ENDEMISM, DIVERSITY AND PRIORITIES FOR THE CONSERVATION OF SERPENTINE AREAS IN THE BARBERTON GREENSTONE BELT, MPUMALANGA, SOUTH AFRICA

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A Thesis submitted to the Faculty of Science, University of the Witwatersrand, in fulfilment of the requirement for the degree of Doctor of Philosophy.

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DECLARATION

I declare that this Thesis is my own, unaided work. It is being submitted for the Degree of Doctor of Philosophy at the University of the Witwatersrand, Johannesburg. It has not been submitted before for any degree or examination at any other University.

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ABSTRACT

This study aims to provide a comprehensive description of the Flora, biogeography and diversity of the serpentine outcrops of Barberton Greenstone Belt in Mpumalanga, South Africa in order to set conservation priorities for these areas. About 30 large and many smaller serpentine outcrops form part of the Barberton Greenstone Belt and consist of various combinations of serpentinized minerals. Seven outcrops were selected to be studied in detail.

A floristic analysis recorded 744 species and subspecies, 319 genera and 94 families. The flora includes 33 taxa endemic to serpentine soils and six taxa, which are hyperaccumulators of nickel. The endemic taxa make up 41 % of the endemics of the Barberton Centre of Endemism. The serpentine flora was found to be different to the surrounding non-serpentine vegetation in terms of numbers of species per family, the ratios of dicotyledons to monocotyledons and familial composition. The Asteraceae and the Anacardiaceae support a higher number of endemics than expected, which suggests genetic pre-adaptation within these families and specifically within the genera *Berkheya, Helichrysum* and *Ozoroa*. Most of the endemic taxa exhibit long-range dispersal suggesting gene flow between populations on different outcrops. The endemics represent a mix of neo-endemics and paleoendemics.

Non-parametric species richness estimators used to predict the species richness of each site, indicated that five serpentine outcrops have higher species richness than the surrounding non-serpentine areas. Indices of diversity calculated showed similar patterns to those of the species richness estimates. The Barberton Greenstone Belt serpentine outcrops show relatively high plant diversity when compared to some other serpentine outcrops around the world. Beta diversity calculated for each site was not correlated with altitude and weakly correlated with the size of outcrops. Species turnover between outcrops is high and is positively correlated with the geographical distance between outcrops. Diversity at higher taxonomic levels were calculated, and results suggest that genera have some potential for facilitating the ranking of outcrops in terms of biological richness to select sites for conservation planning. Less than 30% of serpentine outcrops are adequately conserved. Species and genus richness and endemism were used to select five outcrops that have high conservation priority.

To my husband John

and my children Alex and Christine

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Chapter 1

General Introduction

‘Serpentine’ was a broad term used by biologists to describe a group of ultramafic rocks and the soils derived from them (Proctor and Woodell 1975). These serpentines and their floras of North America and Europe have stimulated the interest of taxonomists, ecophysiologists, soil scientists and evolutionary biologists since the 1940’s (Kruckeberg 1992). In its strictest sense, ‘serpentine’ soils are those derived from serpentinite rocks containing the minerals: Antigorite, Chrysotile and Lizardite and contain high concentrations of magnesium and iron (Brooks 1987). In most studies and this study, the term serpentine has been and will be used to describe rocks containing the serpentinite minerals and the soils derived from these rocks. The term ‘ultramafic’ will be used to describe those rich in Ferro-magnesium minerals, even though they may not contain some of the minerals of serpentines in the strict sense.

The soils derived from serpentine rocks can vary from one site to another as the soil produced during the weathering process is also dependent on the climate, relief, time and biological activity (Brooks 1987). However, most serpentine soils have unusaully high levels of heavy metal such as nickel, chromium, cobalt and iron; high magnesium to calcium ratios (Brooks and Yang 1984, Kruckeberg 2013) and low concentrations of plant nutrients such as nitrogen, phosphorus and potassium (Brooks 1987). Serpentine soils support a very unusual and specialized flora made up of species that have evolved some degree of resistance to these metals. Consequently, there is usually a sharp contrast between the serpentine flora and that of the surrounding areas. The survival of taxa with such restricted distribution depends largely on the availability of suitable habitats (Baker, Brooks and Reeves 1988). Although there is some variation between all serpentine sites, Whittaker (1954) recognised poor plant productivity, high rates of endemism and vegetation distinct from those of neighbouring areas as collective traits.

Serpentines often show distinctive floras and communities that are markedly set apart from those on adjacent non-serpentine substrates. However, in addition to the species composition of serpentine vegetation being different to the species composition of the surrounding non-serpentine vegetation, a further effect of serpentine is the inclusion of a number of taxa that are endemic to the serpentine soils i.e. only grow on the serpentine soils of a particular area.

Biodiversity of serpentine floras

Due to the insular pattern of serpentine soils in the Barberton Greenstone Belt, these sites can be compared to “Terrestrial Islands”, because they are isolated from one another by substrates that the serpentine endemics cannot colonize. The equilibrium theory predicts that the insularity of these substrates will result in low species diversity of the vegetation. However, it has been suggested that some “islands” can re-establish diversity after a sufficiently long geological time period (Brown 1988). It is possible that this could have occurred on the serpentine sites of the Barberton Greenstone Belt as they are thought to be very old (Brooks 1987). One of the main assumptions of Island Diversity is that species numbers increase with “island” area (Bond 1991). Linked to this is the assumption that the diversity of plants in these areas will increase with an increase in niche or habitat diversity (Diamond 1988). For these reasons, it is thought that the large serpentine sites of the Barberton Greenstone Belt would show high botanical diversity.

Many studies also show that biodiversity decreases with an increase in altitude (Brown 1988). This relationship could be tested on serpentine sites due to the range of altitudes at which they are found. The distances between serpentine sites should affect their species composition and species richness. Sites closest to one another should share a high percentage of species and show high species diversity due to the short dispersal distances required for immigration from one site to another.

Diversity measures can be applied in two main areas. The first is in environmental monitoring, where the main assumption is that the adverse effects of pollution will be reflected in a reduction of diversity or a change in the relative abundances of certain species. The second application is in conservation which relies on the concept that species-rich communities are more valuable than species-poor ones (Magurran 1988). Effective conservation and environmental management are dependent on baseline data on biological diversity across a range of taxa and at a variety of scales (Magurran 2004). Environmental monitoring makes extensive use of diversity indices and species abundance distributions while conservation management relies more on measures of species richness. However, it can be said that conservation strategies would probably be improved if data on species abundance patterns were taken into account.

Conservation of Serpentine Floras and their habitats

A review of the recent literature, including the Proceedings of the recent International Conferences on Serpentine Ecology, shows a focus of research on the physiology of hyperaccumulators (Boyd *et al.* 2009), the use of hyperaccumulators for phytoremediation and phytomining operations (O'Dell and Claassen 2009) and the development of ecological and evolutionary theory (Harrison and Rajakaruna 2011). Habitats with serpentine soils provide ample opportunities for conservation- and restoration-oriented research directed at finding ways to improve management of these biodiversity hotspots. However, there are very few recent publications of research into the conservation and management of these floras.

Although serpentine soils occur all over the world, there is only one report of actions being taken to conserve these unique areas. The conservation of serpentine sites of California was discussed in 1986 at a conference on Rare and Endangered Plants (Reeves 1992). This resulted in a number of papers on conservation and management of serpentine endemics and their habitats. These reported on a large number of rare and endemic plants that were threatened by mining activities, industry and recreational traffic. However, implementation of preservation protocols suggested in these reports has lagged, and a number of sites that need to be conserved are still left unprotected.

Importance of serpentine floras

All serpentine sites are considered to be important as they are reservoirs of rarities that have an untapped potential to contribute to fundamental research into of their unique properties (Whiting *et al*. 2004). Serpentine soils are found worldwide and studies of these soils and their floras have been conducted as far afield as California (Kruckeberg 1984, Callizo 1992 and McCarten 1992), Cuba (Borhidi 1992), Brazil (Brooks *et al.* (1992), Italy (Ferrari, Lombini and Carpené 1992 and Verger 1992), Japan (Toyokuni 1992), Australia (Batianoff, Neldner & Singh 2000, Gibson and Lyons 2001) and New Caledonia (Jaffré 1992). Floristic studies in areas that have been well studied (e.g. California and New Caledonia) have laid the foundation for ongoing research in endemism (e.g. Safford *et al.* 2005), plant diversity (e.g. Harrison *et al.* 2006, Selvi 2007) the physiology of hyperaccumulators (Boyd *et al.* 2009) and evolutionary theory (Harrison and Rajakaruna 2011). Research on the less well-studied serpentine outcrops and their biota still has much to contribute to our understanding of evolution and ecology and could show some serpentine areas to be of major economic importance. In addition, serpentines are going to present formidable problems of conservation especially in the area of phytomining and phytoremediation (O'Dell and Claassen 2009). In South Africa, serpentine outcrops occur in the Greenstone Belt near and around Barberton in Mpumalanga and Swaziland and a band from Potgietersrus to Duiwelskloof, Northern Province (Balkwill *et al.* 1997). The sites investigated for this study are all within the Barberton Greenstone Belt.

Characteristics of the Barberton Greenstone Belt

The Barberton Greenstone Belt occurs in south-eastern Mpumalanga, South Africa and is a largely triangular mafic to ultramafic geological intrusion extending from Malalane in the east to Badplaas and Barberton in the south and ending just west of Mbombela (Nelspruit). The Barberton Greenstone Belt is surrounded by extensive granitoid plutons and gabbroid intrusions (Ward 2000). Within this ultramafic intrusion are about 30 large (i.e. >1 km^2) serpentinite (hereafter referred to as 'serpentine') outcrops and many smaller outcrops (Figure 1 of Chapter 2). The serpentine outcrops consist of various combinations of serpentinized dunite, amphibolite, chrysotile asbestos and Peridotite (Morrey *et al.* 1992). The outcrops vary in size from 0.1 km^2 to ca. 19 km^2 with a mean size of 2.6 km^2. Some are separated from other outcrops by up to 20 km (Balkwill *et al.* 1997). The outcrops occur in mountainous areas and are heterogeneous in altitude, slope, soil depth, etc. The serpentine vegetation falls within the Mixed Lowveld Bushveld, Sour Lowveld Bushveld and North-eastern Mountain Grassland vegetation types (Low and Rebelo 1996). The vegetation type has more recently been reclassified as Barberton Serpentine Sourveld by Mucina and Rutherford (2006) due to the unique, stunted woody vegetation that results from the high toxicity of the soils.

The lack of knowledge of the floras of the metalliferous sites in South Africa initiated a program, funded by the Foundation for Research and Development (The National Research Foundation prior to 1999), entitled "Metalliferous Flora" and was focused on the study of floristics, biodiversity, conservation, soils and evolution of these floras. Together with the research conducted by Kidger (1993), Changwe and Balkwill (2003) and McCallum (2006) this study forms part of this program and provides a descriptive analysis and characterization of the flora of the Barberton Greenstone Belt in terms of biodiversity, endemic species and conservation.

Aim and objectives of the project

The overall aim of this project was **to assess which biological and physical characteristics could be used to determine conservation priorities for the vegetation of serpentine outcrops in the Barberton Greenstone Belt.**

To meet this aim the objectives were to:

1) comprehensively characterize endemism, diversity and physical properties of selected serpentine sites that cover the full range of altitude, size and latitudinal and longitudinal position of the sites;
2) compare the species composition of the vegetation of selected sites and characteristics of endemic species to those of the adjacent non-serpentine vegetation;
3) determine the relationship between levels of endemism and diversity to physical properties of these sites and
4) evaluate and critically analyze the potential of models to predict priorities for the conservation of serpentine sites.

In addition to and as a result of the information arising from these objectives, the conservation status of the serpentine outcrops of the Barberton Greenstone Belt was to be assessed and outcrops of high conservation priority identified.

Key Questions and Hypotheses

The following key questions were answered while attaining the first objective:

1. Are levels of endemism higher on serpentine sites than on other terrestrial “islands”?
2. How do the levels of endemism on serpentine sites compare to the levels of endemism in the province?

3. Is each of the serpentine endemics restricted to one site or are they found on a number or all the sites in the Barberton Greenstone Belt?
4. Does the richness of higher taxa reflect levels of species richness in serpentine vegetation?
5. What levels of gamma diversity are shown by the serpentine sites of the Barberton Greenstone Belt?

The following two key questions were answered to attain the second objective:

1. How does the species composition of serpentine vegetation differ from the species composition of the surrounding non-serpentine vegetation?
2. Do serpentine endemics exhibit different profiles to endemic species of other terrestrial "islands" and to species found adjacent to but not on serpentine?

The following key question was answered while attaining the third objective:

> Is there a correlation between levels of beta diversity (within serpentine site diversity) of the vegetation and the diversity of physical properties such as altitude, area, soil characteristics, topography, etc.?

The following key questions were answered while attaining the fourth objective:

1. Which of the 36 mapped sites do the different models, used to select priority areas for conservation, show to have high conservation priorities?
2. Do the different models select the same or different sites as priorities?
3. If different sites are selected by the models, what is the basis for this difference?

The hypotheses formulated for this study are as follows:

1. Serpentine sites show high levels of richness per area at the family, genus and species level when compared to adjacent non-serpentine areas.
2. Levels of endemism on serpentine sites in the Barberton Greenstone Belt will increase with an increase in the range of altitude, size and diversity of habitats of each site.
3. Endemics from the serpentine of the Barberton Greenstone Belt are palaeoendemics with widespread distributions amongst serpentine sites.
4. The profile of serpentine endemics will match the profile of the species found in

the surrounding matrix.

5. The turnover of taxa between sites with different altitudes and latitudinal and longitudinal ranges is higher than that of sites with similar altitudes and that are closer together.
6. A large number of the serpentine sites of the Barberton Greenstone Belt need to be conserved to preserve the endemics and the diversity of the serpentine sites.

Study sites

A few sites have been selected to be studied in detail. See Table 2.1 (Chapter 2) of sites selected and Figure 2.1 (Chapter 2) for their location. These selections were made so that presumably the full range of variation will be accounted for in the study. The Kaapsehoop and Agnes Mine sites, which are intermediate in locality, size and altitude, have already been studied in relative detail (Williamson 1994). Those results will be compared with the results of this study.

The Sawmill and Kalkkloof sites were selected over the Forbes Reef and Motjane sites (which have a higher altitude and are situated further south) as these sites have been studied in detail in a separate investigation (McCallum 2006). Rosentuin was selected over Heemstede and Noisy as the smallest sites. Heemstede is almost completely planted to pine with very little natural vegetation remaining. Noisy is not very accessible as it is even difficult to reach with a four-wheel drive vehicle.

Structure of the thesis

Chapter 1 provides a general introduction to serpentine, the background of the study and the aims and objectives of the study. The general methods employed are outlined with a description of the Barberton Greenstone Belt and the selected study sites.

Chapter 2 describes the floristics of the selected serpentine sites of the Barberton Greenstone Belt and provides an analysis of the important genera and families occurring on these serpentine soils. Taxa that are indicators of serpentine soils, which are excluded from serpentine soils and that are endemic to the serpentine soils are identified and listed.

Chapter 3 presents levels of endemism determined for the serpentine of the Barberton Greenstone Belt and provides a comparison of endemics on other serpentine areas. The levels of endemism on serpentine are compared to the rest of the Barberton Centre of Endemism and the Mpumalanga province. Levels of threat to serpentine endemics are analysed and presented.

Chapter 4 presents a detailed analysis of species richness and species diversity of the serpentine sites and compares the diversity of the serpentine sites to the surrounding non-serpentine areas.

Levels of beta (within-site) and gamma (between site) diversity of the selected serpentine sites are presented in Chapter 5. These levels of diversity are compared to non-serpentine sites and are correlated with the physical characteristics of sample sites.

Chapter 6 assesses the diversity and richness of the serpentine flora at the generic and family levels and compares these to calculated levels of species richness and diversity.

Chapter 7 reports on the application of a number of conservation models to the serpentine flora in an effort to determine the sites that could have high conservation value and whether different models select the same sites as priorities.

Chapter 8 provides concluding comments for the entire study.

Many of the data resulting from this study will provide a base from which further studies on the evolution of serpentine endemics and on the use of serpentine vegetation for revegetating and rehabilitating the mine dumps of the asbestos mines in the area. All the data gained from this study will add to our current knowledge of serpentines in southern Africa and will help us to reach the level of knowledge of serpentines in other parts of the world.

This study makes a substantial contribution to new knowledge in a number of ways. Only a few of the serpentine sites in the Barberton Greenstone Belt have been studied to this depth previously i.e. the Kaapsehoop and Agnes Mine sites, Swaziland sites and a few sites within the Songimvelo Game Reserve. The serpentines of the rest of the world have also not been studied with this approach, which combines taxonomic descriptions and diversity at various hierarchical levels for conservation purposes. This study addresses some topical issues in the literature such as the theory of Island Biogeography and how it applies to the diversity and endemism of the serpentine floras.

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Chapter 2

Floristic analysis of selected serpentine outcrops of the Barberton Greenstone Belt

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Plant census and floristic analysis of selected serpentine outcrops of the Barberton Greenstone Belt, Mpumalanga, South Africa

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ABSTRACT

This paper aims to characterise and describe the species composition of serpentine sites of the Barberton Greenstone Belt as compared to surrounding non-serpentine areas. A floristic analysis of seven serpentine (serpentinite) outcrops of the Barberton Greenstone Belt, in the eastern part of South Africa, recorded 744 species and subspecies, 319 genera and 94 families. 18 taxa remain undescribed. The Barberton Greenstone Belt flora includes 32 taxa endemic to serpentine soils and six taxa considered to be hyperaccumulators of nickel. The taxa considered to be endemic to serpentine outcrops make up 39% of the number of endemics found within the Barberton Centre of Endemism. The serpentine vegetation is characterised by fewer trees than the surrounding vegetation and the dominance of grass species such as *Themeda triandra*, *Heteropogon contortus* and *Loudetia simplex*. The species composition of each outcrop is relatively unique with only about 30% of species shared between any pair of outcrops. The flora of the serpentine outcrops of the Barberton Greenstone Belt is found to be different to the surrounding non-serpentine vegetation in terms of number of species per family, the ratios of dicotyledons to monocotyledons and familial composition.

1. Introduction

Outcrops of serpentinite (henceforth referred to as 'serpentine') rocks are often referred to as edaphic islands due to their sharp boundaries and patchy distribution. Soils derived from serpentine rocks are considered a harsh environment for plants due to low levels of calcium relative to magnesium, low nutrient content, nickel and chromium toxicity and poor water holding capacity (Harrison et al., 2006). The extreme physical and chemical properties of serpentine soils provide conditions that allow colonisation by tolerant species and then strong diversifying selection processes may lead to ecological speciation (Kruckeberg, 1986; Rajakaruna, 2004). Species of plants tolerant to serpentine soils include species found only on serpentine soils i.e. serpentine endemics; species that are local or regional indicators but are not restricted to serpentine and species that are serpentine indifferent (Kruckeberg, 1984). Taxa that are found on adjacent non-serpentine substrates but are completely excluded from serpentine soils (Harrison et al., 2006) are also important for defining the distinctiveness of serpentine floras.

Physiological and evolutionary mechanisms hypothesised to be responsible for adaptations to serpentine soils include the tolerance of a low calcium-to-magnesium ratio, avoidance of Mg toxicity, or a high Mg requirement (Brady et al., 2005). In a floristic analysis of the serpentine vegetation of Central Queensland, Australia, Batianoff et al. (2000) suggested a family tolerance of soil conditions and postulated that some families are characterised by higher proportions of serpentine tolerant species. It is also thought that edaphic conditions strongly influence species diversity and levels of endemism. Batianoff et al. (2000) found that species richness of the serpentines of Central Queensland in Australia decreased as soil nickel concentrations increased in lowland forests and that levels of endemism increased with increasing nickel concentrations. In the Californian serpentine vegetation, soil calcium levels were negatively correlated with the number of serpentine endemic taxa (Harrison, 1999).

The flora of the serpentine outcrops of the Barberton Greenstone Belt in the eastern parts of South Africa have been less well documented than those of Cuba (Borhidi, 1992), New Caledonia (Jaffré, 1992), California (Kruckeberg, 1984; Callizo, 1992), Zimbabwe (Wild, 1965), Australia (Gibson and Lyons, 1998a,b, 2001; Batianoff et al., 2000) and Italy (Ferrari et al., 1992; Verger, 1992). Most of these studies have lead to the identification of many plant species endemic to serpentine soils. The lack of knowledge of the floras of the metalliferous sites in South Africa initiated a funded research programme entitled 'Metalliferous Flora' and focused on the study of the floristics, biodiversity, conservation, soils and evolution of these floras. This study supplements floristic analyses conducted previously on parts of the Barberton Greenstone Belt by Williamson (1994), Changwe and Balkwill (2003) and McCallum (2006).

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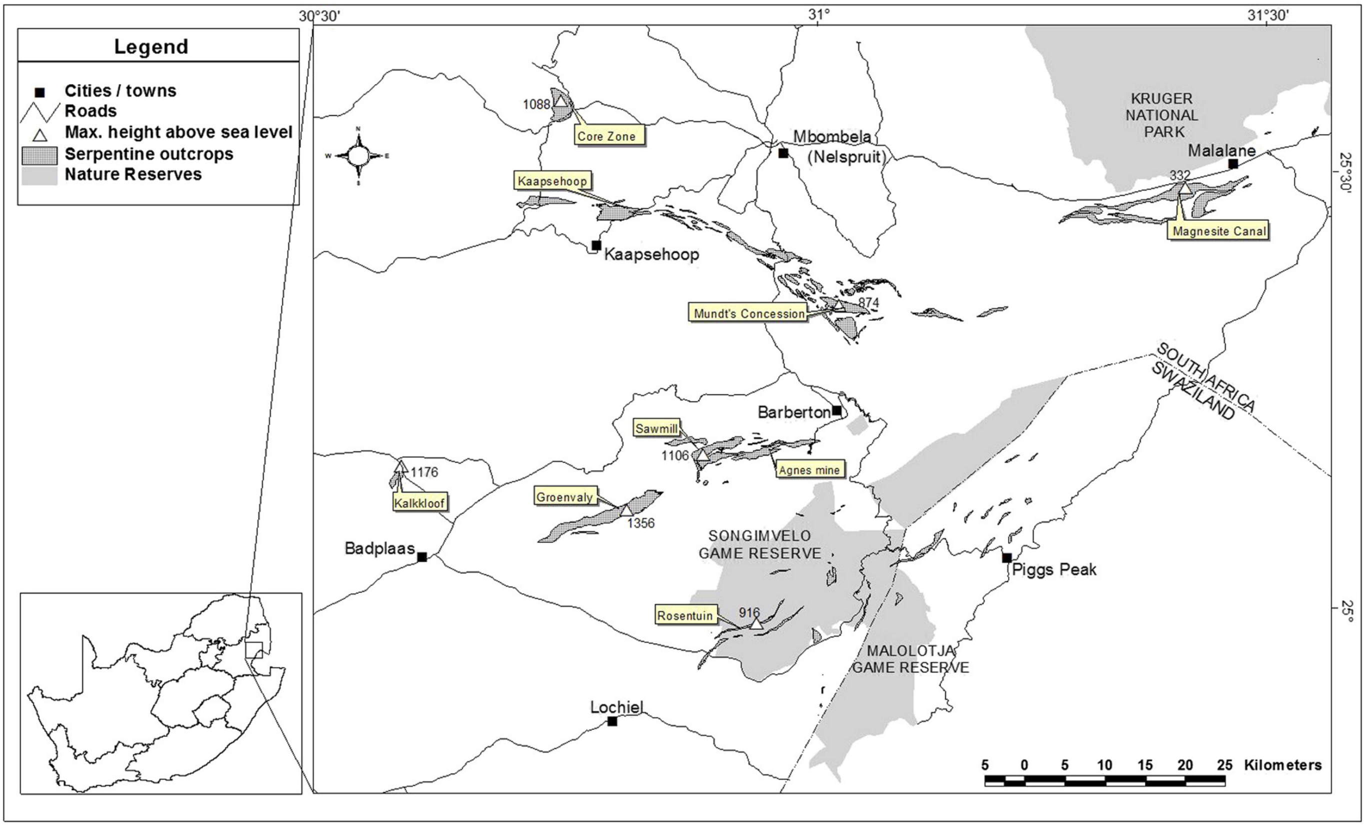

Fig. 1. Map indicating serpentine outcrops of the Barberton Greenstone Belt in Mpumalanga, South Africa. Survey sites are identified by callout labels. Map was prepared using data provided by the Chief Directorate: Surveys and Mapping, Department of Land Affairs, Republic of South Africa.

