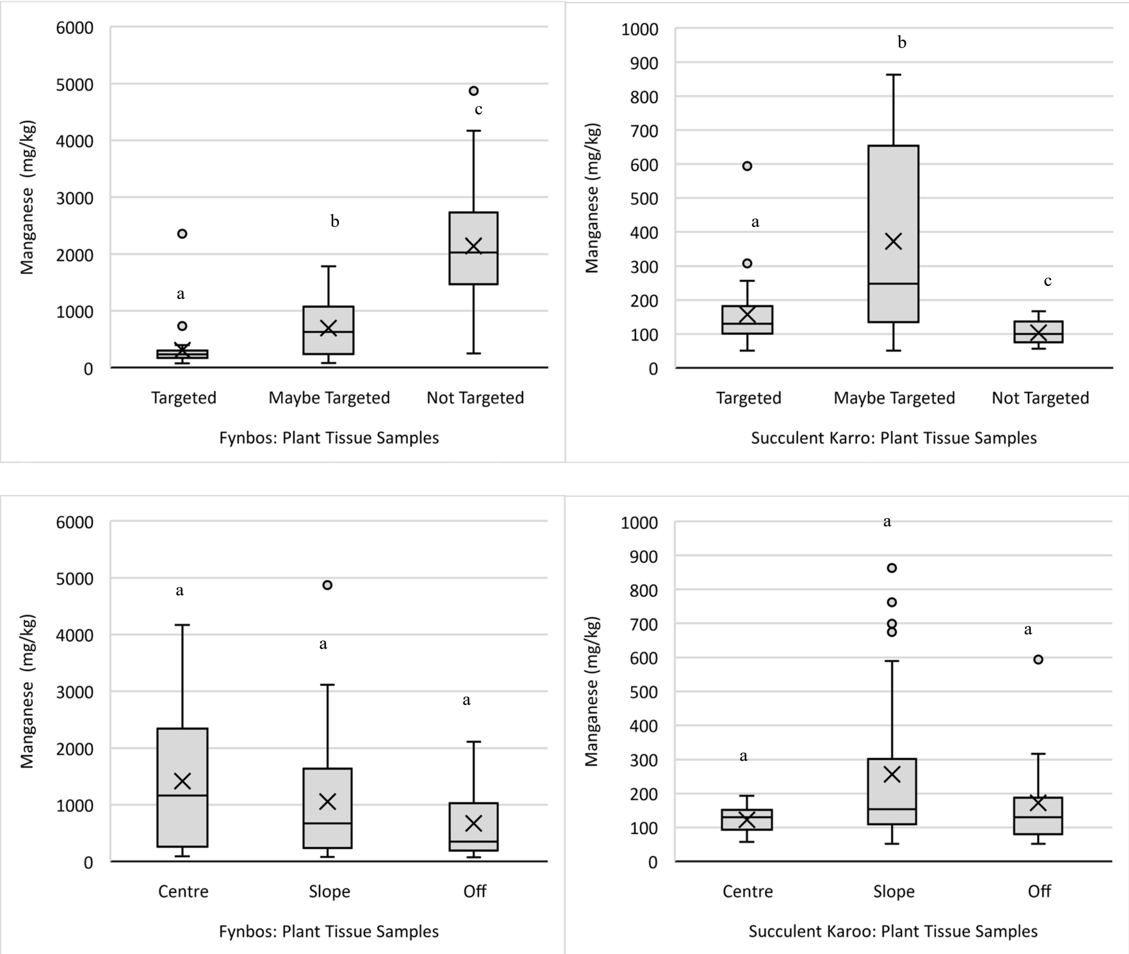


Figure RRR: Boxplots showing the comparison of manganese in dried plant material in 1) plants "Targeted" or "Not Targeted" by termites and rodents and 2) Where the plants can be found growing on the heuweltjie mounds. Groups with the same letter on each graph are not significantly different ($p < 0.05$) from each other, with different letters indication significant differences according to pairwise comparisons run in R using Wilcoxon rank sum tests.