The Barberton Greenstone Belt is located in south-eastern Mpumalanga, South Africa (Fig. 1). This province has an estimated 4946 plant species and infraspecific taxa occurring within its boundaries, yet it only comprises 6.3% of South Africa's surface area (Lötter et al., 2002). This high level of plant diversity is not evenly distributed across Mpumalanga. Two regions and three Centres of Endemism were recognised by Van Wyk and Smith (2001) and an additional one for the Lydenburg area was proposed by Lötter et al. (2002). The Barberton Greenstone Belt falls within the Barberton Centre of Endemism, which has an area of about 4000 km^2, has about 2210 plant species and more than 80 endemics with 3.6% endemism. A large percentage (>29%) of this area is transformed by commercial plantations of species of *Pinus* and *Eucalyptus* (Lötter et al., 2002), threatening many of the endemics on serpentine and other ultramafic substrates (Williamson and Balkwill, 2006).

The Barberton Greenstone Belt consists of approximately 30 large serpentine outcrops in the belt surrounded by several very small outcrops (Ward, 2000). These outcrops are located in an inverted equilateral triangle centred on Barberton and extending to Malelane in the east and to Badplaas in the south. The Barberton Greenstone Belt is surrounded by extensive granitoid plutons and gabbroid intrusions. The serpentine outcrops consist of various combinations of serpentinised dunite, amphibolite, chrysotile asbestos and peridotite (Morrey et al., 1992). The largest of these outcrops is about 19 km^2, and there are several smaller outcrops (from 0.1 km^2). Some outcrops are separated from others by up to 20 km (Balkwill et al., 1997). The outcrops occur in mountainous areas and are heterogeneous in altitude, slope, soil depth and other topographic features. The serpentine vegetation falls within the Mixed Lowveld Bushveld, Sour Lowveld Bushveld and North-eastern Mountain Grassland vegetation types as described by Low and Rebelo (1996). It has more recently been reclassified as Barberton Serpentine Sourveld by Mucina and Rutherford (2006) due to the unique, stunted woody vegetation that results from the high toxicity of the soils. The landscape of the areas surrounding the serpentine outcrops of the Barberton Greenstone Belt is mostly hilly with varied terrain. The outcrops range from 350 to 1400 m above sea level. The climate of the area is characterised by summer rainfall (MAP 600–1150 mm) with dry winters, during which frost is infrequent.

Table 1
List of outcrops selected as study sites with two additional sites studied previously (Williamson, 1994) and a summary of environmental conditions used for selection.

Serpentine site	Approx. area (km^2)	Altitude range (m.a.s.l)	Mean annual rainfall (mm)	Reason for selection of site
CoreZone (CZ)	9.3	780–1189	800–950	Furtherest north
Kalkkloof (KK)	2.0	1176–1300	650–800	Furtherest west and 3rd highest
Magnesite Mine (MM)	13.7	354–575	650–800	Furtherest east and lowest altitude
Groenvaly (GV)	18.7	1000–1540	800–950	Largest
Mundt's Concession (MC)	5.8	620–889	800–950	Intermediate area and altitude
Sawmill (SM)	6.7	900–1192	950–1100	Intermediate area and altitude
Rosentuin (RT)	0.3	1200–1400	950–1100	3rd smallest
Agnes Mine (AM)	9.7	900–1100	950–1100	
Kaapsehoop (KH)	7.6	1430–1580	800–950	

Table 2
Comparison of the number of taxa in different categories represented in the flora of the serpentine outcrops of the Barberton Greenstone Belt.

	Numbers of taxa for each serpentine site							Serpentine sites combined
	CZ	GV	KK	MM	MC	RT	SM	
Families	46	61	41	48	47	45	42	94
Genera	117	185	120	122	142	126	111	319
Species and infraspecific taxa	162	285	182	179	206	184	151	744
Species to genus ratio	1.38	1.54	1.52	1.47	1.45	1.46	1.36	2.33
Species per family	3.52	4.67	4.43	3.73	4.38	4.09	3.60	7.91
Unique species (% of total)	33 (20)	101 (35)	31 (17)	65 (36)	51 (25)	32 (17)	24 (16)	

A 'Resolution' was passed by the delegates of The First International Conference on Serpentine Ecology held in 1991 supporting the conservation of the vegetation of serpentine areas worldwide (Kruckeberg, 1992). Subsequent to this resolution a few publications providing evidence for the need for the conservation of serpentine vegetation have been published (Wolf, 2001; Selvi, 2007). The ability of metallophytes to tolerate extreme metal concentrations commends them for revegetation of mines and metal-contaminated sites and can be exploited in environmental technologies, such as phytostabilisation, phytoremediation and phytomining (Whiting et al., 2004). Conservation of biodiversity is the main objective of most conservation organisations (Brooks et al., 2006) and thus if diversity of serpentine vegetation is conserved then metallophytes and other rare and/or restricted range (endemic) species are also conserved. Serpentine outcrops usually support many rare and endemic species, which are often threatened by a variety of activities and are in need of conservation (Wolf, 2001). However, very few accounts of the actual conservation of serpentine vegetation exist suggesting that such sites continue to be severely under-conserved. The serpentine vegetation of the Barberton Greenstone Belt is threatened by mining activities, commercial plantations of species of *Eucalyptus* and *Pinus* and by urban development.

This paper aims to characterise and describe the plant species composition of serpentine sites in the Barberton Greenstone Belt as compared to surrounding non-serpentine areas. This allows the contribution of the serpentine flora to the overall diversity and endemicity of the Barberton Centre of Endemism to be quantified and the conservation value of the area to be determined in terms of taxon richness at different taxonomic ranks. The plant list presented for the selected serpentine outcrops of the Barberton Greenstone Belt is intended for use in land management, conservation planning and rehabilitation of disturbed serpentine landscapes.

An analysis of the rarity or commonness of taxa on serpentine outcrops compared with their occurrence in the Mpumalanga Province or the Barberton Centre of Endemism will assist in identifying areas or sites that would maximise the conservation of plant diversity on serpentine outcrops. It was predicted that each serpentine outcrop of the Barberton Greenstone Belt is unique in its species composition and a large number of serpentine sites need to be protected in order to adequately conserve the area.

2. Materials and methods

The serpentine outcrops of the Barberton Greenstone Belt have been visited regularly from 1991 and during these visits extensive plant collections were made. In addition taxon data were collected using Modified-Whittaker plots positioned on each serpentine site and adjacent non-serpentine area. A detailed description of the positioning and layout of these plots is given in Williamson and Balkwill (2013).

Field surveys focused on seven outcrops selected in such a way that the full range of variation is presumably accounted for (Table 1). Fig. 1 indicates the location of all the serpentine sites in the Barberton Greenstone Belt and shows the sites studied in detail.

Distribution data for all taxa recorded were obtained from: PRECIS (Pretoria National Herbarium Computerised Information System) a computerised data bank managed by the South African National Biodiversity Institute (SANBI); taxonomic accounts, monographs, herbarium collections and distributional data collected by scientists of the Mpumalanga Tourism and Parks Agency (MTPA). This was done to identify the taxa endemic to the serpentine of the Barberton Greenstone Belt. All specimens collected by the authors were deposited at the C.E.Moss Herbarium of the University of the Witwatersrand, Johannesburg and duplicate specimens were forwarded to various herbaria in South Africa and abroad, as annotated on individual specimen labels. The plant lists produced include sterile specimens that were positively identified, but for which no herbarium specimen was kept. These specimens were added to the plant list as they represent the only record of these taxa from these sites. A number of sterile specimens that could not be identified were noted but not added to the plant list. Taxa on the plant list that are restricted to the area within which the Barberton Greenstone Belt is found and that occur more commonly on serpentine soils than on non-serpentine soils were identified as possible local or regional serpentine indicators as described by Kruckeberg (1984). In addition, taxa that occur commonly in non-serpentine plots but do not appear on the serpentine plant lists were identified to be possible excluded taxa. This list was refined by removing any plants that occur on species lists compiled for other serpentine sites of the Barberton Greenstone Belt (McCallum, 2006; Changwe and Balkwill, 2003; Balkwill and Balkwill, 1999; Kidger, 1993; Williamson, 1994).

Table 3
Comparison of the number of monocotyledonous (M) and dicotyledonous (D) taxa collected at each site to rest of Mpumalanga.

	No of species of each monocotyledons and dicotyledons and percentage of total no of species in parentheses								
	CZ	GR	KK	MM	MC	RT	SM	Serpentine sites combined	Mpumalanga Province[a]
Fern and Fern Allies	1(<1)	2(<1)	2(1)	3(1)	5(2)	3(2)	1(<1)	7 (<1)	172 (4.2)
Monocotyledons	24	65	44	39	49	41	37	155	1194
	(15)	(23)	(24)	(22)	(24)	(22)	(24)	(21)	(29)
Dicotyledons	137	218	136	138	152	140	113	578	2766
	(85)	(77)	(75)	(77)	(74)	(76)	(75)	(78)	(67)
Ratio M:D	0.18	0.30	0.32	0.28	0.32	0.29	0.33	0.27	0.43
Total	162	285	182	179	206	184	151	740	4133

[a] SANBI (2013).

Table 4
Comparison of ratios of monocotyledons (M) and dicotyledons (D) for serpentine and adjacent non-serpentine areas from data collected in Modified Whittaker plots.

Site	Serpentine			Non-serpentine		
	Monocots	Dicots	Ratio M:D	Monocots	Dicots	Ratio M:D
SM	43	110	0.39	39	121	0.32
MM	42	120	0.35	31	114	0.27
KK	48	125	0.38	47	123	0.37
CZ	26	114	0.23	33	145	0.23
GV	45	117	0.38	41	112	0.37
RT	39	122	0.32	34	122	0.28
MC	37	117	0.32	32	134	0.24
Serpentine sites combined	194	552	0.35	165	569	0.29
Mpumalanga Province (including serpentine outcrops)[a]				1194	2766	0.43

[a] SANBI (2013)

About 85% of the collected specimens have been identified to species or infraspecific level; however, some specimens remain unidentified (3%) or need further consultation and/or analyses to confirm identifications (12%). All endemic species and those with relatively restricted distributions on and around serpentine outcrops were subjected to a leaf sap test, in which sap was applied to dimethylglyoxime-impregnated filter paper (1% in ethanol). The test gives a distinct dark pink reaction to leaf sap containing high levels of nickel.

The South African National Botanical Institute's online database (SANBI, 2013) was used to determine the number of taxa recorded for Mpumalanga Province and their familial classification system was adopted when compiling the checklists for each serpentine outcrop (Appendix 1).

Proportions of Fern and Fern allies, monocotyledons and dicotyledons recorded in the plant list for each site (Appendix 1) were compared to the other sites and to the vegetation of Mpumalanga Province as a whole. The ratio of dicotyledons to monocotyledons was calculated for the taxa recorded within the Modified-Whittaker plots at each site and these data were compared to those of the adjacent non-serpentine sites by way of a Student's T-test. The relative proportions of monocotyledons and dicotyledons, calculated by combining the data from all the Modified-Whittaker plots placed on serpentine outcrops and on adjacent non-serpentine areas, were compared using a Chi square test to determine if differences between these proportions are significant.

Species compositions of the serpentine sites were compared by ranking the twenty most diverse families at each site. Site-specific species compositions were compared to the flora of Mpumalanga Province to highlight any deviation in floristic composition at the family level from the regional flora. Spearman rank correlation coefficient tests (Sokal and Rohlf, 1995) were performed to test for significance of correlations in family rankings between pairs of sites. The null hypothesis was that the familial ranking of one site did not covary with the familial ranking of another site. A correlation analysis was conducted on the ranks in a pairwise manner between all combinations of the sites and a correlation coefficient (Spearman's rho) is calculated for each pair of ranks. A value of r (rho) $= 1$ indicates a perfect positive correlation and the value of $r = -1$ indicates a perfect negative correlation. The significance of this coefficient is calculated by determining a P-value for the correlation.

Two additional sites, Agnes Mine (AM) and Kaapsehoop (KH) (Williamson, 1994) were included in this analysis to broaden the basic data. Clustering by the unweighted pair group method, arithmetic average (UPGMA: http://genomes.urv.es/UPGMA/) was used to create a phenogram indicating the relative similarities of the floras of the individual sites.

3. Results and discussion

3.1. Floristic analysis

The plant list (Appendix 1) provides an alphabetical listing of species by family. The number of serpentine-tolerant taxa recorded within the selected sites includes a total of 744 species and infraspecific taxa in 319 genera within 94 families (Table 2). This plant list includes 513 herbaceous plants (70%), 125 small- to medium-sized shrubs (17%), 39 large shrubs or small trees (5%), 35 trees (5%) and 21 climbers (3%). It is noteworthy that the percentage of tree species in the entire Mpumalanga Province at <1% (calculated from the plant list downloaded from SANBI, 2013) is 4% lower than that recorded for the serpentine outcrops. This is unexpected as serpentine vegetation in this area is known for its paucity of trees and shrubs; however the percentages of small shrubs and large shrubs/small trees recorded from the serpentine vegetation are 6% and 3% respectively lower than those

Table 5
List of the highest ranked families and the numbers of species in each family, recorded for each serpentine outcrop, compared to those of the serpentine outcrops combined and to the Mpumalanga Province.

Families	Serpentine outcrops									Combined serpentine sites	Mpumalanga Province
	GV	MC	RT	SM	CZ	KK	MM	AM	KH		
Asteraceae	66	21	26	30	22	40	14	25	28	111	311
Fabaceae	28	28	25	15	21	23	24	15	11	87	392
Poaceae	32	29	23	24	13	20	22	20	21	70	355
Acanthaceae	11	15	7	10	7	8	13	8	7	36	119
Euphorbiaceae	12	8	6	4	5	5	7	5	3	30	85
Lamiaceae	8	11	7	8	6	7	1	9	3	27	131
Malvaceae	6	7	7	6	9	7	1	3	10	27	129
Anacardiaceae	7	7	7	3	8	3	4	5	2	25	47
Rubiaceae	8	4	9	3	1	9	2	8	10	20	110
Apocynaceae	7	4	7	4	3	4	5	7	1	20	190
Cyperaceae	7	7	3	2	1	6	2	8	6	18	169
Convolvulaceae	3	4	3	2	5	2	8	5	3	17	46
Vitaceae	2	4	1	1	4	0	5	2	1	14	26
Hyacinthaceae	7	1	2	1	0	5	2	7	5	12	62
Scrophulariaceae	6	2	3	2	2	1	0	3	6	11	85
Polygalaceae	4	2	3	1	3	1	3	4	3	10	31
Ebenaceae	3	3	3	2	2	2	2	1	1	8	19
Santalaceae	1	3	1	2	2	1	1	0	1	8	32
Celastraceae	3	2	3	1	3	0	3	1	1	8	33
Asphodelaceae	4	2	2	1	1	2	2	3	2	8	102

Table 6
Spearman rank correlation coefficients test (ρ) of the rank of the twenty most diverse families recorded at each serpentine site compared to the combined serpentine flora and to the flora of the Mpumalanga Province. Correlations that are not significant (P > 0.01) are underlined.

	MC	RT	SM	CZ	KK	MM	AM[a]	KH[a]	Combined serpentine sites	Mpumalanga Province
GV	0.738	0.830	0.798	0.704	0.864	0.488	0.866	0.753	0.874	0.811
MC		0.772	0.897	0.767	0.793	0.530	0.680	0.568	0.915	0.691
RT			0.886	0.864	0.823	0.473	0.713	0.677	0.849	0.752
SM				0.795	0.794	0.430	0.700	0.634	0.899	0.768
CZ					0.795	0.522	0.621	0.659	0.854	0.539
KK						0.430	0.804	0.766	0.865	0.806
MM							0.350	0.363	0.576	0.333
AM								0.714	0.766	0.798
KH									0.715	0.705
Combined serpentine sites										0.764

[a] Williamson (1994).

for the Mpumalanga Province. Although the serpentine vegetation supports a relatively high number of tree species, these trees are present in low numbers on the serpentine and individuals are often small and/or stunted.

The species to genus and species to family ratios (2.33 and 7.91 respectively) for the serpentine sites combined are considerably higher than those for each of the individual serpentine outcrops (Table 2). This suggests a family tolerance of the serpentine condition and that there are different members of families and genera on different outcrops contributing to the relative uniqueness of each outcrop.

Less than 1% of the taxa recorded on the serpentine outcrops are Fern and Fern allies as compared to the rest of Mpumalanga Province with 4.2% of the taxa being Ferns (Table 3). The percentage of monocotyledonous taxa is also lower (21%) than the rest of the province (29%). The ratios of the number of monocotyledons to dicotyledons (Table 4) recorded from the plots placed in serpentine vegetation were found to be statistically different ($P < 0.02$) from the ratio recorded from the adjacent non-serpentine vegetation. The ratios calculated from combining the species lists from all the sampled sites showed a significantly ($P < 0.001$) higher monocotyledon to dicotyledon ratio on serpentine (0.35) than on the adjacent non-serpentine vegetation (0.29). The serpentine condition leads to an environment with reduced moisture levels and one would expect that monocotyledons with their well developed fibrous root systems are better able to access soil moisture than plants with tap root systems. In addition plants with fibrous root systems are naturally replacing their roots at a high rate and will thus be less affected by the toxic levels of nickel in the soil (Brady et al., 2005). It is thus surprising that the number of monocotyledons relative to dicotyledons is lower than in the province as a whole.

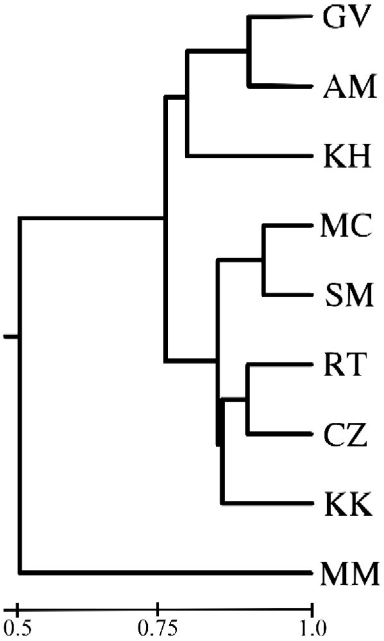

Fig. 2. Dendrogram representing the Spearman rank correlation coefficients of similarities in ranks of important families representing the floras of the sampled serpentine sites of the Barberton Greenstone Belt and the Mpumalanga Province. This was drawn online (http://genomes.urv.es/UPGMA/) using the UPGMA algorithm. Values used are presented in Table 6 and abbreviations are expanded in Table 1.

The five most important plant families represented in the vegetation of the serpentine outcrops of the Barberton Greenstone Belt are presented in Table 5. The comparison to the entire Mpumalanga Province demonstrates the distinctiveness of the serpentine flora. The list of major families shows the decreased representation of the Apocynaceae and Cyperaceae and the increased representation of the Asteraceae on serpentine soils. The Rubiaceae and Euphorbiaceae also show an increased representation on some individual outcrops. The serpentine vegetation has a third of the Asteraceae taxa found in the entire province; however, the serpentine outcrops represent only 0.1% of the area of the province. Batianoff et al. (2000) suggest that such a high representation may be explained by a family tolerance of soil conditions such as high Mg, commonly associated with serpentine soils and that this innate tolerance facilitates prominence of these families on serpentine outcrops. This is borne out by all the nickel accumulators on the Barberton Greenstone Belt being members of the Asteraceae.

Spearman rank correlation coefficients for all pair wise comparisons of ranking of the twenty most diverse families of the sampled serpentine sites of the Barberton Greenstone Belt are listed in Table 6. Lower, non-significant correlations were found with all comparisons with Magnesite Mine (MM) and between CoreZone (CZ) and the flora of Mpumalanga Province (0.539). The UPGMA dendrogram (Fig. 2) shows that Mundt's Concession (MC) and Sawmill (SM) sites are most similar to one another. The Groenvaly (GV) and Agnes Mine (AM) sites show a high level of similarity in their familial diversity. The Rosentuin (RT) and CoreZone (CZ) show higher levels of similarity with each other than with the flora of Mpumalanga Province. The flora of the Magnesite Mine (MM) site shows the least similarity to any of the other sites and also a low degree of similarity with the flora of the Mpumalanga Province. There is no substantial difference in the soil chemistry (Table 7) between the Magnesite Mine site and the other sites. This suggests that the difference in the vegetation could be due to the lower rainfall of the area in which the Magnesite Mine occurs (Table 1), which, possibly results in an exacerbation of the 'serpentine condition'.

The large difference in the total number of taxa from combined lists and individual sample site lists suggests that the composition of taxa on each serpentine outcrop is near-unique and supports a large number of taxa found at one site only. For instance, 35% of the flora on the Groenvaly (GV) outcrop and 36% of that on the Magnesite Mine (MM) outcrop are found on only those sites and not on the others sampled (Table 2). The Sørenson's coefficient of similarity (Table 8), which measures the degree of similarity between sites indicates that less than 26%

Table 7
Mean concentrations of heavy metals, calcium to magnesium ratios and pH of soils sampled from seven outcrops of the Barberton Greenstone Belt. Ranges are given in parentheses (n = 3).

Site	Zn (mg/kg)	Ni (mg/kg)	Cr (mg/kg)	Ca:Mg	pH
Mundts Concession (MC)	0.53 (0.4–0.8)	2689 (1480–4286)	1875 (920–3346)	1.1 (0.87–1.46)	5.68 (5.5–5.82)
Magnesite Mine (MM)	0.53 (0.2–1.1)	543 (1420–800)	1040 (138–1720)	2.5 (1.31–4.09)	6.22 (5.75–6.55)
Kalkkloof (KK)	0.97 (0.5–1.0)	134 (40–214)	544 (330–920)	1.9 (1.22–2.5)	4.95 (4.55–5.65)
Sawmill (SM)	0.85 (0.6–1.1)	2184 (1520–2848)	3128 (2160–4096)	1.0 (0.92–1.07)	5.50 (5.35–5.65)
Rosentuin (RT)	0.70 (0.2–1.1)	651 (370–908)	1006 (500–1738)	2.8 (1.28–4.21)	4.93 (4.85–4.93)
Groenvaly (GV)	1.25 (0.9–1.6)	1288 (344–2242)	2615 (782–4760)	1.4 (0.98–1.91)	5.26 (5.15–5.35)
CoreZone (CZ)	2.7 (0.8–4.6)	510 (485–534)	1463 (1362–1563)	1.7 (1.51–1.79)	6.20 (6.15–6.25)

of the plant taxa recorded are shared between most sites. Two sites, namely the Groenvaly (GV) and Rosentuin (RT) sites are most similar in terms of species composition (35%), which could be explained by the relative proximity of the sites (Fig. 1). The Sawmill (SM) site and the Groenvaly site, which are also relatively close together, share 34% of their plant taxa. The three outlying outcrops i.e. Magnesite Mine (MM), Kalkloof (KK) and CoreZone (CZ) share the lowest number of taxa with the other outcrops. These three outcrops also have the lowest soil nickel concentrations measured (i.e. 0.54, 0.13, 0.51% of metal in air dried soil, respectively). A possible source of error includes the oversampling of common and widespread taxa compared with uncommon, endemic or patchily distributed species. Thus with more sampling the number of taxa shared between sites would decrease.

18 taxa have been found to be undescribed and are listed in the checklists as sp. nov. or subsp. nov. This would suggest that the area is historically under-collected and although the area is considered to be relatively high in diversity, this level may be underestimated. Many of these taxa could be found to be taxa endemic to the Barberton area or to Mpumalanga Province increasing the conservation value of the area.

While the family composition of the flora of the Barberton Greenstone Belt shows some variation from the flora of the surrounding areas (Table 5), this difference is not seen in the flora of the greenstone outcrops of Western Australia. Here the family composition is very similar to and typical of the flora of the South Western Interzone (Gibson and Lyons, 1998a). However, the relative uniqueness of the flora of each group of outcrops of the Western Australian greenstones is similar to that of the Barberton Greenstone Belt with the greenstone of the Bremer Range and the Parker Range only sharing 32% of the recorded flora (Gibson and Lyons, 1998b). The flora of the Central Queensland serpentine has a higher representation of Fabaceae (narrowly defined) (5%), Mimosaceae (2%) and Rubiaceae (1.5%) compared to the Port Curtis District flora (Batianoff et al., 2000). It is postulated that these families are characterised by a higher proportion of serpentine-tolerant species which find the serpentine soils adequate and/or neutral for establishment, growth and reproduction, thus facilitating their relative expansion.

Table 8
Sørenson's coefficient of similarity measuring the degree of similarity of species between sampled serpentine sites, expressed as percentages. Values in italics represent the lowest similarity calculated and the value in bold indicates the highest similarity.

	CZ	GV	KK	MM	MC	RT
GV	17.7	–				
KK	18.3	25.4	–			
MM	*14.0*	18.9	9.4	–		
MC	25.4	27.5	15.7	21.1	–	
RT	17.0	**35.0**	24.9	15.9	22.5	–
SM	16.2	34.1	17.0	*14.0*	21.1	23.3

3.2. Commonness and rarity

The 20 most frequently occurring plants on the serpentine of the Barberton Greenstone Belt are listed in Table 9 and are compared to those of the adjacent non-serpentine areas sampled. The serpentine vegetation and the surrounding non-serpentine vegetation are dominated by grasses such as *Themeda triandra* Forssk., *Heteropogon contortus* (L.) Roem. & Schult. and *Loudetia simplex* (Nees) C.E.Hubb. However, grasses such as *Cymbopogon caesius* (Hook. & Arn.) Stapf, *Tristachya leucothrix* Trin. ex Nees and *Trachypogon spicatus* Kuntze appear to become more common on serpentine than on non-serpentine. Due to their widespread distributions these taxa would not be considered to be indicator species (Kruckeberg, 1986). Grasses such as *Cymbopogon pospischilii* (K. Schum.) C.E.Hubb. and *Hyparrhenia filipendula* (Krauss) Stapf, which are relatively common throughout South Africa, were not recorded on any of the sampled serpentine outcrops.

The tree species *Dichrostachys cinerea* (L.) Wight & Arn. and herbaceous *Oxalis obliquifolia* Steud. ex A.Rich. were found to be relatively common on non-toxic soils but occasional to rare on nearby serpentine outcrops (Table 9). A further 53 species representing 47 genera and 29 families (Appendix 2) are possible 'excluded' taxa as they were not recorded on serpentine sites but were recorded from the Modified-Whittaker plots placed on adjacent non-serpentine areas. Twenty genera and two families (Kirkiaceae and Strychnaceae) are absent from the serpentine flora. Of the 60 dicotyledonous species excluded, 55% are woody species (shrubs, trees or woody climbers), many of which are common in the surrounding areas.

All of the serpentine endemics (Table 11) are considered to be rare because of their restricted distributions on the serpentine outcrops of the Barberton Greenstone Belt. However, 17 taxa are found on only five or fewer outcrops and on those outcrops have small and sparse populations, increasing their measure of rarity.

3.3. Conservation importance

Eighteen of the taxa collected from the seven serpentine outcrops of the Barberton Greenstone Belt, included in this study, have been listed on the South African National Biodiversity Institute Red Data list as either threatened with extinction or of conservation concern (SANBI, 2013). An additional seven taxa are listed on the Mpumalanga Tourism and Parks Agency's list of threatened plants for the province (Lötter pers. comm.[1]). Seven of these taxa are listed as vulnerable, three as near threatened, four as declining and four as rare.

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Table 9
The top 20 taxa ranked according to the number of subplots, of the Modified Whittaker plots, within which they were recorded on the serpentine sites compared with those of the adjacent non-serpentine sites. Relative abundance scores are represented by 0 = absent, 1 = rare, 2 = uncommon, 3 = occasional, 4 = common, 5 = abundant and are given as a range as the relative abundance of individual taxa varies between each outcrop sampled.

Serpentine outcrops				Non-serpentine areas			
Species	Family	No of subplots out of 280	Relative abundance score	Species	Family	No of subplots out of 280	Relative abundance score
Themeda triandra	Poaceae	258	2–5	Themeda triandra	Poaceae	209	2–5
Heteropogon contortus	Poaceae	139	2–5	*Heteropogon contortus*	Poaceae	125	2–5
Cymbopogon caesius	Poaceae	134	0–5	*Loudetia simplex*	Poaceae	125	0–5
Loudetia simplex	Poaceae	100	0–5	*Bewsia biflora*	Poaceae	89	0–5
Panicum natalense	Poaceae	84	0–5	*Diheteropogon amplectens*	Poaceae	76	0–5
Tristachya leucothrix	Poaceae	83	0–5	*Panicum maximum*	Poaceae	74	0–5
Trachopogon spicatus	Poaceae	75	0–5	*Cymbopogon caesius*	Poaceae	67	0–4
Bewsia biflora	Poaceae	72	0–5	*Hilliardiella aristata*	Asteraceae	63	0–3
Diheteropogon amplectens	Poaceae	70	0–5	*Cymbopogon pospischilii*	Poaceae	59	0–4
Kohautia amatymbica	Rubiaceae	67	0–4	*Senecio venosus*	Asteraceae	55	0–3
Ruellia cordata	Acanthaceae	60	0–4	*Setaria sphacelata*	Poaceae	53	0–4
Corchorus asplenifolius	Malvaceae	54	0–4	*Eragrostis racemosa*	Poaceae	47	0–4
Justicia anagalloides	Acanthaceae	53	0–3	*Pentanisia angustifolia*	Rubiaceae	47	0–3
Pearsonia sessilifolia	Fabaceae	49	0–4	*Panicum natalense*	Poaceae	45	0–3
Thunbergia atriplicifolia	Acanthaceae	47	0–4	*Oxalis obliquefolia*	Oxalidaceae	45	0–3
Hilliardiella aristata	Asteraceae	47	0–3	*Schizachyrium sanguineum*	Poaceae	45	0–4
Rhynchosia totta	Fabaceae	46	0–3	*Hyparrhenia filipendula*	Poaceae	43	0–4
Phyllanthus sp.	Phyllanthaceae	45	0–3	*Ruellia cordata*	Acanthaceae	43	0–3
Vernonia sutherlandii	Asteraceae	43	0–3	*Dicrostachys cinerea*	Fabaceae	41	0–3
Acalypha sp. nr *A. wilmsii*	Euphorbiaceae	41	0–3	*Digitaria diagonalis*	Poaceae	41	0–4

Eighteen taxa are considered to be new and undescribed species and some of these are further thought to be endemic to serpentine soils. Further taxonomic study and occurrence data are required to confirm these findings.

Only one of the sampled outcrops (Rosentuin) occurs within an officially proclaimed conservation area, the Songimvelo Nature Reserve. This is one of the smallest outcrops with only three of the more common endemic species. However, three additional sites (Groenvaly, Sawmill and CoreZone) occur in areas considered to be of high intrinsic biodiversity value and the others occur in areas considered to be of medium intrinsic biodiversity value (Emery et al., 2002).

3.4. Indicator and endemic taxa

Fig. 3 shows those taxa that have been more frequently recorded in serpentine plots than plots on adjacent non-serpentine areas. None of these taxa are local or regional indicators as most are relatively widespread through South Africa (Table 10). However, possible indicator species have been identified as those that have the serpentine outcrops as the edge or an extension of their 'natural' range (Kruckeberg, 1984) and are shown in Table 10. Of the 24 taxa that were recorded to be more common in plots placed on serpentine sites than on plots on adjacent non-serpentine sites, seven are considered to be indicator species

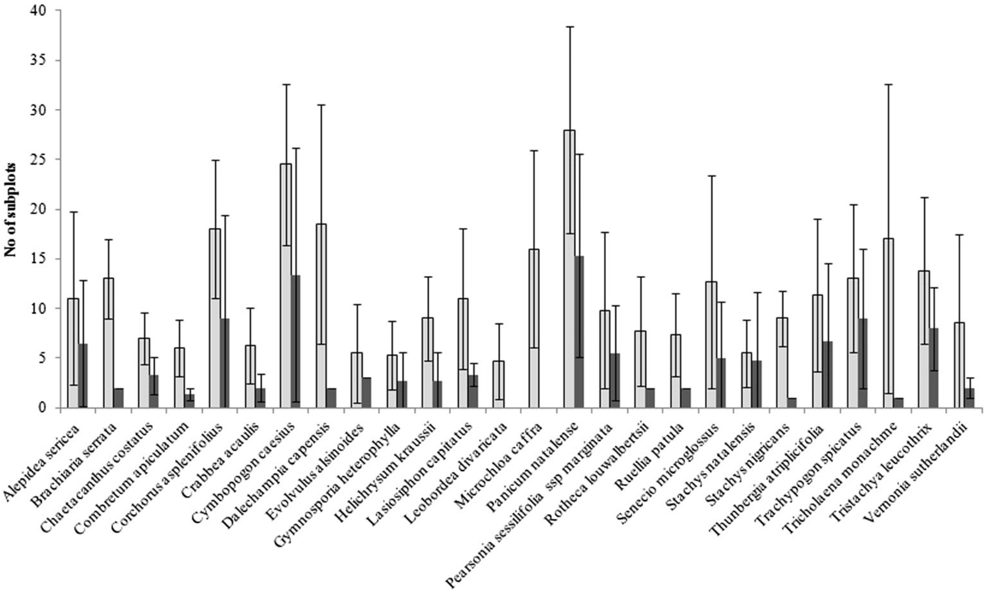

Fig. 3. Graph showing the mean number of sub-plots within which possible indicator species have been recorded on serpentine sites as well as non-serpentine sites. Error bars represent calculated standard deviations of the means.

Table 10
List of taxa occurring more commonly in serpentine plots than in non-serpentine plots with information on their distributions contributing to their selection as indicator species. Possible indicator taxa are indicated in bold font.

Family	Species	Distribution information
Acanthaceae	***Chaetacanthus costatus* Nees**	Widespread but serpentine is the eastern limit of the distribution
	***Crabbea acaulis* N.E.Br.**	Widespread but serpentine is the eastern limit of the distribution
	Ruellia patula Jacq.	Widespread through north-eastern parts of South Africa
	Thunbergia atriplicifolia E.Mey. ex Nees	Widespread and common through eastern parts of South Africa
Apiaceae	***Alepidea setifera* N.E.Br.**	Widespread but serpentine is the south-eastern limit of distribution
Asteraceae	*Helichrysum kraussii* Sch.Bip.	Widespread through eastern parts of South Africa
	***Senecio microglossus* DC.**	Mostly found in eastern Mpumalanga, serpentine is the eastern limit of edge of distribution
	***Vernonia sutherlandii* Harv.**	Widespread but serpentine is the eastern limit of the distribution
Celastraceae	*Gymnosporia heterophylla* (Eckl. & Zeyh.) Loes.	Common in the Barberton area and down to Kwa-Zulu Natal coast
Combretaceae	*Combretum apiculatum* Sond.	Widespread through north-eastern parts of South Africa
Convolvulaceae	*Evolvulus alsinoides* (L.) L.	Widespread through eastern parts of South Africa
Euphorbiaceae	*Dalechampia capensis* A. Spreng.	Widespread through eastern parts of South Africa
Fabaceae	***Leobordea divaricata* Eckl. & Zeyh**	Widespread through eastern parts of South Africa, serpentine is the eastern limit of the distribution.
	Pearsonia sessilifolia (Harv.) Dummer *subsp. marginata* (Schinz) Polhill	Common through Eastern Mpumalanga and into Swaziland
Lamiaceae	***Rotheca louwalbertsii* (P.P.J.Herman) P.P.J. Herman & Retief**	Widespread but serpentine is the southern boundary of the distribution
	Stachys natalensis (Hochst.)	Widespread through eastern parts of South Africa
	Stachys nigricans (Benth.)	Widespread through eastern Mpumalanga into Kwa-Zulu Natal
Malvaceae	*Corchorus asplenifolius* (Burch.)	Widespread through Mpumalanga into Kwa-Zulu Natal
Poaceae	*Brachiaria serrata* (Thunb.) Stapf	Widespread through South Africa
	Cymbopogon caesius (Hook. & Arn.) Stapf	Widespread through eastern parts of South Africa
	Microchloa caffra Nees	Widespread through South Africa
	Panicum natalense Hochst.	Widespread through Mpumalanga into Kwa-Zulu Natal
	Trachypogon spicatus (L.f.) Kuntze	Widespread and relatively common through South Africa
	Tricholaena monachme (Trin.) Stapf & C.E.Hubb.	Widespread through eastern parts of South Africa
	Tristachya leucothrix Trin. ex. Nees	Widespread through South Africa
Thymelaeaceae	*Lasiosiphon capitatus* (L.f.) Burtt Davy	Widespread through eastern parts of South Africa

based on their distributions. Only two taxa, *Leobordea divaricata* Eckl. & Zeyh and *Microchloa caffra* Nees were not recorded from non-serpentine plots but were common in serpentine plots and are widespread through Mpumalanga Province. The small number of indicators identified for the Barberton Greenstone Belt contrasts with the report of 200 indicators and 200 strict endemic species from the Californian serpentine (Kruckeberg, 1984). In contrast to the Barberton Greenstone Belt, Californian serpentine indicator species have more restricted and localised distributions around serpentine outcrops.

Thirty-two taxa are considered to be endemic to the serpentine outcrops of the Barberton Greenstone Belt (Table 11). Twenty-eight of these taxa have been found to only occur on soils derived from serpentinite rocks. The other four taxa have also been found on ultramafics of the Onverwacht group, which show very similar patterns of soil chemistry and toxicity as the serpentine outcrops (Balkwill et al., 2011). Three of these endemic taxa, *Helichrysum* sp. nov. aff. *H. nudifolium* (L.) Less, *Kniphofia umbrina* Codd and *Ocimum motjaneanum* McCallum & K.Balkwill, are found only on the Swaziland serpentine outcrops. Many of these endemic taxa are threatened due to their small population size and by extensive afforestation (Williamson and Balkwill, 2006). Van Wyk and Smith (2001) listed 56 plant species as being endemic or near endemic to the Barberton Centre of Endemism. This list includes 13 taxa, which are considered to be serpentine endemics i.e. 23% of species listed as endemic to the Barberton Centre of Endemism are from serpentine outcrops. Adding undescribed taxa and four other species known to be serpentine endemics, which are not included in Van Wyk and Smith's (2001) list, would increase the contribution of serpentine endemics to 39% of the Barberton Centre of Endemism.

By way of comparison, very few endemics have been recorded from the ultramafic outcrops of Western Australia (Gibson and Lyons, 1998a, b). A hypothesis suggests that this lack of endemic species is due to the lack of soil chemistry imbalances seen on other ultramafic soils, which is probably as a result of a long period of weathering of the ancient soils or due to differences in the parent material of the soils. The serpentine soils of Barberton Greenstone Belt possibly support this hypothesis as the outcrops are relatively ancient yet exhibit the high levels of nickel, chromium and magnesium found in other serpentine soils.

Thirteen endemics have been reported from the Central Queensland serpentine outcrop (Batianoff et al., 2000) and these endemics occur from a range of families. These families differ markedly from the families within which the Barberton Greenstone Belt endemics are found. Two nickel accumulators discovered on the Central Queensland serpentine are from different families to those from the Barberton Greenstone Belt.

The floras of Brazil and Cuba support large numbers of endemics and hyperaccumulators (Reeves et al., 1999, 2007), yet very few of these are found within the Asteraceae. This is in contrast to the Barberton Greenstone Belt where the five hyperaccumulators are all from the Asteraceae.

4. Conclusion

This study expands on the understanding of the floristics of the Barberton Greenstone Belt in Mpumalanga, South Africa. Comparing the serpentine vegetation to the surrounding non-serpentine vegetation and quantifying their differences ultimately makes a case for including serpentine vegetation in the planning for conservation of the vegetation of the eastern portion of Mpumalanga Province.

It is predicted that the unique soil chemistry could render the serpentine outcrops floristically distinguishable from the surrounding non-serpentine vegetation. This study has shown that serpentine outcrops support higher relative numbers of dicotyledons and also larger average numbers of species per family. The serpentine vegetation is relatively rich in tree species, but these species are found in low numbers, when compared to those of the surrounding non-serpentine vegetation. The floristic composition of each serpentine outcrop is near-unique, with only 30 to 35% of taxa being shared between outcrops. In order to adequately conserve the plant diversity of the Barberton Greenstone

Table 11
List of taxa considered to be endemic to the Barberton Greenstone Belt, including those found on outcrops in Swaziland.

Family	Taxon	Nickel hyperaccumulator
Asteraceae	*Athrixia* sp. nov.	
	Berkheya coddii Roessler	✓
	Berkheya nivea N.E.Br.	✓
	Berkheya rehmannii Thell.var. *rogersiana* Thell.	✓
	Berkheya sp. nov. aff. *B. seminivea* Harv. & Sond.	
	Dicoma swazilandica S.Ortiz	
	Helichrysum sp. nov. aff. *H. nudifolium* (L.) Less.[a]	
	Helichrysum sp. nov. aff. *H. albo-brunneum* S.Moore	
	Macledium zeyheri subsp. *thyrsifolium* (Klatt) Netnou	
	Senecio sp. aff. *S. anomalochrous* Hilliard	✓
	Senecio sp. aff. *S. coronatus* (Thunb.) Harv.[b]	✓
Acanthaceae	*Salpinctium hirsutum* T.J.Edwards	
	Asystasia subbiflora C.B.Clarke	
	Sclerochiton triacanthus A.Meeuse	
Anacardiaceae	*Searsia pygmaea* (Moffett) Moffett	
	Ozoroa barbertonensis Retief	
	Ozoroa sp. nov.	
Lamiaceae	*Ocimum motjaneanum* McCallum & K.Balkwill[a]	
	Ocimum sp. nov. 1 (Barberton species)	
	Ocimum sp. nov. 2	
Apocynaceae	*Brachystelma dyeri* K.& M.Balkwill	
	Brachystelma sp. nov.	
Asphodelaceae	*Kniphofia umbrina* Codd[a]	
	Aloe thorncroftii Pole-Evans	
Fabaceae	*Indigofera crebra* N.E.Br.	
	Senegalia loetteri N.Hahn	
Celastraceae	*Gymnosporia* sp. nov.	
Iridaceae	*Gladiolus serpenticola* Goldblatt & J.C.Manning	
Lobeliaceae	*Cyphia bolusii* E.Phillips	
Orobanchaceae	*Graderia* sp. nov.[b]	
Poaceae	*Sartidia dewinteri* J.Munday & L.Fish	
Proteaceae	*Protea curvata* N.E.Br.	

[a] Endemic taxa from the outcrops of the Barberton Greenstone Belt within Swaziland (McCallum, 2006).

[b] Further research is needed to confirm its status as an endemic.

Belt a large number of outcrops would need to be conserved. The Barberton Greenstone Belt flora contributes substantially to the Barberton Centre of Endemism (BCE) as the 32 serpentine endemic taxa represent 39% of the endemic taxa included in the BCE. Five taxa, all from the family Asteraceae, found in the Barberton Greenstone Belt flora are considered to be hyperaccumulators of nickel. The Asteraceae are also known to harbour few hyperaccumulators in Cuba and none in Brazil, New Caledonia and Central Queensland.

Many hypotheses aim to identify the edaphic factor or combination of factors that have provided the evolutionary drive that has resulted in the unique serpentine vegetation and high numbers of endemics and hyperaccumulators. None of the hypotheses have been tested for the vegetation of the Barberton Greenstone Belt, and thus much research is still needed to determine the factors resulting in this unusual flora.

The floristic analyses presented here, species richness and diversity values calculated previously (Williamson and Balkwill, 2013), together with levels of endemism to be published, form part of a larger study which aims at determining the level of conservation needed in order to protect the diversity, endemics and unique flora of the Barberton Greenstone Belt.

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Chapter 3

Endemism of the serpentine soils derived from serpentinite outcrops of the Barberton Greenstone Belt.

Introduction

Ultramafic outcrops, especially those containing the mineral serpentinite, are often referred to as edaphic islands due to their sharp boundaries and patchy distribution. The extreme physical and chemical properties of serpentine soils provide conditions that allow colonisation by tolerant species and then strong diversifying selection processes may lead to ecological speciation (Kruckeberg 1986, Rajakaruna 2004). Plants endemic to soils derived from serpentinite are found in many parts of the world, but Kruckeberg and Rabinowitz (1985) specifically refer to two areas in the tropics that are noted for their high incidences of endemics to serpentine. In New Caledonia two monotypic families, more than 30 genera and 900 species (60% of the flora of the island) are restricted to serpentine outcrops. The second area is the Great Dyke of Zimbabwe, which has at least 20 species restricted to serpentine. Several of these taxa are known to be accumulators of heavy metals in unusually high concentrations (>1000 ppm). In addition, the serpentine outcrops of Cuba are considered to be the second richest in the world, supporting 920 endemic taxa (Brooks 1987).

Current theory of endemism is based on Cain's (1944) notion that a high degree of endemism is usually correlated with age and isolation of an area, and with the diversification of its habitats. These factors influence both evolution (the formation of new endemics) and survival (the persistence of relic endemics). Recent reviews agree that there are multiple causes of rarity and endemism (Kruckeberg and Rabinowitz 1985). An analysis of molecular phylogenies for 23 genera containing serpentine endemics in California (Anacker *et al*. 2010) suggests that serpentine endemics are often derived from speciation within populations of serpentine tolerant ancestors rather than from the splitting of already endemic lineages. These results confirm Kruckeberg's (1985, 1986) description of the process that may lead to the establishment of an edaphically endemic species, which suggests that some populations may be pre-adapted for serpentine tolerance. Disruptive selection, catastrophic selection or gradual divergence may effectively separate the species into serpentine-tolerant and intolerant gene pools. Further genetic divergence could lead to the isolation between tolerant and non-tolerant populations becoming fixed and the two populations being unable to exchange genes. Further divergence of the serpentine ecotype leads to an edaphically endemic species. Therefore, general biological factors that can increase endemism include: life-form and life-cycle, genetic processes, pollination, production of propagules and dispersal mode (Bruchmann and Hobohm 2014). An alternative pathway could be the extinction of all non-serpentine populations (Anacker *et al*. 2010).

Narrow endemic taxa are considered to be those that occur in one or a few small populations and hence are confined to a domain or a few localities (Kruckeberg and Rabinowitz 1985). Palaeoendemics are thought to be ancient vestiges of taxa that were once more widespread. Their present relictual status is presumably the result of increasing constriction of their specialised habitats over time. Therefore, palaeoendemics often have more than one disjunct population (Stebbins and Major 1965). Neoendemics are recent in origin and have recently split off from a parental entity, and may be poised for a further expansion of their ranges and gene pools (Kruckeberg and Rabinowitz 1985). Endemics confined to a single population can be either palaeoendemic or neoendemic (Stebbins and Major 1965). To determine whether a taxon is a neoendemic or a palaeoendemic, Ferreira and Boldrini (2011) suggest that phylogenetic and geological data should be used. If phylogenetic data are unavailable distributional data and morphological characteristics of the restricted taxon should be used to infer whether it diverged early or late within its taxonomic group.