Average of Manganese (mg/kg)				
Row Labels	**Centre**	**Slope**	**Off**	**Targeted Total**
Fynbos	**1414,92**	**1055,57**	**674,16**	**1048,22**
Targeted	414,98		197,50	306,24
Maybe Targeted		624,11	916,49	697,20
Not Targeted	2414,86	2349,96	1385,16	2141,21
Succulent Karoo	**122,48**	**256,27**	**171,91**	**185,67**
Targeted	145,73	140,47	171,91	157,85
Maybe Targeted		372,07		372,07
Not Targeted	103,59			103,59

Boron

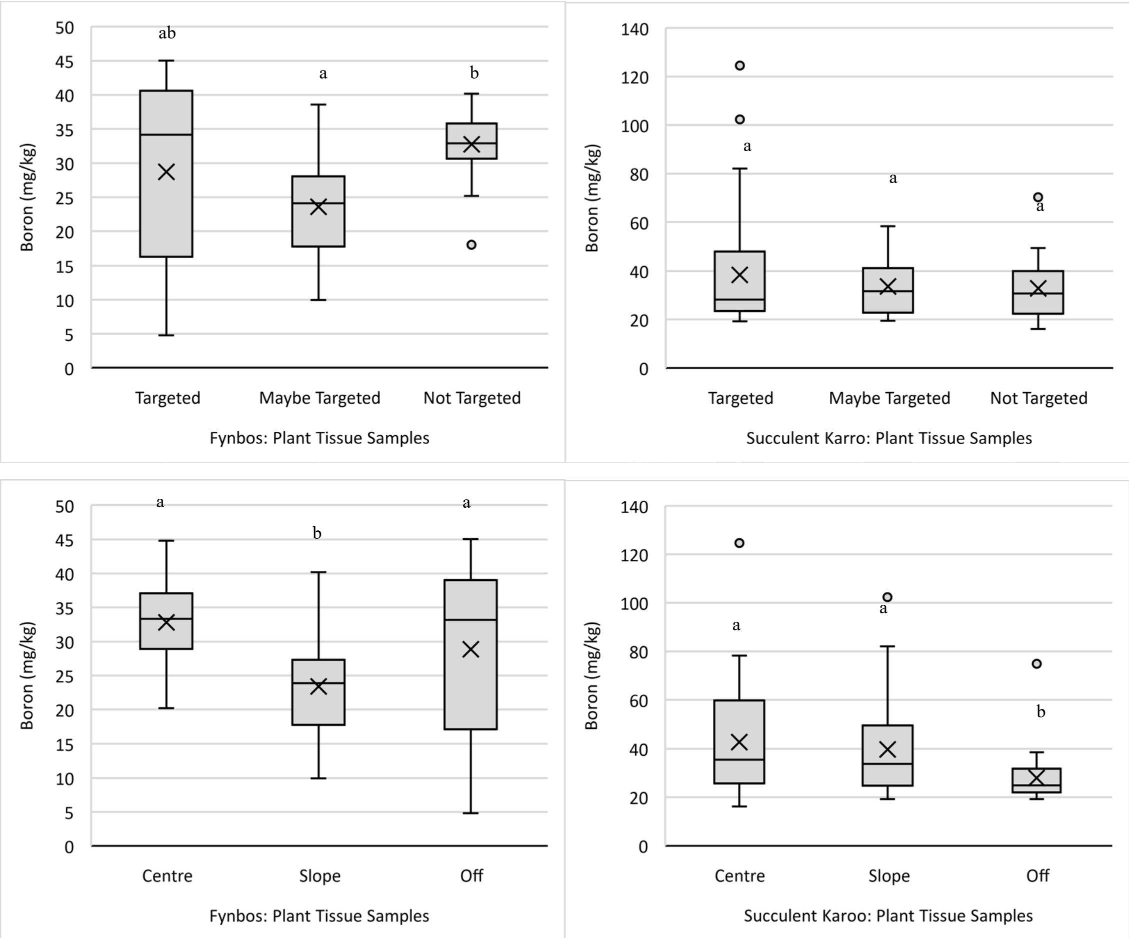


Figure SSS: Boxplots showing the comparison of boron in dried plant material in 1) plants "Targeted" or "Not Targeted" by termites and rodents and 2) Where the plants can be found growing on the heuweltjie mounds. Groups with the same letter on each graph are not significantly different ($p < 0.05$) from each other, with different letters indication significant differences according to pairwise comparisons run in R using Wilcoxon rank sum tests.