Southern Africa has a few, relatively small localised areas with a high occurrence of endemic species. These areas would be considered regions or centres of endemism. These centres hold clues to the origin and evolution of the botanical diversity within a particular area and if these areas are conserved, would safeguard the greatest number of plant species (Van Wyk and Smith 2001). Regional Centres of Endemism, as defined by White (1983) have at least 50% of their species confined to them, as well as having more than 1000 endemic species. Centres of Endemism are identified by the WWF/IUCN to be species rich and/or known to contain large numbers of endemic species. Two Regions and three Centres of Endemism were recognised in the Mpumalanga Province by Van Wyk and Smith (2001). An additional one for the Lydenburg area was proposed by Lötter *et al.* (2002). The Barberton Greenstone Belt falls within the Barberton Centre of endemism, which has an area of about 4000 km^2, approximately 2210 species and more than 80 endemics, which represents 3.6% endemism. A large percentage (>29%) of this area is transformed by *Pinus* sp. and *Eucalyptus* sp. plantations (Lötter *et al.* 2002), threatening many of the serpentine endemics (Williamson and Balkwill 2006). It is known that rare plants of specialised climax habitats are more likely to go extinct if the habitats are narrowly limited in extent (Mahler 1983). Anacker *et al.* (2010) confirmed that if habitat patches are small, isolated, rare, or of poor quality, then small populations with low genetic variation and restricted geographic ranges could face increased extinction risk.

Although the serpentine floras around the world have been relatively well studied, the patterns and determinants of endemism of the Barberton Greenstone Belt have been poorly examined. Studies on the flora of serpentine outcrops in California have shown that endemism and endemic diversity can be determined by climatic conditions such as precipitation and that surface area of an outcrop is a significant predictor of the diversity of endemic plants (Harrison *et al.* 2000). It was also determined for the serpentine endemics of California that endemism richness is increased with increasing isolation of outcrops (Harrison *et al.* 2000). For conservation management, it is important to know whether or not an endemic flora constitutes a random assemblage with respect to taxonomy, habitat preference and biological attributes. If not, then the distinct characteristics of the endemic flora should be used as a guide for management (Willis *et al.* 1996).

Thirty-three plant taxa have been identified as endemic to the serpentine outcrops of the Barberton Greenstone Belt (Williamson and Balkwill 2015). This chapter aims to characterise and describe the endemism of these serpentine sites and to compare the level of endemism on serpentine sites to that of the Barberton Centre of Endemism. We predict that the levels of endemism on serpentine sites in the Barberton Greenstone Belt will increase with an increase in range of altitude, in size of area and diversity of habitats. This chapter also aims to determine whether the serpentine endemics can be considered to be neo- or paleoendemics and to determine whether the profiles of the serpentine endemics differ from the profiles of endemics found on other terrestrial "islands". A further aim is to identify the threats to the serpentine endemics and to determine their conservation status. These descriptions will be used to develop a conservation plan for the serpentine outcrops and their endemics.

Materials and Methods

Extensive plant collections from the serpentine outcrops of the Barberton Greenstone Belt were first made by K. Balkwill and M-J. Balkwill in 1991 and numerous plant collectors, many of which have been listed in Appendix A (Williamson and Balkwill 2015), have visited these outcrops since. These collections were used to identify the taxa restricted to the serpentine outcrops of the Barberton Greenstone Belt. Comprehensive plant lists have been compiled for ten of the larger outcrops within the Mpumalanga Province of South Africa (Williamson and Balkwill 2015, Williamson 1994 and Changwe and Balkwill 2003) (Figure 2.1). Distributions and characteristics of and threats to presumed endemics were noted. Further data on the distributions of endemic species were obtained from PRECIS (Pretoria National Herbarium Computerised Information System) a computerised data bank managed by the South African National Biodiversity Institute, taxonomic accounts and monographs and herbarium collections and distributional data collected by scientists of the Mpumalanga Tourism and Parks Agency. Voucher specimens of the endemics, which are housed in the CE Moss Herbarium of the University of the Witwatersrand, are listed in Appendix C.

As there are no comprehensive lists of plant taxa endemic to the Mpumalanga Province (MP) and the Barberton Centre of Plant Endemism (BCPE), the online South African National Biodiversity Institute’s online plant checklist (http://posa.sanbi.org) was used to compile lists of plants recorded from the relevant quarter degrees squares. These lists were filtered to produce lists of plants recorded from only the MP and BCPE in order to make comparisons with the levels of endemism in families and genera represented by the Barberton Greenstone Belt endemic taxa. The levels of endemism in the MP and BCPE were compared to those of the BGB by using Chi-squared analyses. Endemism is expressed as a percentage, which becomes more meaningful the greater the diversity (Harrison 2013)

Bykov’s (1979 in Major 1988) index of endemism, which allows for the comparison of levels of endemism between different sized areas, was used to assess the levels of endemism of the Barberton Greenstone Belt. The index $l_e = E_f/E_n$ where E_f is the actual percentage of endemism for a particular area and E_n is the ‘normal’ percentage of endemism read off a nomogram, where the ordinate is area and the abscissa percentage endemism. A value of $l_e = 1$ indicates that an area has the expected level of endemism for its size. A value >1 implies an area has higher and a value < 1, lower than ‘normal’ levels of endemism. The published nomogram does not accommodate areas as small as those of the serpentine outcrops. Extrapolation of the *x* and *y*-axes allowed an estimate of what the ‘normal’ percentage of endemism for an area equal to the study sites should be.

The number of endemics and percentage endemism were regressed against a variety of physical and biological characteristics of the outcrops, using the data analysis tools of Microsoft Excel 2010. These characteristics included average and range of altitude above sea level, average latitude, surface area, soil Ni and Cr concentration, soil Mg:Ca ratios as well as the plant alpha diversity of the vegetation of each outcrop and the surrounding non-serpentine vegetation.

In order to determine if there is a biological profile characteristic of the serpentine endemic taxa, data on growth form, pollination and dispersal mechanisms were compiled from observations in the field and taxonomic monographs and descriptions in the literature. The types of growth forms recognised include herbs, graminoids, small shrubs and shrubs to small trees. The pollination vectors of plants were categorised as bird, insect or wind. The dispersal mode categories recognised were: wind, bird, insect, ballistic and unknown. The frequency of each of the biological traits within the BGB endemic taxa was compared to that of the excluded taxa (listed in Appendix B) using Chi-square analysis.

The nearest possible relative of each serpentine endemic taxon was identified from monographs and taxonomic accounts. The geographical distribution of the nearest relative was determined from the South African National Biodiversity Institute’s online plant checklist (http://posa.sanbi.org) and compared to the distribution of the serpentine endemic. The distribution and number of populations of the endemic taxon, its extent of occurrence and dispersal profile were used to determine whether the endemic is palaeoendemic or neoendemic.

Results and Discussion

Twenty-seven plant taxa were found to be restricted (i.e. endemic) to the serpentine soils derived from the outcrops of serpentinite of the Barberton Greenstone Belt (Williamson and Balkwill 2015) with an additional species, *Dioscorea strydomiana* Wilkin reported here. Five additional taxa are mainly restricted to these serpentine outcrops but have also been recorded from a few sites that are also ferromagnesium ultramafics but do not contain all the serpentinite minerals (Table 3.1). There are more than 80 taxa endemic or near endemic to the Barberton Centre of Endemism (BCE) (Van Wyk and Smith 2001). Of these 13 are serpentine endemics, which represent 16% of species listed as endemic to the BCE. This, however, does not include the twelve undescribed taxa and eight other species known to be serpentine endemics. Adding these would increase the percentage to 33%.

Taxonomic aspects of endemism

The plant taxa that are considered to be endemic to the Barberton Greenstone Belt are relatively widespread taxonomically representing 14 families (15% of total). In contrast, the endemic taxa only represent 24 genera out of the 319 recorded from the Barberton Greenstone Belt (7.5% of total) (Table 3.1). Only the genus *Berkheya* is represented by four endemic taxa, *Ocimum* has three endemic taxa, with *Brachystelma, Senecio, Helichrysum* and *Ozoroa* each having two endemic taxa. Five endemics, namely *Aloe thorncroftii* Pole-Evans, *Dioscorea strydomiana*, *Kniphofia umbrina* Codd, *Gladiolus serpenticola* Goldblatt and J.C.Manning and *Sartidia dewinteri* J.Munday and L.Fish are monocotyledons, with the remaining taxa representing the dicotyledons. As yet, the endemic taxa of the Barberton Greenstone Belt do not include any gymnosperms or ferns. Some of the taxa endemic to the ultramafic outcrops of the Great Dyke are from the same genera as those from the serpentine of the Barberton Greenstone Belt. These genera include *Aloe*, *Dicoma, Ozoroa* and *Searsia* (Brooks and Malaisse 1985).

Comparing the levels of endemism of the Barberton Greenstone Belt with that of the MP and the BCPE reveals that there is a higher number of endemics than expected within the Asteracease ($P<0.005$) and the Anacardiaceae ($P<0.05$) (Table 3.2). A lower number of endemics than expected were recorded for the Fabaceae, although this is considered to be slightly or not statistically significant at $P = 0.09$. The high number of endemics within the Asteraceae and Anacardiaceae would suggest some genetic

Table 3.1: List of taxa considered to be endemic to the Barberton Greenstone Belt, including those found on outcrops in Swaziland. Site names are abbreviated as follows: Groenvaly (GV), Magnesite Mine (MM), Mundt's Concession (MC), Rosentuin (RT), Sawmill (SM), Agnes Mine (AM), Kaapsehoop (KH) and Dunbar (DB).

Family	Taxon	Nickel hyper-accumulator	GV	MM	MC	RT	SM	AM	KH	DB	Serpentinite specialist (S)/ also on non-serpenitinite ultramafics (U)
Asteraceae	*Athrixia* sp. nov.		X								S
	Berkheya coddii Roessler	✓	X			X	X	X		X	S
	Berkheya nivea N.E.Br.	✓			X						S
	Berkheya rehmannii Thell. var. *rogersiana* Thell.	✓	X			X	X	X	X	X	S
	Berkheya sp. nov. aff. *B. seminivea* Harv. & Sond.										U
	Dicoma swazilandica S.Ortiz		X							X	S
	Helichrysum sp. nov. aff. *H. nudifolium* (L.) Less.**										S
	Helichrysum sp. nov. aff, *H. albo-brunneum* S.Moore									X	S
	Macledium zeyheri subsp. *thyrsifolium* (Klatt) Netnou			X	X						S
	Senecio sp. aff. *S. anomalochrous* Hilliard	✓							X		S
	Senecio sp. aff. *S. coronatus* (Thunb.) Harv.*	✓	X					X	X		S
Acanthaceae	*Asystasia subbiflora* C.B.Clarke		X		X		X	X			S

Family	Taxon	Nickel hyper-accumulator	GV	MM	MC	RT	SM	AM	KH	DB	Serpentinite specialist (S)/ also on non-serpenitinite ultramafics (U)
	Salpinctium hirsutum T.J.Edwards								X		S
	Sclerochiton triacanthus A.Meeuse				X						S
Anacardiaceae	*Searsia pygmaea* (Moffett) Moffett		X		X		X	X			S
	Ozoroa barbertonensis Retief						X	X			S
	Ozoroa sp. nov.									X	S
Lamiaceae	*Ocimum motjaneanum* McCallum & K.Balkwill**										S
	Ocimum sp. nov.1 (Barberton species)					X		X		X	S
	Ocimum sp. nov. 2						X				S
Apocynaceae	*Brachystelma dyeri* K.& M.Balkwill							X			U
	Brachystelma sp. nov.aff. *B. longifolium* (Schltr.) N.E.Br.										S
Asphodelaceae	*Kniphofia umbrina* Codd**										S
	Aloe thorncroftii Pole-Evans		X					X			S
Fabaceae	*Indigofera crebra* N.E.Br.		X				X	X			U
	Senegalia loetteri N.Hahn				X						S
Celastraceae	*Gymnosporia* sp. nov.				X						S
Dioscoreaceae	*Dioscorea strydomiana* Wilkin										S
Iridaceae	*Gladiolus serpenticola* Goldblatt & J.C.Manning				X						S
Lobeliaceae	*Cyphia bolusii* E.Phillips								X		U
Orobanchaceae	*Graderia* sp. nov.*					X					?

Family	Taxon	Nickel hyper-accumulator	GV	MM	MC	RT	SM	AM	KH	DB	Serpentinite specialist (S)/ also on non-serpenitinite ultramafics (U)
Poaceae	*Sartidia dewinteri* J.Munday & L.Fish		X			X	X	X	X	X	U
Proteaceae	*Protea curvata* N.E.Br.				X						S

* further research needed to confirm its status as an endemic (R. Reddy *pers. comm.*.)[1]

** endemic taxa from the outcrops of the Barberton Greenstone Belt within Swaziland (McCallum 2006)

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[1] Renee Reddy, C.E.Moss Herbarium, University of the Witwatersrand, Johannesburg.

Table 3.2: The number of endemic species in each family represented by the Barberton Greenstone Belt endemics compared to the relative levels of endemism in the Mpumalanga Province (MP) and Barberton Centre of Plant Endemism (BCPE), excluding the serpentine endemics. The chi-square analysis (χ^2) tests the null hypothesis that the observed frequency of endemics in the BGB is not different from that expected based on the number of endemics per family in the rest of the flora (NS = not significant).

Family	Number of species in family			Significance	
	Mp	BCPE	BGB endemics	MP and BGB	BCPE and BGB
Asteraceae	22	11	10	$P<0.005$	NS
Acanthaceae	10	2	3	NS	NS
Anacardiaceae	5	3	3	$P<0.05$	NS
Lamiaceae	10	3	2	NS	NS
Apocynaceae	15	4	2	NS	NS
Asphodelaceae	11	3	1	NS	NS
Fabaceae	27	3	2	NS	NS
Celastraceae	2	2	1	NS	NS
Iridaceae	19	1	1	NS	NS
Lobeliaceae	4	1	1	NS	NS
Orobanchaceae	2	1	1	NS	NS
Poaceae	12	3	1	NS	NS
Proteaceae	2	1	1	NS	NS

pre-adaptation to the chemical characteristics of ultramafic soils. This possible genetic pre-adaptation to serpentine soils seems most prevalent within *Berkheya* and *Helichrysum* of the Asteraceae and *Ozoroa* of the Anacardiaceae (Table 3.3), although statistical results show this trend to be only slightly significant ($P=0.08$ and 0.06 respectively). The Lamiaceae does not show significantly higher endemism on the BGB compared with the BCPE and MP (Table 3.2). However, the genus *Ocimum* is represented by a significantly larger number of endemic species on the BGB than expected ($P<0.02$) (Table 3.3). BGB endemism within *Salpinctium* (Acanthaceae) and *Sartidia* (Poaceae) is seen as significantly higher than expected ($P<0.001$) (Table 3.3) as there are no other representatives of these genera found within the BCPE and MP.

Table 3.3: The number of species in each genus, represented by the Barberton Greenstone Belt endemics, in the Mpumalanga Province (MP) and Barberton Centre of Plant Endemism (BCPE) compared to the relative levels of endemism on the Barberton Greenstone Belt (BGB). The chi-square analysis (χ^2) tests the null hypothesis that the observed frequency of endemics in the BGB is not different from that expected based on the size of the genus in the rest of the flora (NS = not significant).

Genus	Number of species in Genus			Significance	
	MP	BCPE	BGB endemics	MP and BGB	BCPE and BGB
Athrixia	5	1	1	NS	NS
Berkheya	20	13	4	NS	NS
Dicoma	4	3	1	NS	NS
Helichrysum	83	54	2	NS	NS
Macledium	1	1	1	P<0.05	NS
Senecio	70	39	2	NS	NS
Asystasia	3	1	1	NS	NS
Salpinctium	0	0	1	P<0.001	P<0.005
Sclerochiton	3	1	1	NS	NS
Searsia	32	19	1	NS	NS
Ozoroa	5	4	2	P<0.05	NS
Ocimum	8	5	3	P<0.02	P<0.02
Brachystelma	24	9	2	NS	NS
Kniphofia	12	6	1	NS	NS
Aloe	43	22	1	NS	NS
Indigofera	6	37	1	NS	NS
Senegalia	8	7	1	NS	NS
Gymnosporia	12	9	1	NS	NS
Dioscorea	5	5	1	NS	NS
Gladiolus	30	16	1	NS	NS
Cyphia	9	1	1	NS	NS
Graderia	3	2	1	NS	NS
Sartidia	0	0	1	P<0.001	P<0.005
Protea	10	6	1	NS	NS

The widespread taxonomic representation of the Barberton Greenstone Belt serpentine endemic taxa is also seen in the Californian serpentine vegetation (Anacker *et al*. 2010). Serpentine habitat specialists (i.e. "endemics") in the Californian flora are from 103 genera and 41 families, including angiosperms, gymnosperms, and ferns (Safford *et al.* 2005). The four families with the largest number of serpentine endemics are the Asteraceae, Acanthaceae, Anacardiaceae and Lamiaceae (Table 3.1). However, endemism in the Barberton Centre of Plant Endemism (BCPE) is pronounced in the Iridaceae, Lamiaceae and Liliaceae *sens. lat.* (van Wyk and Smith 2001). Families that include endemic species on the escarpment of the Limpopo Province (North-eastern Transvaal escarpment) (Matthews *et al.* 1993) are similar to those of the BCPE, i.e. Liliaceae *sens. lat.*, Iridaceae, Asteraceae and Lamiaceae. The families with the largest numbers of endemics on the Dolomite outcrops of the escarpment are the Liliaceae *sens. lat.*, Euphorbiaceae, Lamiaceae and Acanthaceae (Matthews *et al.* 1993). It is noticeable that both dolomite and serpentine outcrops have endemic species within the Lamiaceae and Acanthaceae. However, the Euphorbiaceae are not represented in the endemics of the Barberton Greenstone Belt and the Asteraceae is well represented, but there is only one dolomite endemic in the Asteraceae. The serpentine endemics represent a number of different families to those of other areas, which suggests that these families have a genetic pre-adaptation for the edaphic conditions of serpentine soils.

Levels of endemism

No endemic taxa have been found on the Core Zone (CZ) and Kalkkloof (KK) sites and only one endemic (*Macledium zeyheri* subsp. *thyrsifolium* (Klatt) Netnou) has been found on the Magnesite Mine (MM) site. The Agnes Mine (AM) and Groenvaly (GV) sites have the largest numbers of endemics with eleven and ten, respectively. However, the Sawmill (SM) site has fewer endemic species (8), although it has a higher percentage of endemic species (5.3%) than GV (Table 3.4).

The estimated values for Bykov's index of endemicity ($l_{e)}$ (Table 3.4) show that all the serpentine sites with endemic taxa have a higher than expected degree of endemism (l_e >1) based on Bykov's quantitative index of endemism (1979 in Major 1988). The points

Table 3.4: Endemism data collected for each of the selected serpentine outcrops of the Barberton Greenstone Belt

Study site	No. of specific and infra-specific taxa	No. of endemics	% endemism	Bykov's Index of Endemicity (l_e)
AM *	210	11	5.3	24.7
GV	285	10	3.5	12.9
MC	206	9	4.4	24.9
SM	151	8	5.3	28.8
Swaziland sites **	600	8	1.3	7.3
DB #	254	7	2.8	27.0
KH *	181	6	3.3	17.1
RT	184	5	2.7	25.6
MM	179	1	0.6	2.3
CZ	162	0	0	0
KK	182	0	0	0
Total	1202	33	2.7	4.8

*Williamson (1994)

**McCallum (2006)

Changwe and Balkwill (2003)

on the graph (Figure 3.1) showing the percentage endemism for the combined data of the sampled sites of the Barberton Greenstone Belt occur below Bykov's line of 'normal' endemicity indicating higher than 'normal' percentage endemism. The vegetation of the Barberton Greenstone Belt shows similar levels of endemism when compared to larger areas known to support high numbers of endemic taxa.

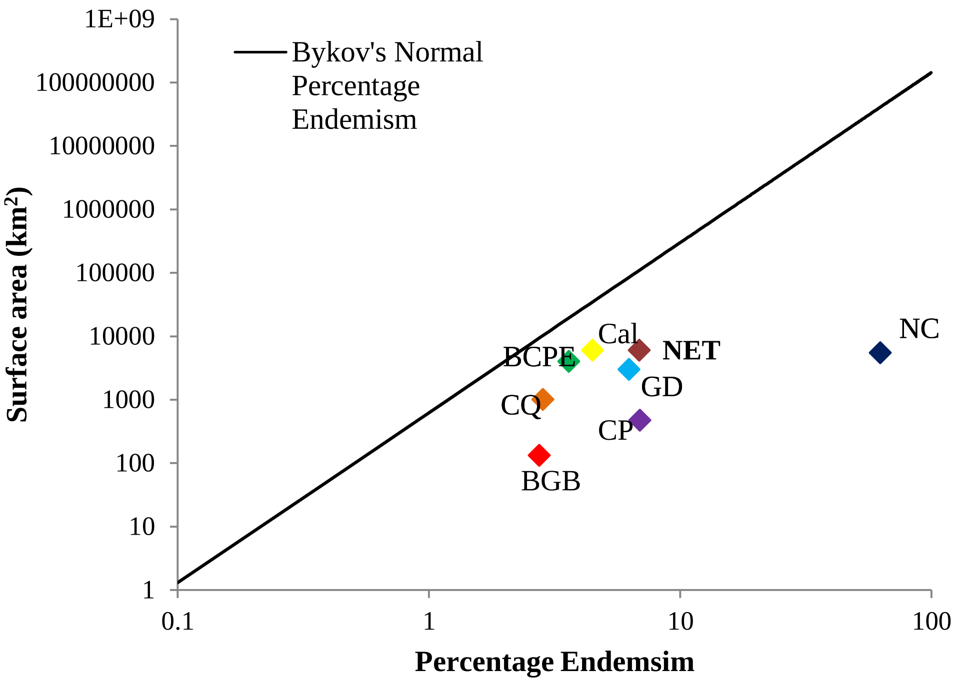

Figure 3.1: Graph showing Bykov's normal percentage endemism (solid line) as compared to the endemism of the serpentine vegetation of the Barberton Greenstone Belt (BGB) and the serpentine/ultramafic areas of New Caledonia (NC) (Jaffré 1992), California (Cal) (Safford *et al.* 2005), Central Queensland (CQ) (Batianoff *et al*. 2000), the Great Dyke (GD) (Wild 1965) as well as the Barberton Centre of Plant Endemism (BCPE) (van Wyk and Smith 2001), North Eastern Transvaal (NET) (Matthews *et al.* 1993) and the Cape Peninsula (CP) (Helme and Trinder-Smith 2006). The distance from the line is proportional to the deviation from normal levels of endemism. Points below the line indicate greater than expected endemism.

The generalisation that number of endemics increases with increasing area (Cowling 2001) does not apply to the endemics of the Barberton Greenstone Belt as there was no statistical correlation between the number of endemics or the percentage endemism and the surface area of the outcrops ($P > 0.5$) (Table 3.5). There is also no significant correlation between the number of endemics or percentage endemism and mean altitude and latitude of the outcrops ($P > 0.2$). The number of endemics and percentage endemism of each outcrop does seem to be dependent on the soil chemistry. There is a positive correlation between the levels of endemism and Ni concentration ($P < 0.05$) and between percentage endemism and Cr concentration ($P < 0.05$) and a weak negative

Table 3.5: R-values calculated from the regression analysis conducted to correlate the number of endemics and percentage endemism with various environmental and biological conditions as described in Chapters 1 and 2. Significant correlations ($P<0.05$) are indicated with bold font

	R-values calculated for the correlation with various environmental conditions								
	α - diversity of serpentine sites	α - diversity of non-serpentine areas	No of recorded taxa	Mean altitude of outcrop	Mean latitude of outcrop	Surface area of outcrop	[Ni]	[Cr]	Ca: Mg
No of endemics	0.150	-0.589	0.534	0.420	0.095	0.197	**0.781**	**0.651**	**-0.570**
% endemism	0.004	-0.599	0.222	0.366	0.110	0.060	**0.895**	**0.794**	**-0.565**

correlation with the Ca: Mg ($P > 0.1$). Similar results have been reported from serpentine outcrops in Queensland, Australia and California. Batianoff and Singh (2001 found a direct correlation between soil Ni and the number of endemic species in open forests i.e. the greater the soil Ni concentration, the higher the number of endemics. Harrison (1999) regressed the local diversity of endemic species, in Californian serpentine outcrops, on calcium levels for patches and continuous sites and found that endemic diversity declined significantly with calcium levels on continuous sites ($r = -0.74$, $P < 0.001$) but not on small patches ($r =-0.22$, $P = 0.29$). The regression correlation between Ca: Mg and percentage endemism of the BGB serpentine outcrops strengthens to $r = 0.84$ when the more isolated sites are removed from the analysis. These results show that there are similar interactions between endemic diversity and the patchiness of outcrops as on the Californian serpentine outcrops.

A negative statistical correlation between the levels of endemism and the species richness of the surrounding non-serpentine vegetation was calculated (Table 3.5). This suggests that sites that have lower surrounding diversity support higher numbers of endemics. However, this correlation was found to be not significant ($P > 0.1$). It is possible that with additional data from more sites, this relationship could become significant.

Patterns of edaphic endemism have in the past been compared to the patterns of endemism on oceanic islands (Major 1988). Shmida and Werger (1992) showed that the numbers of island endemics, on the Canary Islands, are positively correlated with island area and height of the highest peak (which is a proxy for altitudinal range). The outcrops of the Barberton Greenstone Belt, however, do not show similar patterns of endemism to those of the Canary Islands.

Distribution of endemic taxa within the Barberton Greenstone Belt

The map showing the number of endemics recorded at each of the sampled outcrops (Figure 3.2) shows that the larger, central outcrops of the Barberton Greenstone Belt harbour the largest number of endemics. However, as previously noted, there is no significant statistical correlation between the size and position of the outcrops and the levels of endemism. Eight of the 29 (28%) known endemics (disregarding *Graderia* sp.

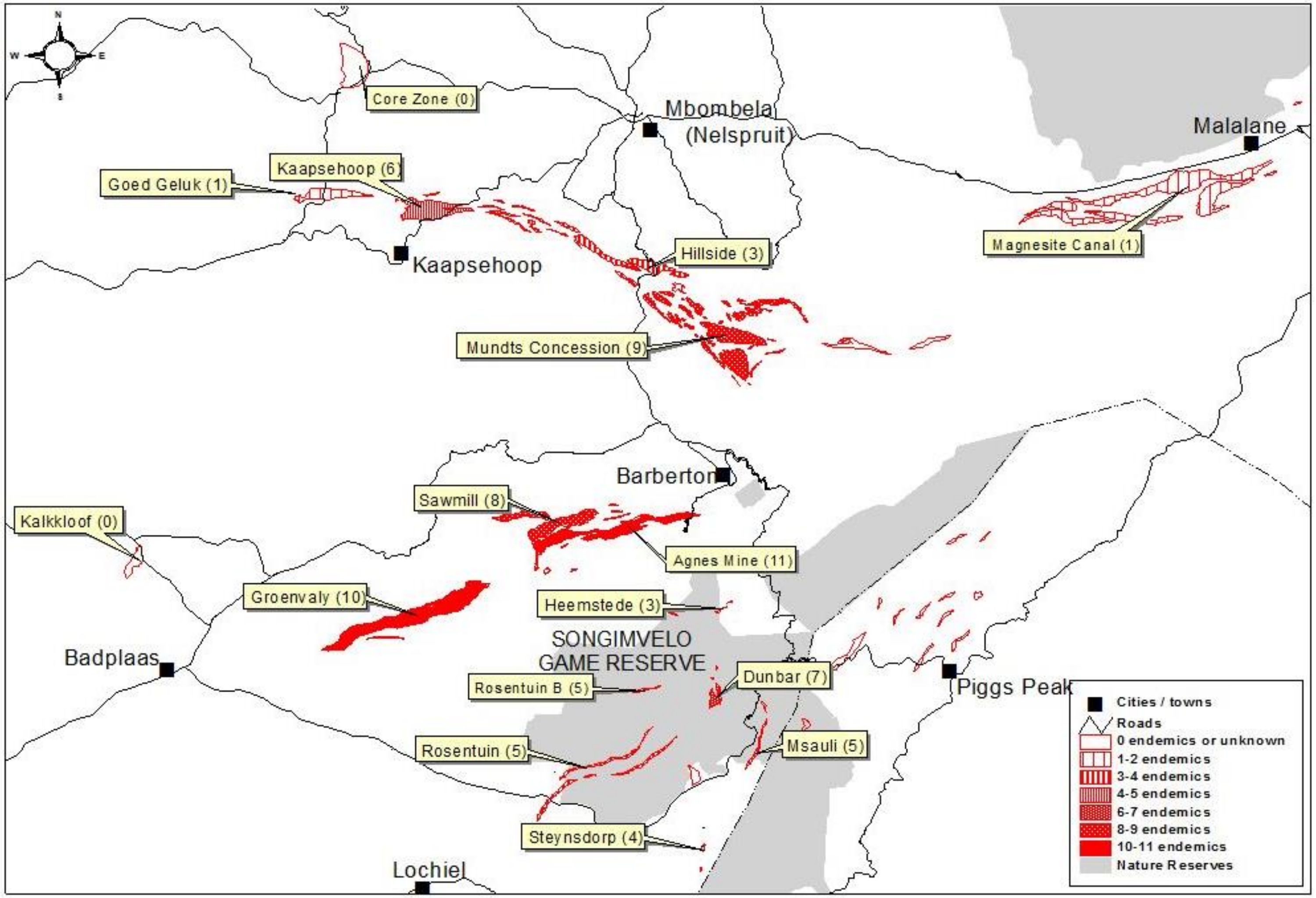

Figure 3.2: Map of the serpentine outcrops of the Barberton Greenstone Belt showing the number of endemics recorded from each outcrop.

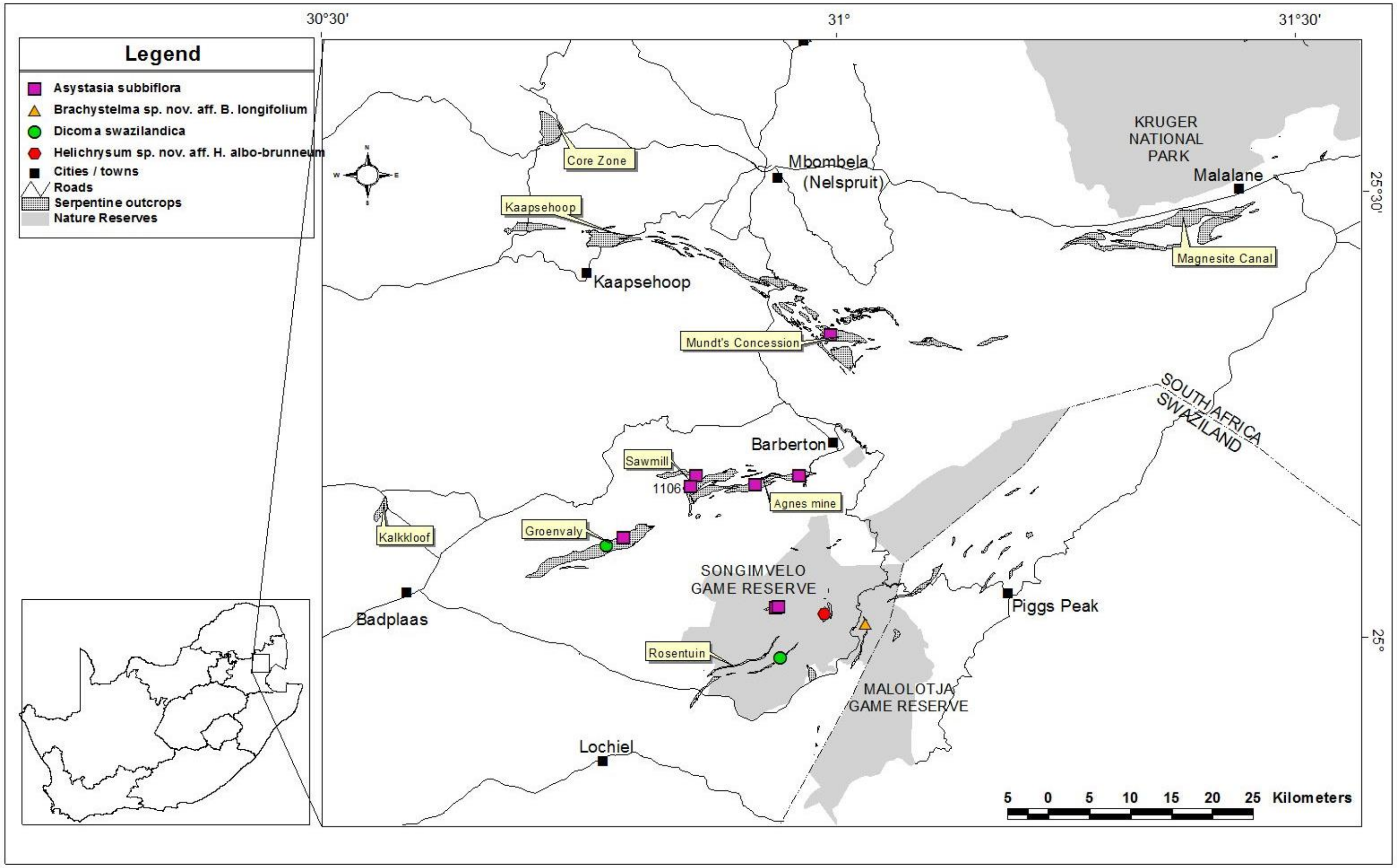

Figure 3.3: Maps showing the known distributions of *Asystasia subbiflora*, *Brachystelma* sp. nov. aff. *B. longifolium, Dicoma swazilandica* and *Helichrysum* sp. nov. aff. *H. albo-brunneum,* which are considered to be endemic to the South African outcrops of the Barberton Greenstone Belt.

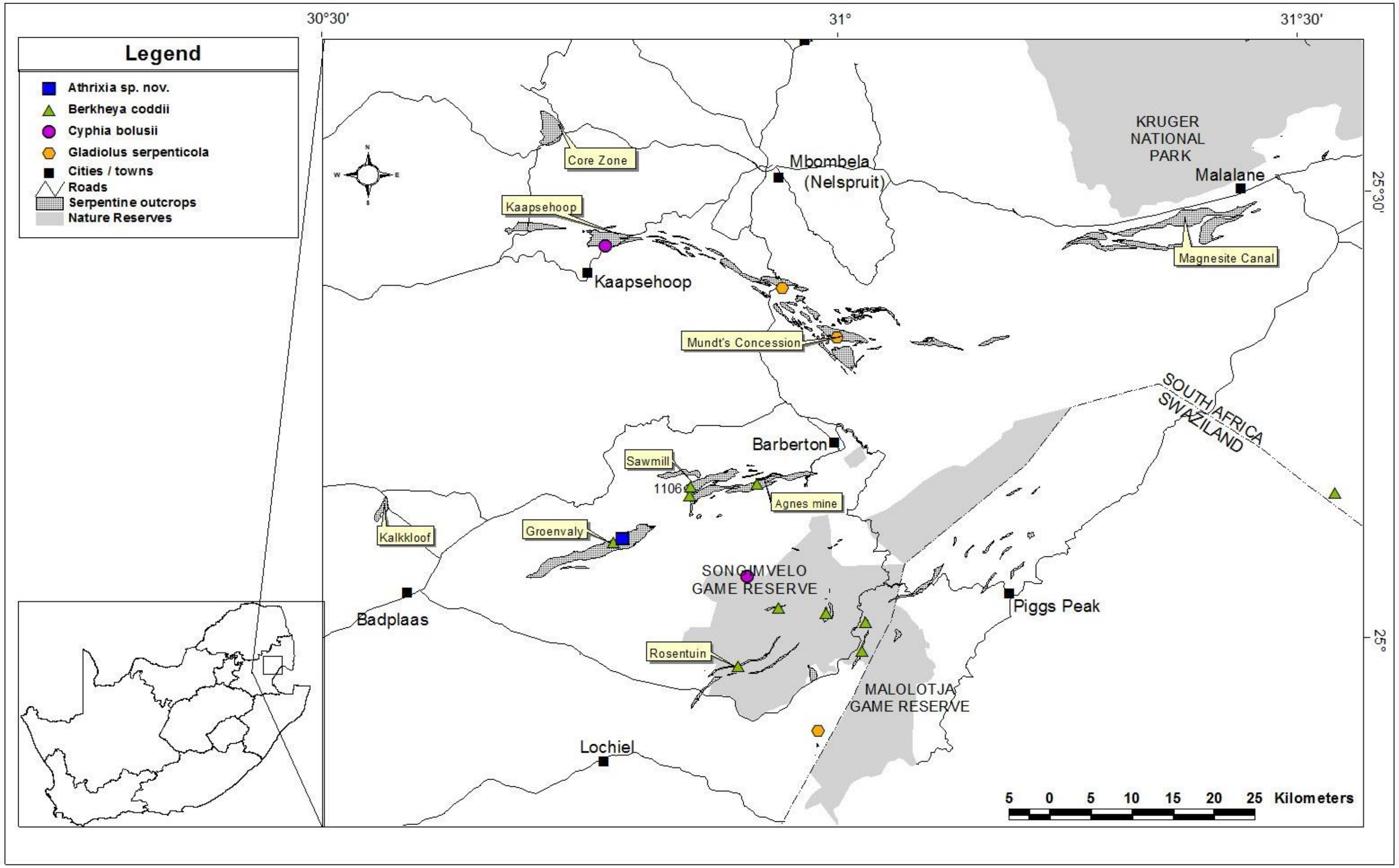

Figure 3.4: Maps showing the known distributions of *Athrixia* sp. nov., *Berkheya coddii*, *Cyphia bolusii* and *Gladiolus serpenticola*, which are considered to be endemic to the South African outcrops of the Barberton Greenstone Belt.

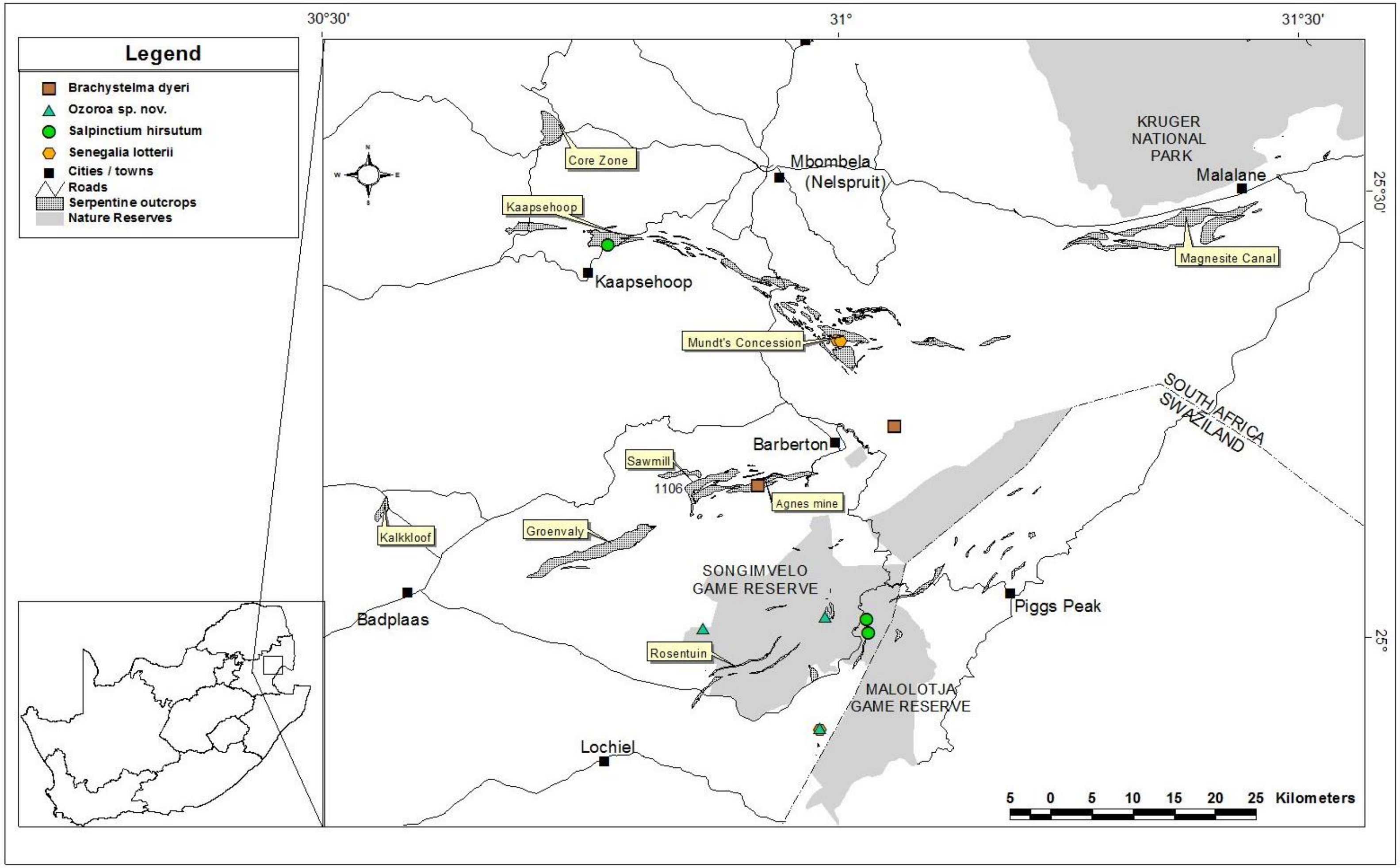

Figure 3.5: Maps showing the known distributions of *Brachystelma dyeri*, *Ozoroa* sp. nov., *Salpinctium hirsutum* and *Senegalia loetteri*, which are considered to be endemic to the South African outcrops of the Barberton Greenstone Belt.

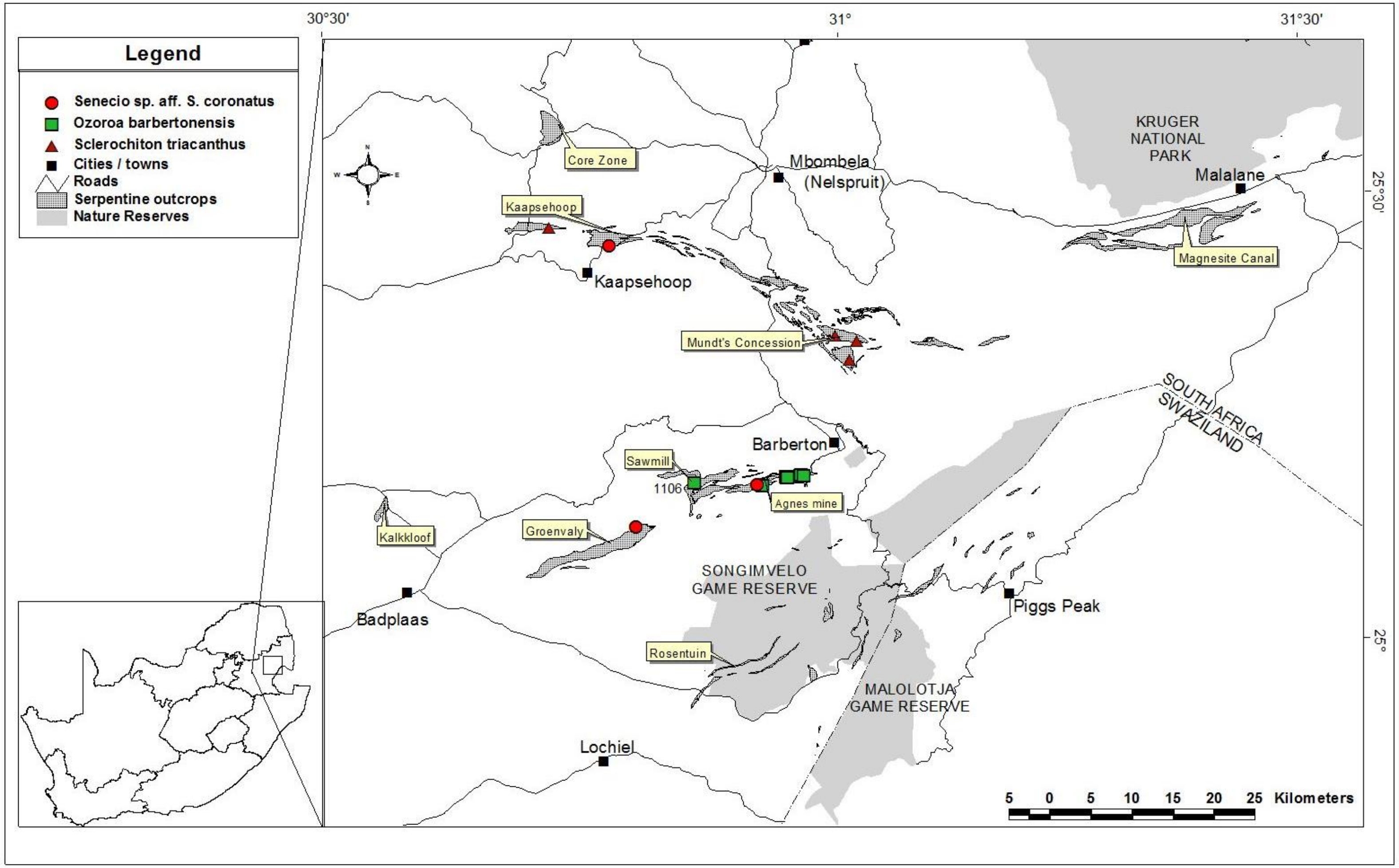

Figure 3.6: Maps showing the known distributions of *Senecio* sp. nov. aff *S. coronatus*, *Ozoroa barbertonensis* and *Sclerochiton triacanthus*, which are considered to be endemic to the South African outcrops of the Barberton Greenstone Belt.