Average of Boron (mg/kg)				
Row Labels	**Centre**	**Slope**	**Off**	**Targeted Total**
Fynbos	**32,79**	**23,39**	**28,84**	**28,34**
Targeted	32,61		24,79	28,70
Maybe Targeted		20,65	32,33	23,57
Not Targeted	32,98	31,62	33,44	32,75
Succulent Karoo	**42,70**	**39,65**	**27,90**	**36,65**
Targeted	54,79	45,69	27,90	38,47
Maybe Targeted		33,61		33,61
Not Targeted	32,88			32,88

<u>Aluminium</u>

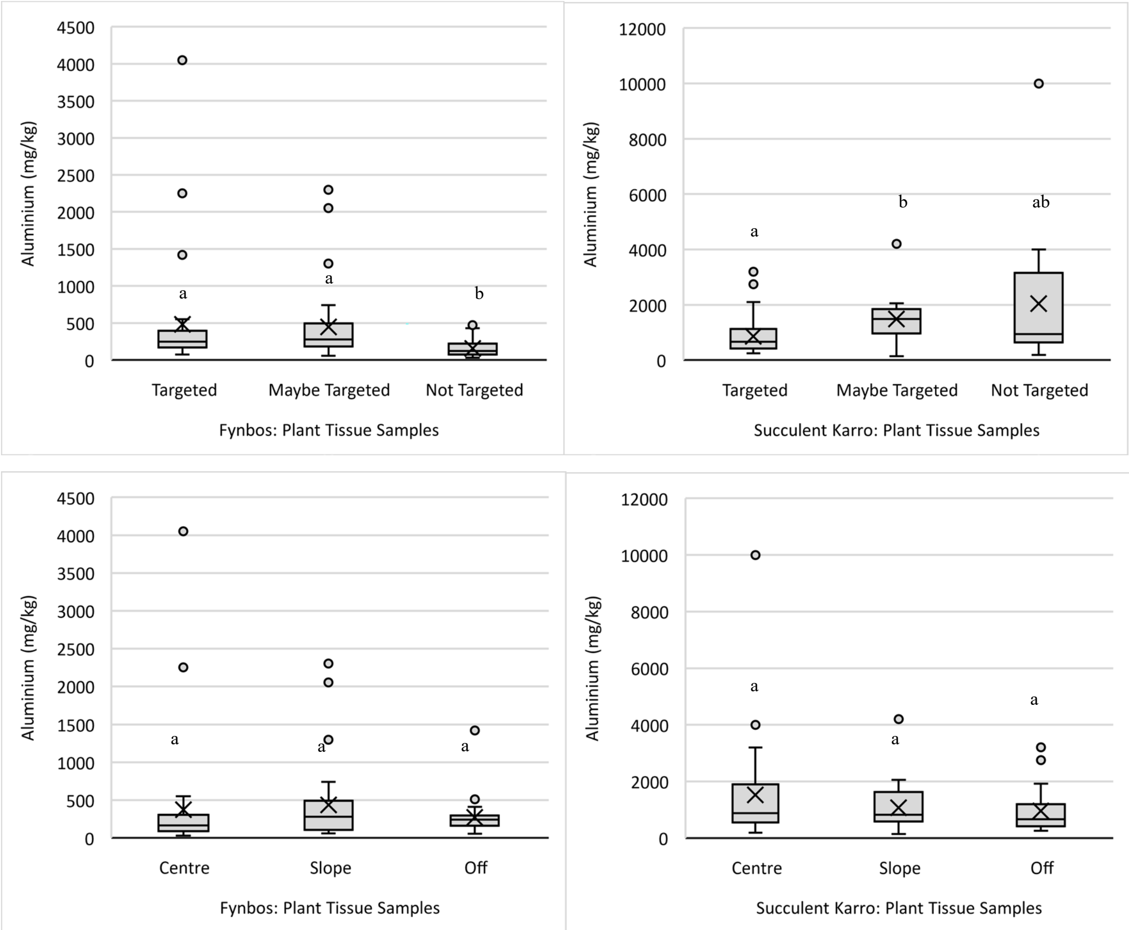


Figure TTT: Boxplots showing the comparison of aluminium in dried plant material in 1) plants "Targeted" or "Not Targeted" by termites and rodents and 2) Where the plants can be found growing on the heuweltjie mounds. Groups with the same letter on each graph are not significantly different ($p < 0.05$) from each other, with different letters indication significant differences according to pairwise comparisons run in R using Wilcoxon rank sum tests.

Average of Aluminium (mg/kg)				
Row Labels	**Centre**	**Slope**	**Off**	**Targeted Total**
Fynbos	**374,03**	**437,34**	**272,88**	**361,42**
Targeted	615,88		345,00	480,44
Maybe Targeted		510,63	252,50	446,09
Not Targeted	132,19	217,50	149,00	157,72
Succulent Karoo	**1528,97**	**1066,88**	**953,27**	**1174,26**
Targeted	896,17	641,16	953,27	857,67
Maybe Targeted		1492,59		1492,59
Not Targeted	2043,13			2043,13