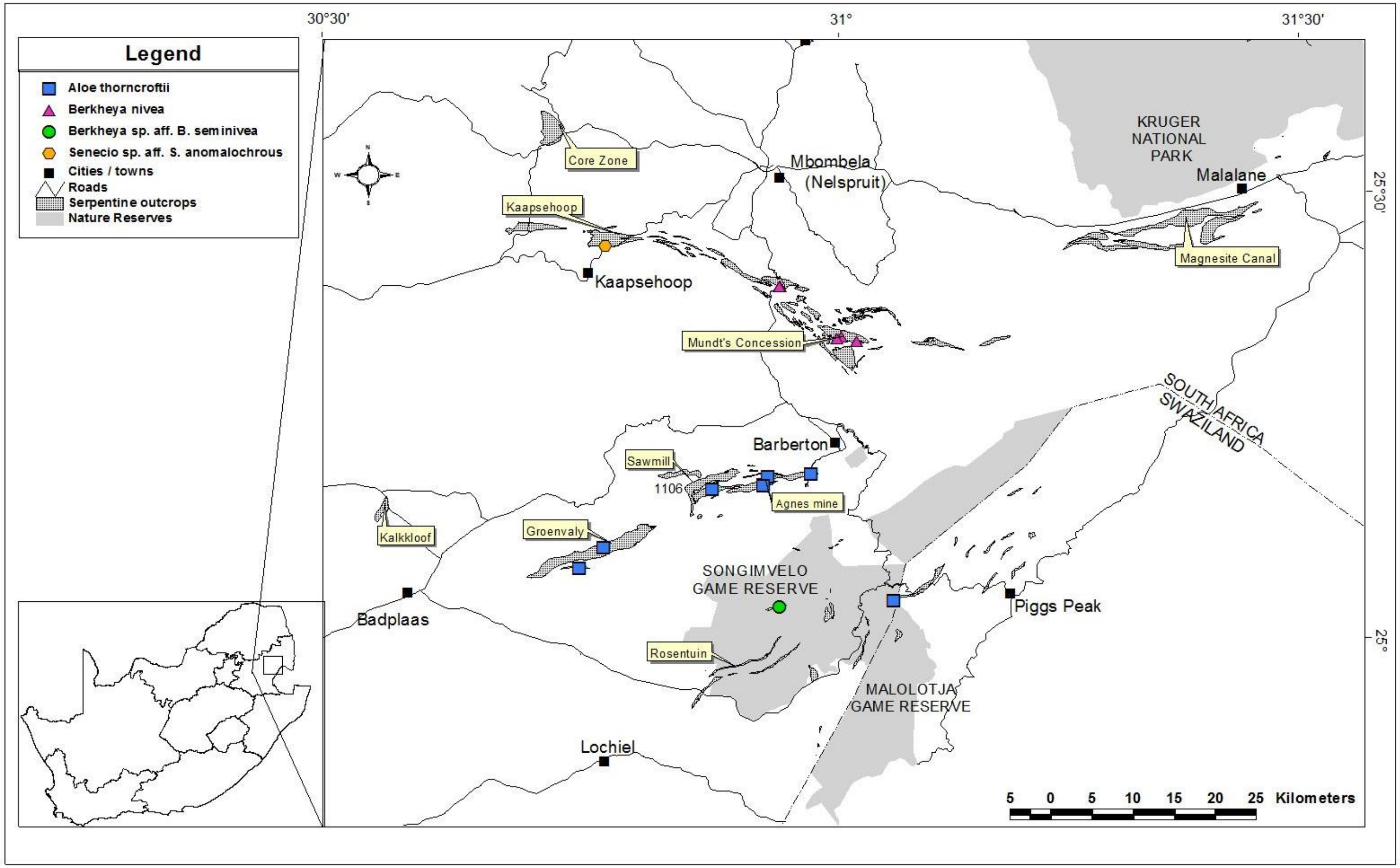

Figure 3.7: Maps showing the known distributions of all *Aloe throncroftii*, *Berkheya nivea*, *Berkheya* sp. nov. aff *B. seminivea* and *Senecio* sp. nov. aff *S. anomalochrous*, which are considered to be endemic to the South African outcrops of the Barberton Greenstone Belt.

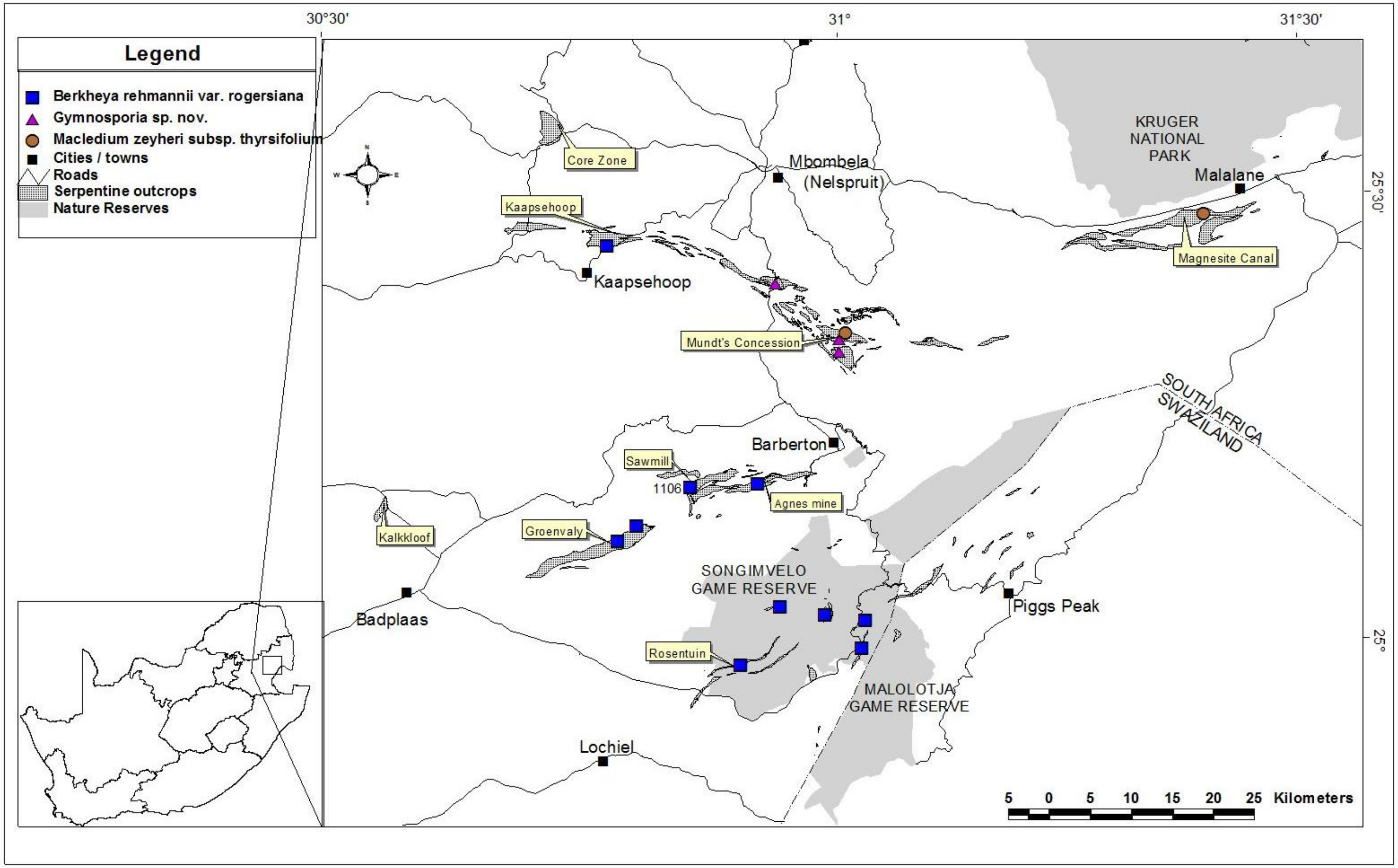

Figure 3.8: Maps showing the known distributions of *Berkheya rehmannii* var. *rogersiana*, *Gymnosporia* sp. nov. and *Macledium zeyheri* subsp. *thyrsifolium*, which are considered to be endemic to the South African outcrops of the Barberton Greenstone Belt.

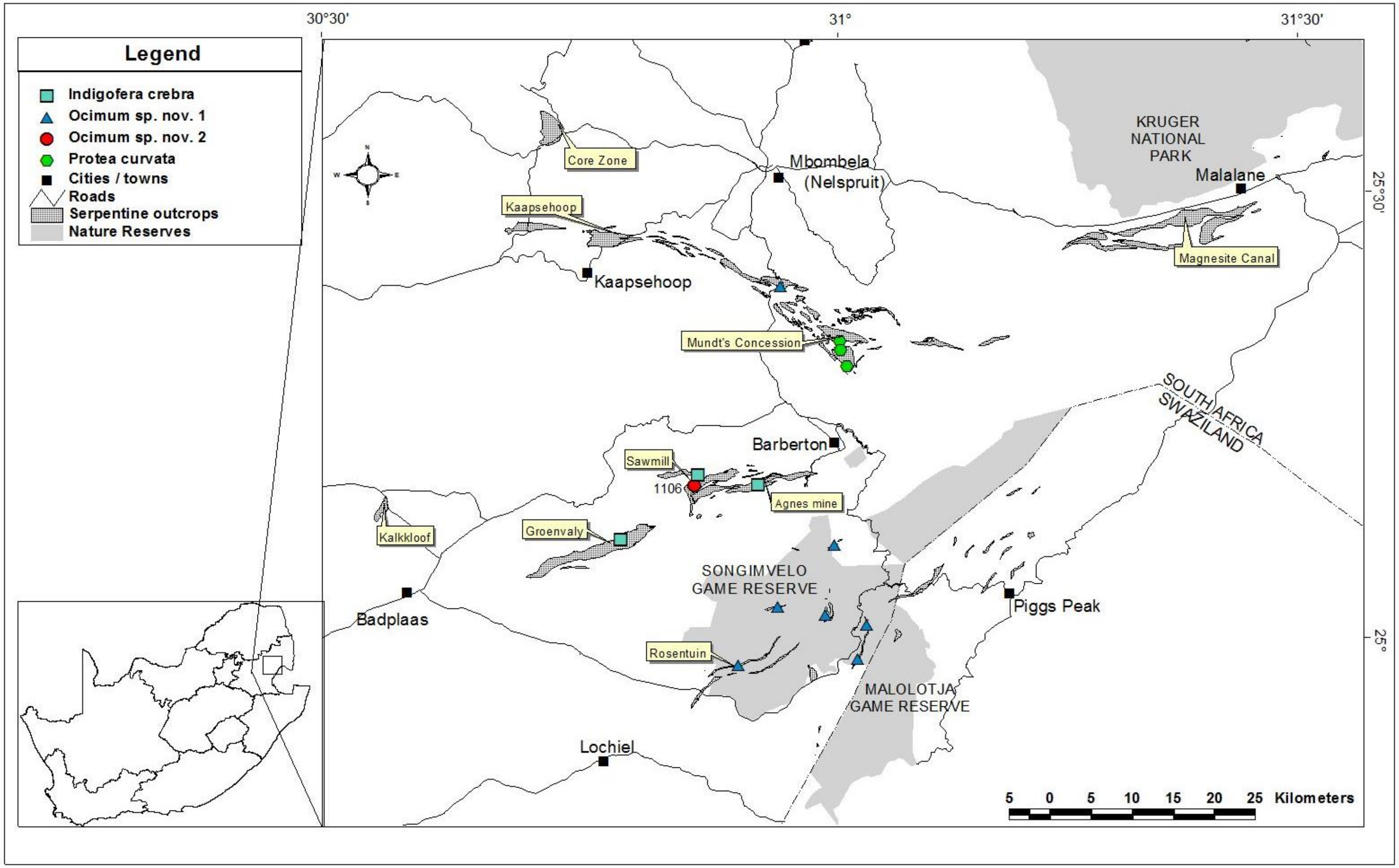

Figure 3.9: Maps showing the known distributions of *Indigofera crebra*, *Ocimum* sp. nov. 1, *Ocimum* sp. nov. 2 and *Protea curvata*, which are considered to be endemic to the South African outcrops of the Barberton Greenstone Belt.

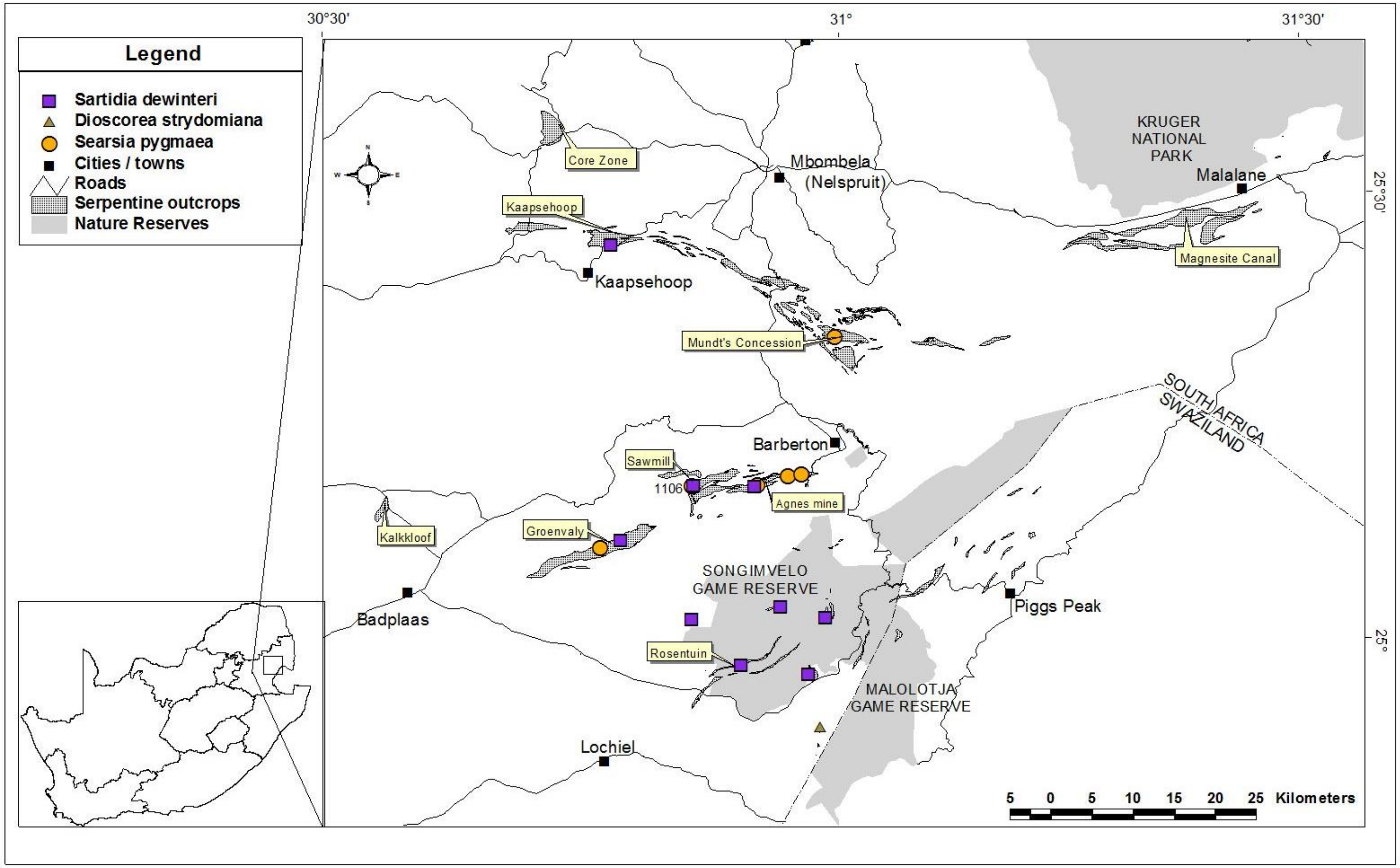

Figure 3.10: Maps showing the known distributions of *Sartidia dewinteri*, *Dioscorea strydomiana* and *Searsia pygmaea*, which are considered to be endemic to the South African outcrops of the Barberton Greenstone Belt.

nov. and the Swaziland taxa) have been recorded from only one outcrop (Figures 3.3 – 3.10) and each of these taxa occurs on a different outcrop. Eight endemics (28%) have been recorded from two of the sampled sites and the rest occur on three or more of the sampled sites. *Sartidia dewinteri* and *Berkheya rehmannii* Thell. var. *rogersiana* have been recorded from six, and *Berkheya coddii* Roessler has been recorded from five, of the sampled sites considered in this study (Table 3.1) and each of these taxa have been recorded from a total of nine outcrops of the Barberton Greenstone Belt and they are thus the most widespread endemic taxa. Many of the endemic taxa are restricted to particular ‘groups’ of outcrops (Figures 3.3 – 3.10) with the exception of *Berkheya coddii* (Figure 3.4), *Berkehya rehmannii* var. *rogersiana* (Figure 3.8), *Sartidia dewinteri* (Figure 3.10), *Asystasia subbiflora* C.B.Clarke (Figure 3.3), *Searsia pygmaea* (Moffett) Moffett (Figure 3.10), *Senecio* sp. aff. *S. coronatus* (Thunb.) Harv. (Figure 3.6) and *Ocimum* sp. nov. 1 (Figure 3.9), which are more widespread across the Barberton Greenstone Belt. Three endemic taxa are found only on the Swaziland serpentine outcrops (McCallum 2006). Of the sites investigated during this study, the Groenvaly site hosts the most endemic taxa, with ten endemics having been recorded from this outcrop (Table 3.4). However, the Agnes Mine site harbours eleven endemics (Williamson 1994). Of the nine endemics that occur at the Mundt’s Concession outcrop, three taxa i.e. *Berkheya nivea* N.E.Br. (Figure 3.7), *Gymnosporia* sp. nov. (Figure 3.8) and *Protea curvata* N.E.Br. (Figure 3.9) occur only in that cluster of outcrops and not on sites further south or north. *Senegalia loetteri* N.Hahn and *Gladiolus serpenticola* have a similar affinity to that cluster of outcrops but have also been recorded from the Ebutsini site further south and *S. loetteri* also occurs at its type locality at Opsaal (Figures 3.4 and 3.5).

Some taxa e.g. *Sartidia dewinteri* (Figure 3.10), *Indigofera crebra* N.E.Br (Figure 3.9), *Berkheya* sp. nov. aff. *B. seminivea* Harv. & Sond. (Figure 3.7) and *Cyphia bolusii* E.Phillips (Figure 3.3) are found predominantly on outcrops of the BGB where the soils contain the mineral serpentinite (i.e. serpentine). Each of these taxa has also been collected at locations that are not recorded on geological maps as serpentine outcrops. These localities are however shown on geological maps as other ferro-magnesium rich ultramafics of the Onverwacht group, which show very similar patterns of soil chemistry and toxicity as the serpentine outcrops (Balkwill *et al.* 2011). These taxa would still be considered to be edaphic specialists restricted to ultramafic outcrops as opposed to strict serpentine endemics (restricted to only the serpentinite outcrops) of the Barberton Greenstone Belt.

No endemic taxa have been recorded for two of the sampled sites i.e. Corezone and Kalkloof and only one endemic taxon, *Macledium zeyheri* subsp. *thyrsifolium*, has been recorded on the Magnesite Mine site (Figure 3.8). Until recently this species was thought only to occur on the northeastern group of outcrops but a recent discovery of *M. zeyheri* subsp. *thyrsifolium* at the Mundt's Concession site (M. Lötter *pers comm.*[2]) suggests that its distribution is wider.

The numbers of endemics shared between pairs of serpentine outcrops (Table 3.6) indicate that many of the outcrops are relatively unique in terms of the composition of endemic species. The Agnes Mine site seems to be the least unique (but richest), sharing large numbers of its endemics with the Groenvaly (GV) site sharing eight out of 11 endemics, Sawmill (SM) site sharing six out of 11 endemics and Rosentuin (RT) site sharing five out of 11 AM endemics. The Mundts Concession (MC) group of outcrops is possibly the most unique of the Barberton Greenstone Belt outcrops sharing only two endemic species i.e. *Senegalia loetteri* and *Gladiolus serpenticola* with the most southern site, Ebutsini.

Harrison (1999) found that small serpentine patches in California were poorer in endemic diversity at the local level, and higher in among-site differentiation. No endemics were found on all continuous sites but not on small patches, nor on all patches but not on continuous sites. This pattern is similar to that found on the serpentine outcrops of the Barberton Greenstone Belt if we consider the outlying outcrops (KK, CZ and MM) as being patchy based on their isolation relative to the other outcrops and the central outcrops as being more continuous due to their relative proximity to one another. One possible way to explain this is that small patches are typically poor in endemic species because of low rates of colonisation and/or high rates of extinction. For the same reason, large continuous sites can be richer in endemic species, but are only so if they are also low in calcium (Harrison 1999). Further experiments are needed to determine if this also the case for the serpentines of the Barberton Greenstone Belt. The

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[2] Mervyn Lötter, Mpumalanga Tourism and Parks Agency, Lydenburg, South Africa.

Table 3.6: The number of endemic species common to pairs of serpentine outcrops in the Barberton Greenstone Belt. Only sites that have endemic species are included in the table. The total number of endemics found on each site is indicated in parentheses after the site abbreviation.

	Number of endemic taxa in common (% of total number of endemics)						
	GV	MC	SM	DB	KH	RT	MM (1)
AM[°] (11)	8 (24)	2(6)	6 (18)	4 (12)	3 (9)	5 (15)	0
GV (10)		2 (6)	5 (15)	3 (9)	4 (12)	4 (12)	0
MC (9)			2 (6)	0	0	1 (3)	1 (3)
SM (8)				2 (6)	2 (6)	4 (12)	0
DB# (7)					3	4 (12)	0
KH[°] (6)						2 (6)	0
RT (5)							0

[°] (Williamson 1994)

[#] (Changwe and Balkwill 2003)

results thus far suggest that the soil chemistry and the geographical isolation of individual outcrops could account for the distribution of the endemic taxa.

Correlation analysis of geographical distance between sites and the Sørensen's index of similarity between sites based on endemic species shows that as the geographic distance between sites increases the similarity in endemic composition decreases ($R = -0.426$, $P< 0.02$) (Figure 3.11). The similarity between KH and RT in terms of endemic species shared is higher than predicted, considering the large geographical distance between the sites. In a comparison of physiological traits between restricted endemics and their more widespread congeners in the North Queensland tropics, Richards *et al.* (2003) concluded that the cause of narrow endemicity seems to be unique to each of the studied species. It is beyond the scope of this study to determine a physiological profile for the taxa endemic to the serpentine soils of the Barberton Greenstone Belt. However, we can determine whether the life history strategies of the endemics could account for the

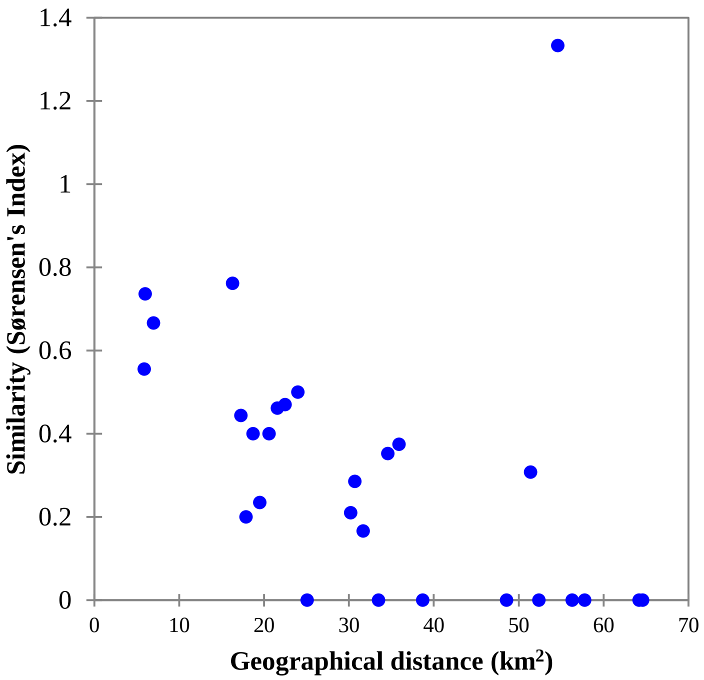

Figure 3.11: Pearson correlation analysis of geographical distance between serpentine outcrops and Sørensen's index of similarity of endemic species between sites calculated using a Mantle test.

differences in their distribution through the Barberton Greenstone Belt i.e. do the more widespread endemics have a different biological profile to the more restricted endemic taxa.