Iron

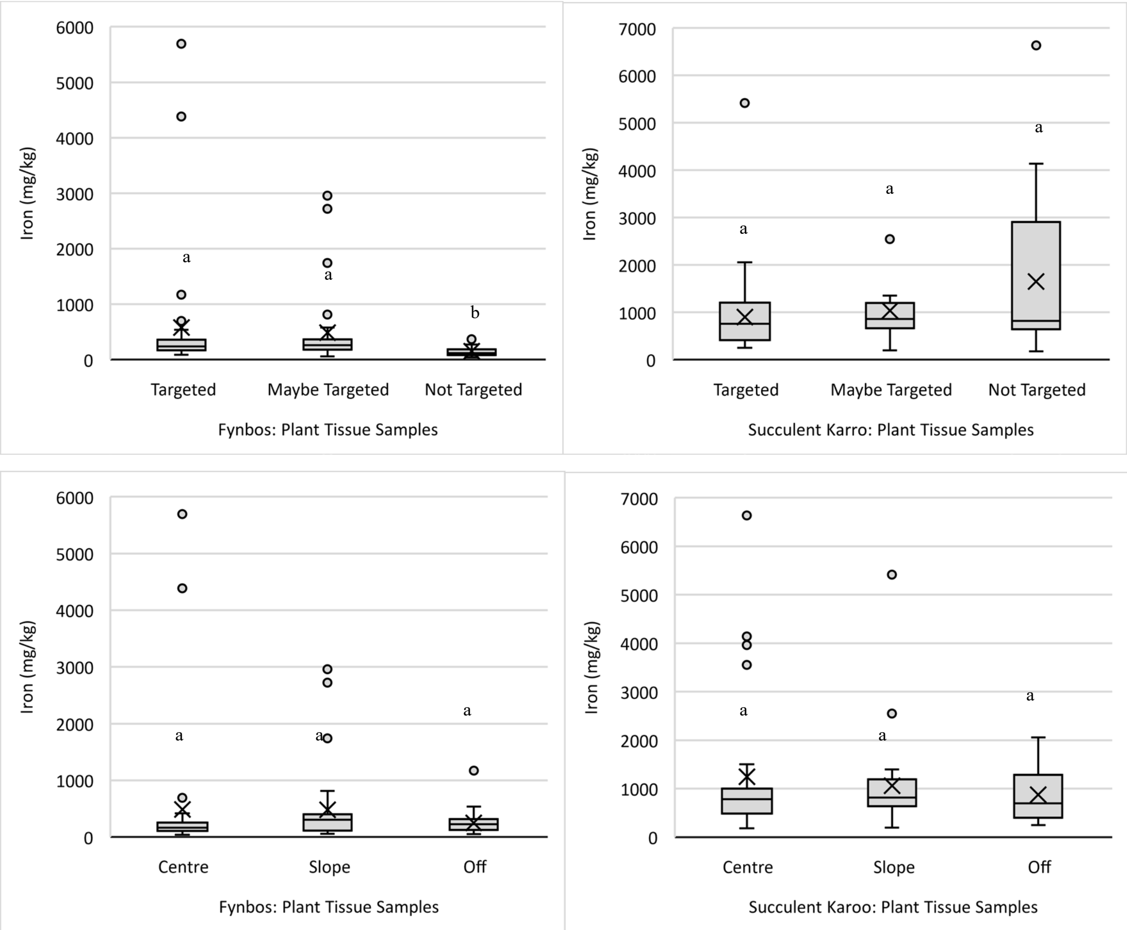


Figure UUU: Boxplots showing the comparison of iron in dried plant material in 1) plants "Targeted" or "Not Targeted" by termites and rodents and 2) Where the plants can be found growing on the heuweltjie mounds. Groups with the same letter on each graph are not significantly different (p < 0.05) from each other, with different letters indication significant differences according to pairwise comparisons run in R using Wilcoxon rank sum tests.

Average of Iron (mg/kg)				
Row Labels	**Centre**	**Slope**	**Off**	**Targeted Total**
Fynbos	**485,95**	**479,10**	**251,02**	**405,35**
Targeted	835,04		325,21	580,12
Maybe Targeted		569,43	228,69	484,24
Not Targeted	136,86	208,13	124,96	151,70
Succulent Karoo	**1244,62**	**1060,86**	**873,75**	**1055,73**
Targeted	741,63	1086,42	873,75	901,84
Maybe Targeted		1035,29		1035,29
Not Targeted	1653,29			1653,29

Appendix 11: XRD results showing mineral presence in all species

Table A: XRD results showing mineral presence in all plant species