Biological profile of endemic taxa

With the exception of growth form, no biological attributes were over or under represented for the serpentine endemics (Table 3.7) when compared to those of the taxa excluded (listed in Appendix B) from the Barberton Greenstone Belt (Table 3.8). Of all the serpentine endemics, 83% have an herbaceous or suffrutex growth form and only five of the endemics are shrubs to small trees. This over representation of the herbaceous growth form is also seen in the entire serpentine flora in which herbs represent over 70% of the recorded taxa (Chapter 2). Although the herbaceous taxa are predominant within the Barberton Greenstone Belt vegetation, there is no significant difference between the frequencies of herbs within the endemic taxa as compared to the taxa excluded from the Barberton Greenstone Belt (Table 3.8). There are a significantly

Table 3.7: Some biological characteristics of taxa restricted to the serpentine and other ultramafic outcrops of the Barberton Greenstone Belt.

Taxon	Habitat	Size of populations	Growth form	Vegetative reproduction	Perennial/ Annual	Flower colour	Flowering time	Pollinator	Fruit/ seed/ spore Dispersal
Aloe thorncroftii	Crests of hills	5-10	Upright herb	None	Perennial	Orange-red	Spring	Birds	Wind
Asystasia subbiflora	Bases, lower facing slopes	50 - 100+	Prostrate herb	Rhizomatous and rooting at nodes	Perennial	Mauve	Spring - Summer	Bees	Ballistic
Athrixia sp. nov.	Bases, slopes and crests of hills	unknown	Upright herb	Possibly from rootstock	Perennial	Lilac-pink	Summer	unknown	Wind
Berkheya coddii	Bases of hills	>100	Upright suffrutex	Possibly from rootstock	Annual shoots, perennial rootstock	Yellow	Spring	Insects	Wind
Berkheya nivea	Bases and N-facing slopes	20 - 100	Upright suffrutex/ robust herb	Possibly from rootstock	Annual shoots, perennial rootstock	Yellow	Spring	Insects	Wind
Berkheya rehmannii var. *rogersiana*	Mostly N-facing slopes	>100	Upright suffrutex/ robust herb	Possibly from rootstock	Annual shoots, perennial rootstock	Yellow	Spring	Insects	Wind
Berkheya sp. nov. aff. *B. seminivea*	Slopes of hills	unknown	Robust, prickly herb	Possibly from rootstock	Annual shoots, perennial rootstock	Yellow	unknown	Insects	Wind

Taxon	Habitat	Size of populations	Growth form	Vegetative reproduction	Perennial/ Annual	Flower colour	Flowering time	Pollinator	Fruit/ seed/ spore Dispersal
Brachystelma dyeri	N-facing slope	<20	Upright herb becoming prostate	None	Annual shoots, perennial tuber	Creamy	Spring	Flies	Wind
Brachystelma sp. nov. aff. *B. longifolium*	unknown	unknown	Upright herb with fusifom roots	None	Perennial	Cream flushed pink	Early summer	unknown	Wind
Cyphia bolusii	Slopes of hills	unknown	Upright herb	None	Perennial	Pale pink	Spring	Likely bees	unknown
Dicoma swazilandica	unknown	unknown	Erect herb	None	Perennial	White	unknown	unknown	Wind
Dioscorea strydomiana	Slopes of hills	100	Shrub	None	Annual stems from woody tuber	Purplish-brown	Summer	Insects	Wind
Gladiolus serpenticola	Bases of hills	>40	Upright herb	Corm	Annual shoots - perennial rootstock	Pale pink to white	Late Summer	Bees?	Wind
Gymnosporia sp. nov.	Bases of hills	unknown	Shrub to small tree	None	Perennial	Cream	Spring	Wasps	Birds
Helichrysum sp. nov. aff. *H. albo-brunneum*	unknown	unknown	Erect herb	Possible	Perennial	Cream/ yellow	Summer	unknown	Wind
Indigofera crebra	Open grassland	500+	Upright herb	None	Perennial	Orange-red	Spring	Bees	Insects?

Taxon	Habitat	Size of populations	Growth form	Vegetative reproduction	Perennial/ Annual	Flower colour	Flowering time	Pollinator	Fruit/ seed/ spore Dispersal
Macledium zeyheri subsp. *thyrsiflorum*	N- facing slopes	unknown	Upright herb	Possible	Perennial	White	Spring	unknown	Wind
Ocimum sp. nov. 1	Bases and lower slopes	>50	Upright herb	Possible from rootstock	Annual shoots - perennial rootstock	Mauve	Spring	Bees	Rain drops
Ocimum sp. nov. 2	Lower slopes	unknown	Upright herb	Possible from rootstock	Annual shoots- perennial rootstock	Pale pink	Spring	Bees	Rain drops
Ozoroa barbertonensis	N facing slopes	20+	Upright shrublet	None	Perennial	White	Summer	Wasps	Ants/Birds
Ozoroa sp. nov.	Rocky hillsides	unknown	Suffrutex	Unlikely	Annual or biennial shoots from perennial rootstock	White or cream	Summer/ autumn	Insects	Ants/Birds
Protea curvata	Bases of hills	500+	Erect, slender tree	None	Perennial	Deep red to pink	Winter	Insects / birds	Wind?
Salpinctium hirsutum	Slopes of hills	200-250	Erect herb	None	Perennial	White	Summer	Bees	Ballistic
Sartidia dewinteri	Slopes and crests	>100	Caespitose herb	Yes	Perennial	Pale brown	Summer	Wind	Ectozoochory (mammals)

Taxon	Habitat	Size of populations	Growth form	Vegetative reproduction	Perennial/ Annual	Flower colour	Flowering time	Pollinator	Fruit/ seed/ spore Dispersal
Sclerochiton triacanthus	S - facing slopes	>30	Upright suffrutex	Stems touch ground and root	Perennial	Blue	Summer through Autumn	Bees	Ballistic
Searsia pygmaea	N facing slopes	10-20	Underground shrub	Possible from rootstock	Perennial	Greenish	Summer	Wasps	Birds
Senecio sp. aff *S. anomalochrous*	S - facing slopes	>100	Upright herb	Possible from rootstock	Annual shoots, perennial rootstock	Yellow	Spring	Insects	Wind
Senecio sp. aff *S. coronatus*	Slopes of hills	>100	Upright herb	Possible from rootstock	Annual shoots, perennial rootstock	Yellow	Spring	Insects	Wind
Senegalia loetteri	Bases of hills	unknown	Woody shrub to small tree	None	Perennial	Cream	Summer	Insects	Ballistic or birds

Table 3.8: Comparison of biological traits exhibited by the taxa excluded from the BGB and taxa endemic to the Barberton Greenstone Belt using Chi-squared analysis (NS = No significant difference).

Characteristic		Excluded taxa	Endemic taxa	Total	Significance
Growth form	Herb	20	17	37	NS
	Climber	6	0	6	NS
	Suffrutex	4	4	8	NS
	Small shrub	1	4	5	P<0.05
	Shrub/ tree	12	2	14	NS
	Tree	10	1	11	NS
	Total	53	28	81	
Vegetative reproduction	None	46	13	59	NS
	From rootstock	1	9	10	P<0.005
	Rhizomatous and nodes	0	2	2	P<0.05
	Bulb or corm	6	1	7	NS
	Total	53	25	78	
Longevity	Perennial	46	15	61	NS
	Annual	5	0	5	NS
	Annual shoots with perennial rootstock	2	11	13	P<0.005
	Total	53	26	79	
Flower colour	White	15	9	24	NS
	Cream/yellow	22	8	30	NS
	Pink/red/orange	5	6	11	NS
	Purple/blue	4	4	8	NS
	Total	46	27	73	
Flowering time	Year round	5	0	5	NS
	Spring	13	14	27	P<0.05
	Summer	32	11	43	NS
	Autumn/ Winter	3	1	4	NS
	Total	53	26	79	
Pollinator	Wind	5	1	6	NS
	Insects/bees	41	16	57	NS
	Specialist	3	4	7	NS
	Birds	1	1	2	NS
	Total	50	22	72	
Seed dispersal	Wind	16	16	32	NS
	Birds	15	4	19	NS
	Mammals	4	1	5	NS
	Mechanical	9	5	13	NS
	Total	44	25	69	

higher (P<0.05) number of small shrubs within the endemic taxa of the Barberton Greenstone Belt than there are within the excluded taxa (Table 3.8). More of the excluded taxa are trees when compared with the endemic taxa. However, this result is only slightly significant (P=0.08) (Table 3.8).

Of the approximately 250 endemic taxa reported for the serpentine outcrops of California, more than 80% are herbaceous and about 10% are woody shrubs and trees (Safford *et al.* 2005). However, the endemic floras of the New Caledonian (Jaffré 1992), Cuban (Borhidi 1992) and Central Queensland (Batianoff and Singh 2001) serpentines, include large numbers of woody shrubs and trees. The floras of the ultramafic outcrops of the Great Dyke (Brooks and Malaisse 1985) and the dolomite outcrops of the Eastern Transvaal Escarpment (Matthews *et al.* 1993) include a large proportion of herbaceous endemic taxa.

There are no annual taxa amongst the endemics of the Barberton Greenstone Belt; however, nine endemics have annual shoots that sprout from a perennial rootstock (Table 3.7). In comparison, only one of the excluded taxa exhibits this form of vegetative reproduction (Table 3.8), which is probably due to the large number of tree species representing the excluded taxa. The ability to produce annual shoots from a perennial rootstock is a characteristic found predominantly among the endemic taxa from the Asteraceae family. In contrast, the endemic flora of California includes 71 (28%) annual forbs (Safford *et al.* 2005).

Five of the endemic taxa of the Barberton Greenstone Belt may have specialist biotic pollinators (Table 3.7), with *Gymnosporia* sp. nov., *Ozoroa barbertonensis* and *Searsia pygmaea* pollinated by wasps, *Brachystelma dyeri* pollinated by flies and *Aloe thorncroftii* pollinated by birds. Further research and analysis are needed to determine whether the taxa that are pollinated by long range pollinators such as bees and birds have higher genetic diversity than those pollinated by short range pollinators (Willis *et al.* 1996). Long range pollinators will promote gene-flow between populations on separated outcrops, promoting the genetic stability of populations. Taxa with short range pollinators have reduced gene-flow between populations on separated outcrops, which will promote isolation of these populations. It is this type of isolation which would have resulted in speciation events and the development of these edaphic endemics from their congeners.

A large proportion of the endemic taxa of the Barberton Greenstone Belt belongs to the species-rich family Asteraceae, in which long-distance dispersal by wind is common (Bruchmann and Hobohm 2014). Including the taxa from this family, up to 80% of the endemic taxa of the Barberton Greenstone Belt show long distance dispersal modes including dispersal by wind and birds. These results are similar to those from studies of endemics in isolated island floras, where many endemic plants show long-distance dispersal modes (Bruchmann and Hobohm 2014). As *Salpinctium hirsutum* exhibits short-range dispersal and the two known populations are relatively far apart (55 km), this taxon could have been more common and widespread in the past, with some populations dying out. Genetic analysis is needed to determine whether there is any gene-flow between the populations of *S. hirsutum*.

Ferreira and Boldrini (2011) propose recommendations to use geological data and phylogenetic data to determine the age of endemics. However, as phylogenetic data are not available, we could conclude, on the basis of geological data only, that all the serpentine endemics are palaeoendemics as the ultramafic areas of the Barberton Greenstone Belt have been shown to be relatively old and have been exposed for about 50 million years (Annheuser *pers. comm.*[3]). However, Ferreira and Boldrini (2011) further propose that if phylogenetic data are not available distributional data and sets of morphological characteristics of a restricted taxon should be used to infer whether it diverged early or late within its group. Distribution information of the endemic taxa of the Barberton Greenstone Belt and their possible congeners (Table 3.9) and their morphological similarity to their congeners suggest that *Berkheya* sp. nov. *B. seminivea*, *Brachystelma* sp. nov. aff. *B. longifolium*, *Helichrysum* sp. nov. aff. *H. albo-brunneum*, *Macledium zeyheri* subsp. *thyrsifolium*, *Ocimum* sp. nov. 2 and *Senecio* sp. nov. aff. *S. coronatus* are neoendemics. Neoendemics are considered to be recently evolved, phylogenetically young and often rank at low taxonomic levels (Kruckeberg and Rabinowitz 1985). Many of these taxa are each found at a single locality, which

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Table 3.9: An analysis of the possible close relatives of the taxa endemic to the serpentine outcrops of the Barberton Greenstone Belt in order to determine the type of endemism represented. Nearest relatives for some endemic species were identified by examination of collected plant material and thus no published sourced are listed.

Endemic Taxon	Distribution of endemic within BGB	Number of sub-populations	Possible nearest relative	Source	Distributions sympatric / allopatric	Palaeoendemic or Neoendemic
Aloe thorncroftii Pole-Evans	Continuous	4–7	*Aloe suprafoliata* Pole-Evans	Glen & Hardy (2000)	Sympatric	Palaeoendemic
Asystasia subbiflora C.B.Clarke	Continuous	5–8	*Asystasia retrocarpa* T.J.Edwards	Edwards (1991)	Allopatric	Palaeoendemic
Athrixia sp. nov.	Single locality	2–3	*Athrixia subsimplex* Brenan		Allopatric	Palaeoendemic
Berkheya coddii Roessler	Continuous	10–15	*Berkheya angolensis* O. Hoffm.	Williamson (1994)	Allopatric	Palaeoendemic
Berkheya nivea N.E.Br.	Continuous	4–5	*Berkheya rehmannii* Thell. var. *rehmannii*	Roessler (1959)	Sympatric	Palaeoendemic
Berkheya rehmannii Thell.var. *rogersiana* Thell.	Continuous	10–14	*Berkheya rehmannii* Thell. var. *rehmannii*	Williamson *et. al* (1997)	Sympatric	Intermediate
Berkheya sp. nov. aff. *B. seminivea* Harv. & Sond.	Single locality	Unknown	*Berkheya seminivea* Harv. & Sond.		Sympatric	Neoendemic
Brachystelma dyeri K.& M.Balkwill	Disjunct	2	*Brachystelma gracile* E.A.Bruce	Balkwill & Balkwill (1988)	Sympatric	Palaeoendemic
Brachystelma sp. nov. aff. *B. longifolium* (Schltr.) N.E.Br.	Single locality	3–4	*Brachystelma longifolium* (Schltr.) N.E.Br.		Sympatric	Neoendemic
Cyphia bolusii E.Phillips	Disjunct	1–2	*Cyphia alba* N.E.Br.	Changwe & Balkwill (2003)	Sympatric ?	Palaeoendemic

Endemic Taxon	Distribution of endemic within BGB	Number of sub-populations	Possible nearest relative	Source	Distributions sympatric / allopatric	Palaeoendemic or Neoendemic
Dicoma swazilandica S.Ortiz	Continuous	Unknown	*Dicoma anomala* Sond.	Ortiz & Pulgar (2000)	Sympatric	Neoendemic
Dioscorea strydomiana Wilkin	Single locality	2	*Dioscorea elephantipes* (L'Hér.) Engl. or *D. sylvatica* Eckl.	Wilkin *et. al* (2010)	Sympatric/ allopatric	
Gladiolus serpenticola Goldblatt & J.C.Manning	Disjunct	5–6	*G. hollandii* L.Bolus or *Gladiolus crassifolius* Baker	Goldblatt and Manning (1998)	Sympatric	Palaeoendemic
Gymnosporia sp. nov.	Disjunct	3–4	*Gymnosporia buxifolia* (L.) Szyszyl.	Schmidt *et al.* (2002)	Sympatric	Palaeoendemic
Helichrysum sp. nov. aff. *H. albo-brunneum* S. Moore.	Single locality	1	*Helichrysum albo-brunneum* S. Moore.		Sympatric	Neoendemic
Indigofera crebra N.E.Br.	Continuous	6–7	*Indigofera ingrata* N.E.Br. or *Indigofera tristoides* N.E.Br.	Brown (1925)	Allopatric /Sympatric	Palaeoendemic
Macledium zeyheri subsp. *thyrsifolium* (Klatt) Netnou	Disjunct	2–3	*Macledium zeyheri* (Sond.) S.Ortiz subsp. *zeyheri*	Netnou & van Wyk (2003) & Pope (1991)	Sympatric	Neoendemic
Ocimum sp. nov.1 (Barberton species)	Continuous	9–10				Palaeoendemic
Ocimum sp. nov. 2	Single locality	2	*Ocimum obovatum* E.Mey. ex Benth. subsp. *obovatum* var. *obovatum*		Sympatric	Neoendemic

Endemic Taxon	Distribution of endemic within BGB	Number of sub-populations	Possible nearest relative	Source	Distributions sympatric / allopatric	Palaeoendemic or Neoendemic
Ozoroa barbertonensis Retief	Continuous	4–8	*Ozoroa albicans* R.Fern. & A.Fern. or *Ozoroa schinzii* R.Fern. & A.Fern.	Retief (1990)	Allopatric	Palaeoendemic
Ozoroa sp. nov.	Continuous	4–5	*Ozoroa barbertonensis*		Sympatric	Palaeoendemic
Protea curvata N.E.Br.	Continuous	4–5	*Protea laetans* L.E.Davidson or *Protea comptonii* Beard	Rourke (1980)	Allopatric/ Sympatric	Palaeoendemic
Salpinctium hirsutum T.J.Edwards	Disjunct	2	*Salpinctium natalense* T.J. Edwards	Edwards and Getliffe Norris (1989)	Allopatric	Palaeoendemic
Sartidia dewinteri J.Munday & L.Fish	Continuous	10–14	*Sartidia jucunda* (Schweick.) De Winter	Balkwill *et. al* (2011)	Allopatric	Palaeoendemic
Sclerochiton triacanthus A.Meeuse	Disjunct	2–4	*Sclerochiton ilicifolius* A.Meeuse	Vollesen (1991)	Allopatric	Palaeoendemic
Searsia pygmaea (Moffett) Moffett	Continuous	5–6	*Searsia pondoensis* Schönl.	Moffett (1999)	Sympatric	Palaeoendemic
Senecio sp. aff. *S. anomalochrous* Hilliard	Single locality	1–2	*Senecio anomalochrous* (Hilliard)		Allopatric	Neoendemic
Senecio sp. aff. *S. coronatus* (Thunb.) Harv.	Continuous	4–5	*Senecio coronatus* (Thunb.) Harv.		Sympatric	Neoendemic
Senegalia loetteri N.Hahn	Disjunct	6–7	*Senegalia rovumae* (Oliv.) Kyal. & Boatwr. or *Senegalia senegal* (L.) Britton	Hahn (2013)	Allopatric	Palaeoendemic

suggests that they are recently evolved and have not had enough time to extend their distributions.

The remaining endemics are geographically separated from their congeners at higher taxonomic levels, which suggests a relatively long time since speciation. In addition, they have relatively widespread distributions further suggesting sufficient time to extend their original range. Considering these factors together with the relatively long exposure of the serpentine outcrops, these taxa are classified as palaeoendemic. Some taxa (*Cyphia bolusi*, *Gladiolus serpenticola*, *Brachystelma dyeri*, *Senegalia loetteri*, *Sclerochiton triacanthus*, *Gymnosporia* sp. nov., and *Macledium zeyheri* subsp. *thyrsifolium*) have disjunct distributions (Figures 3.3 – 3.10), with populations separated by distances ranging from sixteen to 48 km, suggesting that they were once widespread, but some of the intermediate populations have since died out. The endemic taxa that have distributions separated from the distribution of their congeners (allopatric) are possibly older than those endemics that have overlapping distributions with their congeners. *Berkheya rehmannii* var. *rogersiana* is considered to be neither palaeoendemic nor neoendemic but an intermediate between the two based on the high levels of genetic separation between the endemic and its congener, which contrasts with the overlapping distribution of the two taxa (Williamson *et. al.* 1997). This is similar to the *Strepthanthus glandulosus* complex, found on serpentine in California, which is a widespread species that has become fragmented (similar to paleoendemism), with some of its members having speciated recently (Harrison 2013).

It is difficult to determine the status of *Dioscorea strydomiana*, *Ozoroa barbertonensis* and *Protea curvata* as there is little information available to help determine their closest relatives. In each case, one of the possible close relatives that have been identified is sympatric with the endemic species and the other possible relative is allopatric. This highlights the difficulty of using questionable close relatives based on morphological similarities rather than a full phylogenetic analysis (Bruchmann and Hobohm 2014). In order to accurately determine the true closest relative of an endemic taxon, phylogenetic analyses should be used preferably including genetic data. *Graderia* sp. nov. was excluded from the analysis in Table 3.9, as insufficient data exist to determine its status as an endemic and its distribution within the Barberton Greenstone Belt.

Conservation of endemics

A threat assessment has been completed for each serpentine endemic and the appropriate Red Data List (RDL) categories have been suggested (Appendix D). Since the publication of these data, four additional taxa have been recognised as being endemic to the serpentine outcrops of the Barberton Greenstone Belt. These taxa include *Dioscorea strydomiana*, *Graderia* sp. nov., *Ocimum* sp. nov. 2 and *Senegalia loetteri*. Specimens originally identified as *Brachystelma longifolium* are now considered to be an undescribed species and has been replaced on the list of endemics as *Brachystelma* sp. nov. aff. *B. longifolium*. *Inezia speciosa* is no longer considered to be endemic as it has been determined that the specimens collected on serpentine outcrops are not morphologically different from those found on the more northern localities. Although the vegetative parts of the southern populations appear silver and the northern ones golden, there does not seem to be any difference in the indumentum, nor any other character.

There are not enough data on the estimated population sizes of *Brachystelma* sp. nov. aff. *B. longifolium* and *Ocimum* sp. nov. 2 to make an accurate recommendation for an RDL status. However, based on extent of occurrence (EOO) and area of occupancy (AOO) and the lack of threat on *Brachystelma* sp. nov. aff. *B. longifolium*, this taxon could be classified as VuD1 + 2 (Table 3.10).

Brachystelma dyeri¸ *Gymnosporia* sp. nov. and *Gladiolus serpenticola* have each been recorded and collected from an additional locality since the original assessment. However, the recommended status has not changed as none of the criteria (population size, EOO and AOO) have been exceeded by the additional data.

Conclusion

Twenty-seven plant taxa have been found to be restricted to serpentine soils of the Barberton Greenstone Belt with a further five taxa restricted to serpentine soils and other related ultramafic soils. Higher than expected levels of endemism, based on Bykov’s Index of Endemicity, have been calculated for serpentine outcrops. Each outcrop has a distinctive composition of endemic species, with fewer than 24% of the endemic taxa shared between any two sites. Eight of the serpentine endemics have been found to occur on only a single outcrop.

Table 3.10: Current and recommended conservation status (according to the IUCN categories and criteria, IUCN (2001) of serpentine endemics not included in the original assessment (Appendix D)

Taxon	Estimated population size	Estimated future decline (%)	Threats and potential threats	Extent of occurrence (EOO) (km^2)	Area of occupancy (AOO) (km^2)	Number of subpopulations	Recommended status	Current status in RDL
Brachystelma sp. nov. aff. *B. longifolium*	unknown	0	None known	2–5	1–2	3–4	DD	Not listed
Dioscorea strydomiana	200	20–30	Burning, mining and collection for medicinal use	2–5	1–2	2	CR B1ab(v)+2ab(v), C1	CR B1ab(v)+ 2ab(v);C1 (Wilkin *et al*. 2010)
Ocimum sp. nov. 2	unknown	5–15	Afforestation	6–8	3–4	2	DD	Not listed
Senegalia loetteri	200 +	0–5	Mining claims	30–40	6–10	6–7	Vu D1 + 2	Not listed

Most of the endemic taxa exhibit long-range dispersal, but different kinds of research are needed to determine the levels of gene-flow between isolated populations. Endemic taxa with one or two localities and/or are morphologically similar to their congeners are considered to be neoendemic. Taxa with relatively widespread, larger populations that are morphologically distinct from their congeners are thought to be palaeoendemic.