Biome	Species code	Species name	Functional type	Weddellite $Ca(C_2O_4)\cdot 2H_2O$	Whewellite ($CaC_2O_4\cdot H_2O$)	Sodium oxalate	Pentahydrite ($MgSO_4.5H_20$)	Halite (NaCl)
Fynbos	CH (n=1)	Searsia undulata	Small tree		*			
	Cl (n=1)	Ruschia lunulata	Succulent Shrub	*	*		*	*
	CS (n=1)	Metalasia muricata	Low Shrub					
	CT (n=1)	Thamnochortus insignis	Graminoid-Restio					
	FB (n=2)	Chlorophytum triflorum	Geophytic Herb		*			*
	FE (n=4)	Ruschia suaveolens	Succulent Shrub	*	*		*	*
	FH (n=1)	Wiborgia sericea	Low Shrub					
	FI (n=1)	Wiborgia sericea	Low Shrub		*			
Succulent Karoo	Sp10 (n=4)	Tetragonia microptera	Succulent Herb		*	*	*	*
	Sp15 (n=1)	Ruschia leucosperma	Succulent Shrub		*	*		*
	Sp16 (n=1)	Jordaaniella cuprea	Succulent Shrub		*	*	*	*
	Sp25 (n=25)	Lotononis falcata	Herb		*	*		
	Sp40 (n=1)	Ruschia centrocapsula?	Succulent Shrub		*	*		
	Sp5 (n=1)	Mesembryanthemum barklyi	Succulent Herb		*	*		*
	Sp8 (n=1)	Cheirodopsis denticulata	Succulent Shrub		*	*		*
	Sp99 (n=1)	Mesembryanthemum hypertrophicum	Succulent Herb		*	*		*

Appendix 12: Correlations: Surface washes

Table B: correlations of EC, surface area, plant wash concentrations (Meq/l) and corresponding plant tissue samples (%)

All data r	*EC Avg*	*surface area*	*Cl MeqL*	*Na MeqL*	*K MeqL*	*Mg MeqL*	*Ca MeqL*	*Sum Cations*	*Sum anions*	*Charge balance*	*CBE*	*K (%)*	*Ca (%)*	*Mg (%)*	*Na (%)*
EC Avg	1,000														
surface area	0,031	1,000													
Cl_MeqL	0,936	0,039	1,000												
Na_MeqL	0,978	0,056	0,980	1,000											
K_MeqL	0,767	-0,081	0,561	0,650	1,000										
Mg_MeqL	0,870	-0,057	0,723	0,791	0,849	1,000									
Ca_MeqL	0,000	-0,129	-0,034	-0,057	0,140	0,319	1,000								
Sum Cations	0,991	0,036	0,965	0,994	0,724	0,847	0,002	1,000							
Sum anions	0,947	0,034	0,999	0,984	0,595	0,747	-0,024	0,973	1,000						
Charge balance	0,745	0,027	0,484	0,639	0,842	0,829	0,084	0,697	0,512	1,000					
CBE	-0,001	-0,027	-0,094	-0,038	0,150	0,192	0,279	-0,002	-0,085	0,255	1,000				
K (%)	0,088	0,214	0,059	0,063	0,158	0,087	0,018	0,076	0,063	0,086	-0,119	1,000			
Ca (%)	0,114	0,228	0,111	0,112	0,057	0,120	-0,028	0,111	0,115	0,056	0,112	0,184	1,000		
Mg(%)	-0,018	0,072	-0,028	-0,030	0,010	0,068	-0,005	-0,023	-0,024	-0,011	0,042	0,505	0,528	1,000	
Na (%)	0,775	0,252	0,808	0,816	0,304	0,553	-0,060	0,784	0,800	0,431	-0,054	0,183	0,150	0,084	1,000

Appendix 13: Photos of foraging termites foraging

Appendix 14- Photos of rodent nest and faeces samples

Fynbos rodent nests collected from midden located under a bush on a heuweltjie

Fynbos rodent nests collected inside the heuweltjie

S Karoo rodent nests collected and excavated burrow/hole

Appendix 15: Photos of different termite stick-piles

Succulent Karoo more disturbed (buffelsivier) and less disturbed (kommagas)

Fynbos More Disturbed Site

Fynbos Less Disturbed Site

Appendix 16: Termite observations Plant species list

Table C: List of plant species collected by termites from observations

Code	Family	Species name	Growth form
Fynbos			
AA=Q	Asteraceae	Arctotheca calendula	Herb
AW	Aizoaceae	Aizoaceae sp	Succulent shrub
CF	Asteraceae	Eriocephalus africanus	Low shrub
CH	Anacardiaceae	Searsia undulata	Small tree
CJ		Heliophila arenaria	Herb
CL =EO	Aizoaceae	Ruschia lunulata	Succulent Shrub
CP= CT	Restionaceae	Thamnochortus insignis	Graminoid-Restio
CQ		Paranomus?	Low Shrub
CS	Asteraceae	Metalasia muricata	Low Shrub
CX			Climber
DF			Low Shrub
DO	Rutaceae	Macrostylis hirta	Low shrub
DV	Asparagaceae	Lanchenalia sp	Geophytic Herb
E	Asteraceae	Cotula pruinosa	Herb
FA	Oxalidaceae	Oxalis purpurea	Herb
FB	Agavaceae	Chlorophytum triflorum	Geophytic Herb
J	Iridaceae	Lapeirousis fabicii	Geophytic Herb
	Poaceae	Poaceae sp	
	Oxalidaceae	Oxalis sp	
S Karoo			
Sp10	Aizoaceae	Tetragonia microptera	Succulent Herb
15	Aizoaceae	Ruschia leucosperma	Succulent Shrub
16	Aizoaceae	Jordaaniella cuprea	Succulent Shrub
46	Aizoaceae	Mesembryanthemum deciduum	Succulent Shrub
4=57	Oxalidaceae	Oxalis annae	Herb
44	Asteraceae	Gazania difusa	Herb

Appendix 17: Diversity Variables pooled and averaged across the study sites

Table K: The means and SD of the vegetation and diversity variables, and their significance, following Welch Two Sample t-tests, to compare each biome and disturbance subsite. Differences between biomes are shown using capital letters, comparing each variable vertically, while differences between the disturbance levels are shown using uncapitalised letters, compared horizontally; p < 0.05; p < 0.01**; p < 0.001***.*

Variable	Biome	Biome Means ± SD (Fynbos: n=72 S Karoo n=99)	Significance level (p value):	More Disturbed (MD) Means ± SD (n=36)	Less Disturbed (LD) Means ± SD (Fynbos: n=36 S Karoo n=63)	Significance level (p value):
Vegetation cover (%)	Fynbos	72.78 ±16.78A	***	67.69±15.68 a	77.86±16.50 b	**
	S. Karoo	37.38 ±16.98B		41.33±15.67 a	35.13±17.40 a	
Average height (cm)	Fynbos	44.37 ±21.08A	***	38.19±16.83 a	50.56±23.23 b	*
	S. Karoo	9.26 ±3.33B		8.66±2.67 a	9.61±3.63 a	
Abundance (m^2)	Fynbos	23.02± 16.50A		24.01±17.64 a	22.03±15.47 a	
	S. Karoo	26.08± 18.49A		42.37±18.28 a	16.78±10.48 b	***
Species richness (m^2)	Fynbos	3.12± 1.07 A	***	2.94±1.04 a	3.30± 4.33 a	
	S. Karoo	2.13± 0.70 B		2.13± 0.50 a	2.14± 3.18 a	
Shannon (H) diversity	Fynbos	1.90 ±0.38 A	***	1.83±0.36 a	1.97±0.39 a	
	S. Karoo	1.43±0.45 B		1.19±0.37 a	1.57±0.44 b	***
Simpson (H) diversity	Fynbos	0.78 ±0.10 A	***	0.77±0.11 a	0.79±0.10 a	
	S. Karoo	0.65 ±0.16 B		0.56±0.17 a	0.70±0.14 b	***
Pielou's evenness (J)	Fynbos	0.78 ±0.11 A	***	0.77±0.12 a	0.78±0.11 a	
	S. Karoo	0.68 ±0.17 B		0.55±0.15 a	0.75±0.13 b	***
Boxplots and t-tests can be found in Appendix 2						

Appendix 18: Figure 3.1 without the plants with bladder cells

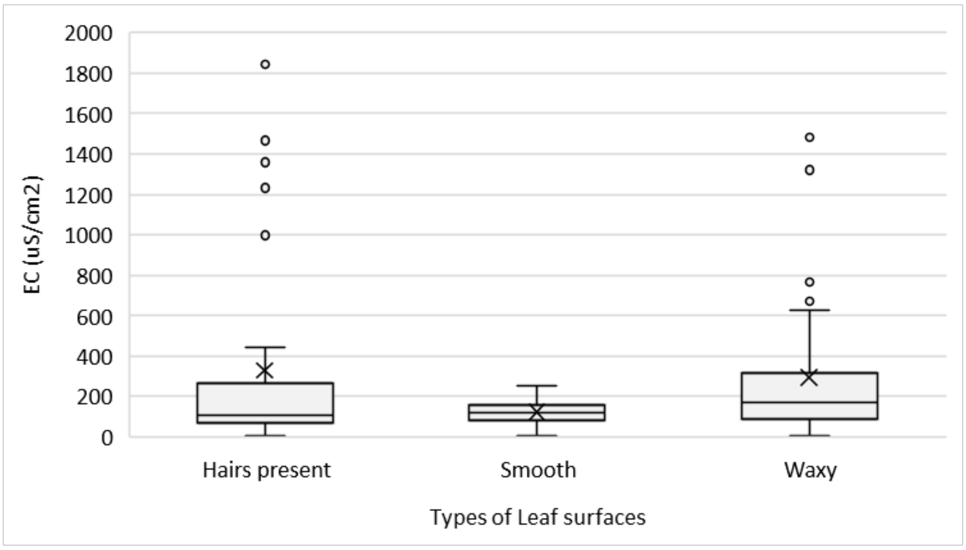


Figure #:: EC (µS/cm) of the solutions washed off of plant surfaces catergorised by the different plant surface types / epidermis.