Detailed study of a Californian serpentine endemic, *Thlaspi montanum* var. *montanum*, revealed that individuals collected from non-serpentine areas will accumulate nickel when grown on nickel rich serpentine soils (Boyd and Martens 1998). Therefore, the authors conclude that nickel hyperaccumulation is a constitutive trait of *Thlaspi montanun*. This type of detailed research is necessary to answer questions about the Barberton Greenstone Belt serpentine endemic *Senecio* sp. nov. aff. *S. coronatus.* These could include:

- are the hyperaccumulating and non-hyperaccumulating individuals interfertile?
- is there a genetic difference between these individuals / populations?
- does this taxon represent one of the first stages in the evolution of genetically and/ or morphologically distinct populations that could result in a new species?
- could there be a level of racial differentiation that has taken place representing early stages of diversification?

The extensive analysis of DNA of endemic species and leaf analyses of nickel and other elements that has been done on the Brazilian and Cuban endemics has not been done on South African edaphic endemics. These analyses are necessary to identify evolutionary processes that resulted in endemism and hyperaccumulation on serpentine soils.

The plant species endemic to serpentine and ultramafic outcrops of the Barberton Greenstone Belt contribute significantly to the endemicity of the Barberton Centre of Endemism and Mpumalanga Province. The high levels of endemicity and unique composition of endemic species found at each site gives these outcrops high conservation priority.

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Chapter 4

Species richness and diversity of a terrestrial insular environment: serpentine of the Barberton Greenstone Belt, South Africa

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Species richness and diversity of a terrestrial insular environment: serpentine of the Barberton Greenstone Belt, South Africa

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The serpentinite outcrops of the Barberton Greenstone Belt are considered to be amongst the oldest in the world and have been exposed for over 50 million years. Of the 30 large outcrops in eastern Mpumalanga, South Africa, seven were selected for detailed comparison of species richness and diversity with those of surrounding non-serpentine vegetation. Various non-parametric species richness estimators were calculated to predict the species richness of each site and these indicated that four serpentine outcrops have higher and three have lower species richness than the surrounding non-serpentine areas. The Shannon, Simpson's and Fisher's alpha indices of diversity were calculated for each site on and off serpentine and compared. The diversity values calculated show similar patterns to those of the species richness estimates. Significant differences in species composition between floras on and off serpentine were estimated using Sørenson's Index of similarity. The serpentine outcrops of the Barberton Greenstone Belt show relatively high diversity when compared to some other serpentine outcrops around the world.

Key words: Diversity indices, *EstimateS*, Modified-Whittaker plots, serpentine, species accumulation curves, species richness estimation.

INTRODUCTION

The diversification of habitats that can flow from differences in substrates such as limestones, dolomite, shale, gypsum and serpentine becomes the stimulus for evolutionary diversification by speciation. These azonal substrates have been found to harbour unique plant associations, including endemic species, and foster morphological and physiological modifications of plants as well as unusual distributions of taxa (Kruckeberg, 1986). Serpentine soils often support a flora distinct from the surrounding vegetation due to phytotoxic concentrations of heavy metals such as nickel and chromium and high ratios of magnesium to calcium (Roberts and Proctor, 1992). In addition, serpentine outcrops support endemics that usually occur in a few small populations and are confined to single or few localities (Kruckeberg and Rabinowitz, 1985), which have conservation and scientific value. A 'Resolution' passed by delegates of The First International Conference on Serpentine Ecology held in 1991 supports the conservation of the vegetation of serpentine areas worldwide (Kruckeberg, 1992).

These serpentine outcrops provide a classic system for understanding the origins and maintenance of plant diversity. It is usually predicted that the insularity of serpentine outcrops will result in low plant species richness (α diversity) but high ß diversity, that is, spatial

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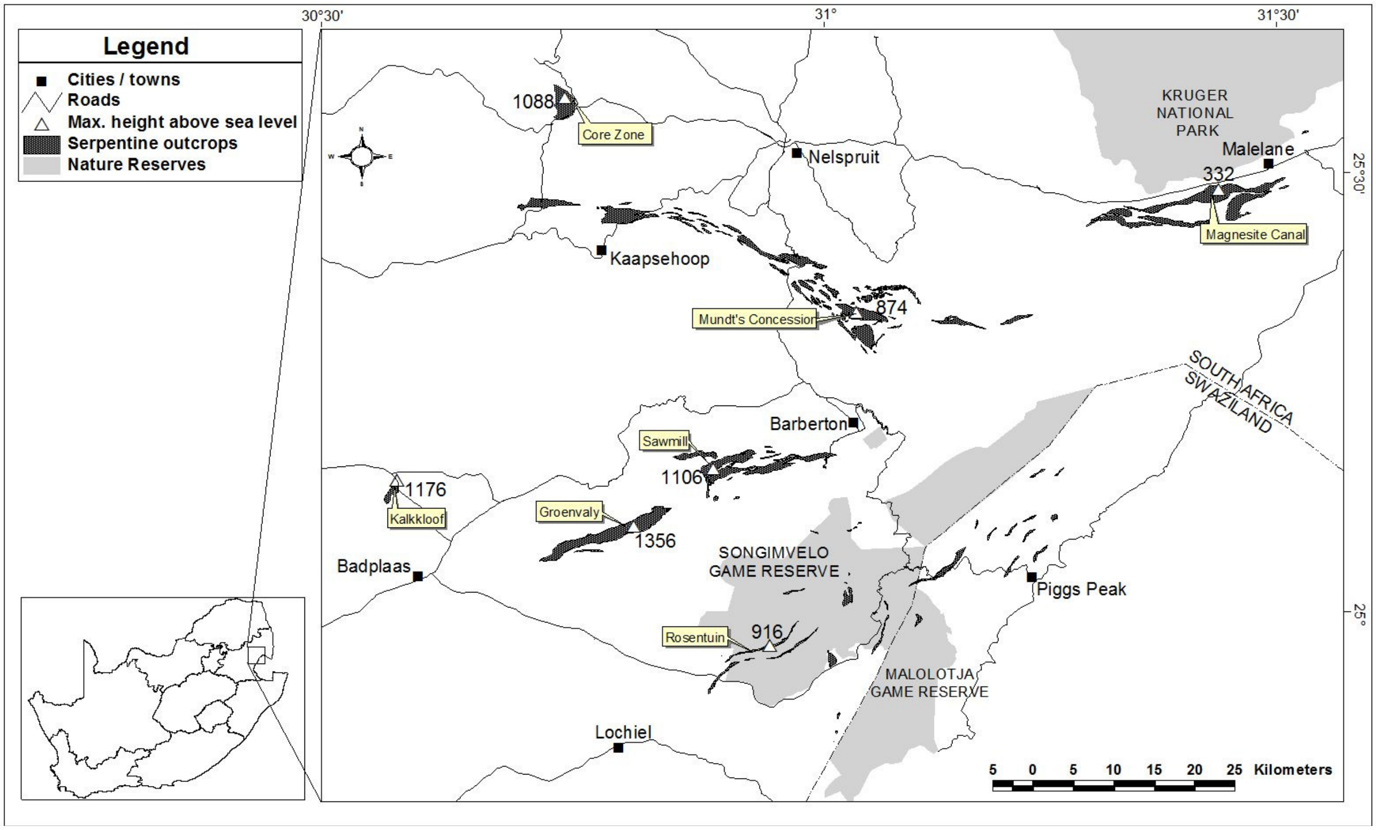

Figure 1. Map indicating serpentine outcrops of the Barberton Greenstone Belt in Mpumulanga, South Africa. Survey sites are identified by callout labels. Map was prepared using data provided by the Chief Directorate: Surveys and Mapping, Department of Land Affairs, Republic of South Africa.

variation in species composition among regions (Harrison and Inouye, 2002). In contrast, the serpentine of the Siskiyou and Wenatchee Mountains in the United States of America were shown to support higher species richness than the surrounding non serpentine areas (Proctor and Woodell, 1975). The high species richness recorded on the serpentine outcrops of South Central Africa (Wild and Bradshaw 1977) suggests that have been exposed sufficiently long to re-establish diversity following the geological changes associated with this exposure (Brown 1988). It is suggested that the Barberton Greenstone Belt is between 3.3 and 3.4 billion years old and is considered to be the second oldest greenstone belt after the Isua Greenstone Belt in Greenland (Condie, 1981). The rocks of the Barberton Greenstone Belt are thought to have been exposed for about 50 million years (Anhaeusser *pers. comm.*). This extended time of exposure could have resulted in the establishment of high diversity on the serpentine sites of the Barberton Greenstone Belt (Brooks, 1987). The serpentines in central Cuba are believed to have been exposed for only around 1 m.y. (Reeves et al., 1996) and those of California were exposed between the Late Pliocene to Early Quaternary (Raven and Axelrod, 1978).

The term ‘serpentine’ is often applied to ultramafic rocks in general, but in this study it is used in its strict sense and applied to rocks derived from serpentinite. There are about 30 large (that is, >1 km^2) serpentine outcrops, with the largest about 19 km^2, and many smaller outcrops (from 0.1 km^2) in eastern Mpumalanga (Figure 1), which form part of the Barberton Greenstone Belt (Ward, 2000). These outcrops are located in an inverted equilateral triangle centred on Barberton extending to Malelane in the east and to Badplaas in the south. Some outcrops are separated from others by up to 20 km (Balkwill et al., 1997). The outcrops occur in mountainous areas and are heterogeneous in altitude, slope, soil depth and other topographic features. The serpentine vegetation falls within the Mixed Lowveld Bushveld, Sour Lowveld Bushveld and North-eastern Mountain Grassland vegetation types (Low and Rebelo, 1996). A more recent classification recognises the unique character of the serpentine vegetation of the Barberton Greenstone Belt and labels it as Barberton Serpentine Sourveld (Mucina and Rutherford, 2006). In addition, the vegetation of these serpentine outcrops forms part of the Barberton Centre of Endemism (van Wyk and Smith, 2001).

As on other serpentine outcrops around the world, the flora of the Barberton Greenstone Belt seems to be distinct from the surrounding vegetation, although this has yet to be quantified. These outcrops do, however, support a large number of relatively rare endemic species most of which have red data status and thus are of conservation interest (Williamson and Balkwill, 2006).

Ecologists have long been intrigued by patterns of spatial and temporal variation in the diversity of plants and animals, and there are now a large array of indices and models that have been developed for quantifying species diversity (Magurran, 1988). Determining species diversity for selected areas and an understanding of the factors that regulate species diversity are central to the science of conservation (Spellerberg, 1992; Cowling, 1983). Species diversity consists of two components, namely: species richness, which is the number of species present, and evenness or the relative abundances of species. A "diversity index" is thus a single statistic that incorporates both richness and evenness into a single value (Magurran, 2004). An area is said to have high diversity if it has many species and their abundances are fairly even. Diversity is low when there are few species with uneven abundances (George and Hanumara, 1989). Selecting an appropriate measure of diversity depends largely on the question the index is being used to answer, the component of diversity being measured and whether the index is simple to use and understand (Williams et al., 2005).

In most studies, the diversity of serpentine vegetation has been expressed in terms of endemic species richness only (Selvi, 2007) and the focus for determining the conservation value of serpentine outcrops has been in terms of the number of endemics and the percentage endemism (Brooks, 1987). Few authors have described the diversity of the vegetation of various serpentine outcrops and the factors affecting diversity, which is essential for determining conservation priorities for these outcrops. The species richness of serpentine outcrops in Tuscany (Italy) was used to identify centres of diversity and endemism and to evaluate the environmental factors that affect the abundance of these plant communities (Chiarucci et al., 2001). There is evidence that the species richness of the Tuscan serpentine outcrops decreases with increasing isolation of outcrops and decreasing surface area (Selvi 2007). These data were used to develop conservation programs for the serpentine outcrops in Tuscany (Italy). The diversity of plant species on ultramafics and nearby schist substrates in New Zealand was analysed to determine trends in diversity in relation to altitude (Bastow Wilson et al., 1990). The serpentine areas of California are considered to support one of the world's richest serpentine floras. The vegetation of the Californian serpentines shows higher diversity on the more continuous serpentine areas and lower diversity on the areas which are naturally more patchy (Harrison, 1997). The high endemic diversity of the Californian serpentine outcrops is determined by high precipitation and high soil Mg/Ca ratios (Grace et al., 2007).

Describing the species diversity of metallophytes (organisms with the ability to tolerate or avoid the toxic effects of metals) is essential to understand and conserve serpentine outcrops (Whiting et al., 2004). The serpentine vegetation of the Barberton Greenstone Belt is considered to have conservation value in an area that is heavily utilised for *Eucalyptus* and pine plantations (Balkwill et al., 1997) and thus for effective conservation and environmental management the baseline species richness and diversity needs to be described.

Numerous theories have been advanced to account for patterns of species diversity across edaphic boundaries such as that of serpentine outcrops and their surrounding vegetation. One is that which edaphic 'islands' have re-established diversity after a sufficiently long period after perturbation (Brown, 1988). In contrast it has been suggested that the insularity of serpentine outcrops will result in low plant species α diversity (Harrison and Inouye, 2002). As the first part of a wider study to determine whether serpentine areas have high conservation value in terms of diversity, this paper attempts to test the hypothesis that the vegetation of the serpentine outcrops of the Barberton Greenstone Belt has higher species richness and diversity than the adjacent non-serpentine areas. The differences in species richness and diversity between serpentine and adjacent non-serpentine areas of the Barberton Greenstone Belt are quantified using a range of applicable richness and diversity indices. These differences would determine whether the vegetation of the Barberton Greenstone Belt supports either of the predictions made regarding species diversity patterns caused by edaphic variation. This paper also aims to determine whether all serpentine outcrops of the Barberton Greenstone Belt have similar plant species richness and diversity and to provide preliminary data to help determine the factors contributing to any measured differences in diversity. The species richness and diversity values calculated could provide a strong case for including serpentine vegetation in the planning for conservation of the vegetation of the southern portion of Mpumalanga Province. Land-use decisions are most often based on variation and species diversity on the scale of landscapes or 'park-sized' units and have significant impact on the long-term future of biodiversity (Colwell and Coddington, 1994). Lastly, this paper aims to compare the diversity of the serpentine sites of the Barberton Greenstone Belt to other serpentine sites around the world.

MATERIALS AND METHODS

There are approximately 30 large serpentine outcrops in the Barberton Greenstone Belt surrounded by several very small outcrops. Of the large ones, seven were selected to be studied in detail. This selection was made so that presumably the full range of variation in terms of size, altitude and position is accounted for and excluded the Swaziland outcrops and other outcrops which have previously been studied in detail (McCallum, 2006; Changwe and Balkwill, 2003; Williamson, 1994).

Four modified-Whittaker plots of 1000 m^2 with ten 1 m^2, two 10 m^2 and a single 100 m^2 subplot within the largest plot (Stohlgren et al., 1995), were positioned on each serpentine site and adjacent non-serpentine area. These plots were positioned at the base, on the mid-slopes and crests of hills. This provided data from 56 modified-Whittaker plots for analysis. Within each plot and subplot, the numbers of taxa present and their relative abundances were recorded.

Species richness

There are two main methods of expressing estimates of species richness. The first method computes numerical species richness, which is the number of species per specified number of individuals or biomass. The second method describes species density, which is the number of species per specified collection area or unit. For the purpose of comparison between serpentine vegetation and adjacent non-serpentine vegetation and to compare the species richness of the sampled serpentine outcrops to other areas, species richness has been calculated with both methods using the data collected in the Modified-Whittaker plots.

For the measurement and comparison of species richness, the use of taxon sampling or accumulation curves is recommended (Colwell and Coddington, 1994; Gotelli and Colwell, 2001) recommend. Public-domain software, EstimateS (Version 7.5, Colwell, 2005) was used to compute randomised species accumulation curves with 95% confidence intervals for each serpentine and adjacent non-serpentine site. To eliminate any variation in curve shape due to sampling error or heterogeneity among sampled units (Colwell and Coddington, 1994); EstimateS randomises the sample order and calculates the mean and standard deviation of $S(n)$ (S represents observed species richness and n the number of samples) computed for each value of n.

Determining species richness from samples invariably underestimates the total species richness of the plant communities, and the total enumeration of species richness within a large study area is generally not feasible (Chiarucci et al., 2003). Non-parametric estimates of species richness have been shown to approach the actual species richness values; however, they are still thought to underestimate total species richness (Chiarucci et al., 2003). EstimateS was used to calculate seven non-parametric estimators of total species richness. Non-parametric methods for estimating species richness were used in preference to fitting parametric models of relative abundance as the non-parametric methods require no assumption about community structure (Colwell and Coddington, 1994).

Chao 2 is simple estimator of absolute number of species in an assemblage, which is calculated on presence/absence data taking into consideration the distribution of species amongst samples (Chao 1984). Jackknife 1, a first-order jackknife estimator, employs the number of species that occur in only a single sample and can be calculated to include the number of species in two samples (Jackknife 2) (Magurran, 2004). The bootstrap estimator used, is based on the proportion of quadrats containing each species and requires only incidence data (Smith and van Belle 1984). Plotting the performance of the estimator on a species accumulation curve illustrates the rate at which new species are found (Magurran, 2004) and demonstrates the performance of the estimator and differences in relative abundance as sample size increases (Williams et al., 2007). Species richness estimators may form the basis of community comparison, providing a convincing asymptote is reached (Magurran, 2004).

The observed species richness calculated for each site was correlated with various physiographic factors using Multiple Regression Analysis to determine the factors affecting species richness. These factors include: area of outcrop (km^2); mean elevation (meters above sea level); the latitude of the outcrop; available nickel and chromium; and the calcium to magnesium ratio of the soil.

Species diversity

Indices of diversity or heterogeneity incorporate measures of species richness and evenness into a single value and are based on the proportional abundance of species in a sample (Magurran, 1988). Three diversity indices that is, Simpson's Index expressed as lambda (λ), the Shannon Index (H') and Fisher's Alpha, were selected to quantify and compare the diversity of the vegetation of the serpentine outcrops and the surrounding non-serpentine areas.

Simpson's Index is a diversity index which is weighted by abundances of the commonest species and measures the probability that two randomly selected individuals from a sample will belong to the same species (Magurran, 2004). A rarely cited function is $-\ln \lambda$, is preferred by Williams et al. (2005) as it does not represent a probability, but a single diversity statistic that increases as diversity increases and gives numbers of similar magnitude to those of the Shannon index (Magurran, 2004). Simpson's index of diversity was selected as it provides a good estimate of diversity at relatively small sample sizes, will rank assemblages consistently and confidence limits can be attached to it (Magurran, 1988).

The Shannon index (H') is sensitive to sample size (Magurran, 2004) as it assumes that individuals are randomly sampled from an infinitely large community and that all species are represented in the sample (Pielou, 1975). The Shannon index is, therefore, one of the information statistics which tend to emphasize the species richness component of diversity (Magurran, 2004) as it measures the average degree of 'uncertainty' in predicting to what species individuals chosen at random will belong. It was selected for this study due to the simplicity of calculating H' and its widespread use as a benchmark measure of biological diversity. Large values of H' (high diversity) indicate a greater uncertainty in correctly predicting the identity of the next species chosen at random.

Fisher's alpha (α) describes the relationship between the number of species and the number of individuals of those species with a logarithmic distribution. It represents a species abundance model that could shed light on the processes that determine the biological diversity of an assemblage (Magurran, 2004). Alpha is low when the number of species is low and, therefore, smaller samples have low values of α. The index is less affected by the abundance of the rarest or commonest species than either H' or λ respectively and depends more on the number of species of intermediate abundance.

In order to compare the diversity calculated for different assemblages statistically, Magurran (2004) suggests using a jackknifing method. This method involves producing a series of 'pseudovalues' and the means of these pseudovalues represent the best estimate of the diversity statistic. Approximate confidence limits can then be attached to the estimate. This was done to determine whether the diversity of each serpentine site is significantly different to that of the adjacent non-serpentine area.

RESULTS AND DISCUSSION

Species richness

The number of recorded species per specified collection area or species density, mostly represented as species per m^2, is a commonly used measure of species

Table 1. Number of species recorded in 1000 m^2 (0.1ha) for each Barberton Greenstone Belt serpentine outcrop and adjacent non serpentine plots including the Swaziland serpentine outcrops. For all sites the differences in species density between serpentine sites and their adjacent non-serpentine areas are not statistically different as P>0.05 (◆, data not available).

Site	Species density (no of species per 0.1ha)				t value and probability of difference
	Serpentine outcrops		Adjacent non-serpentine area		
	Mean ± SD	Range	Mean ± SD	Range	
Sawmill	74.5±6.6	67-83	78.0±10.8	66-92	t(cal) = -0.668, P>0.05
Rosentuin	71.8±13.4	55-87	69.8±6.6	64-78	t(cal) = 0.215, P>0.05
Mundt's	65.3±4.7	62-71	67.3±4.7	63-73	t(cal) = -0.627, P>0.05
Magnesite	73.3±7.5	67-82	54.0±9.4	44-66	t(cal) = 2.339, P>0.05
Kalkkloof	83.3±7.3	73-90	82.8±9.3	71-93	t(cal) = 0.099, P>0.05
Groenvaly	80.3±9.4	69 - 92	72.5±13.4	55 - 86	t(cal) = 0.847, P>0.05
Core Zone	74.8±5.1	70 - 82	84.3±5.7	79 - 92	t(cal) = -1.956, P>0.05
Dunbar[1]	29.0±4.3	◆	31.7±2.9	◆	t(cal) ◆, P>0.05
Forbes Reef [2]	81.7±5.6	◆	73.0±2.3	◆	t(cal) ◆, P>0.05
Malolotja[2]	76.3±11.2	◆	54.5±6.5	◆	t(cal) ◆ P>0.05
Motjane[2]	43.5±0.5	◆	59.0 (single)	◆	t(cal) ◆, P>0.05

[1]Changwe and Balkwill (2003). [2]McCallum (2006).

richness (Magurran, 1988). There is considerable variation in the mean number of species per 0.1ha between the selected sample sites, ranging from 29 to 65 species per 0.1 ha (Table 1). Although the number of species per 0.1 ha for four of the sites is shown to be considerably higher than that of the adjacent non-serpentine areas and for two of the sites the species number is considerably lower, these differences were found to be not significantly different (P> 0.05) when compared using paired T-tests. However, the Magnesite, Corezone, Malolotja and Motjane sites P < 0.2, which suggests a true biological difference.

Randomised species accumulation curves for species recorded in plots for each serpentine outcrop and the adjacent non-serpentine areas per sample are shown in Figure 2. The species accumulation curves in Figure 3 represent a comparison of species richness where the effect of density has been removed by pooling individuals of the same species. The species richness values were plotted together with the 95% confidence intervals for each curve as Colwell et al. (2004) conclude that where confidence levels do not overlap the differences in species richness are significant at *P*<0.05. The graphs of numerical species richness (*S*) for each site (Figure 3) indicate that sampling was slightly inadequate as some of the curves are approaching an asymptote, but most curves are increasing at a rate of 0.08 to 0.12 species per individual at the end of each curve. At this level of sampling, results show that the non-serpentine vegetation has higher species richness than the serpentine vegetation for two of the seven sites and only Groenvaly serpentine shows significantly higher species richness than the surrounding non-serpentine area (P<0.05). Although the species richness on and off serpentine of most sites is quite similar, the percentage of species shared is 33% or less for all sites (Table 2), indicating a large change in species composition across soil substrates.

The differences in the number of shared species on and off serpentine (Table 2) could be due to the different vegetation types surrounding the serpentine outcrops, and possibly show that grassland species are less able to colonise serpentine soils than bushveld species. The graphs in Figures 2 and 3 differ because the relative abundances (measured as the mean number of individuals per sample) are greater in the serpentine vegetation than the non-serpentine vegetation for six sites. The relative abundance for the serpentine of Core Zone is considerably lower (9.9 individuals per species) than the adjacent non-serpentine (12.7 individuals per species).

The dendrogram (Figure 4a) prepared from the Sorensøn's Similarity Index comparing species composition between serpentine sites (Table 3) groups outcrops in a way that correlates with geography and altitude to a large extent and

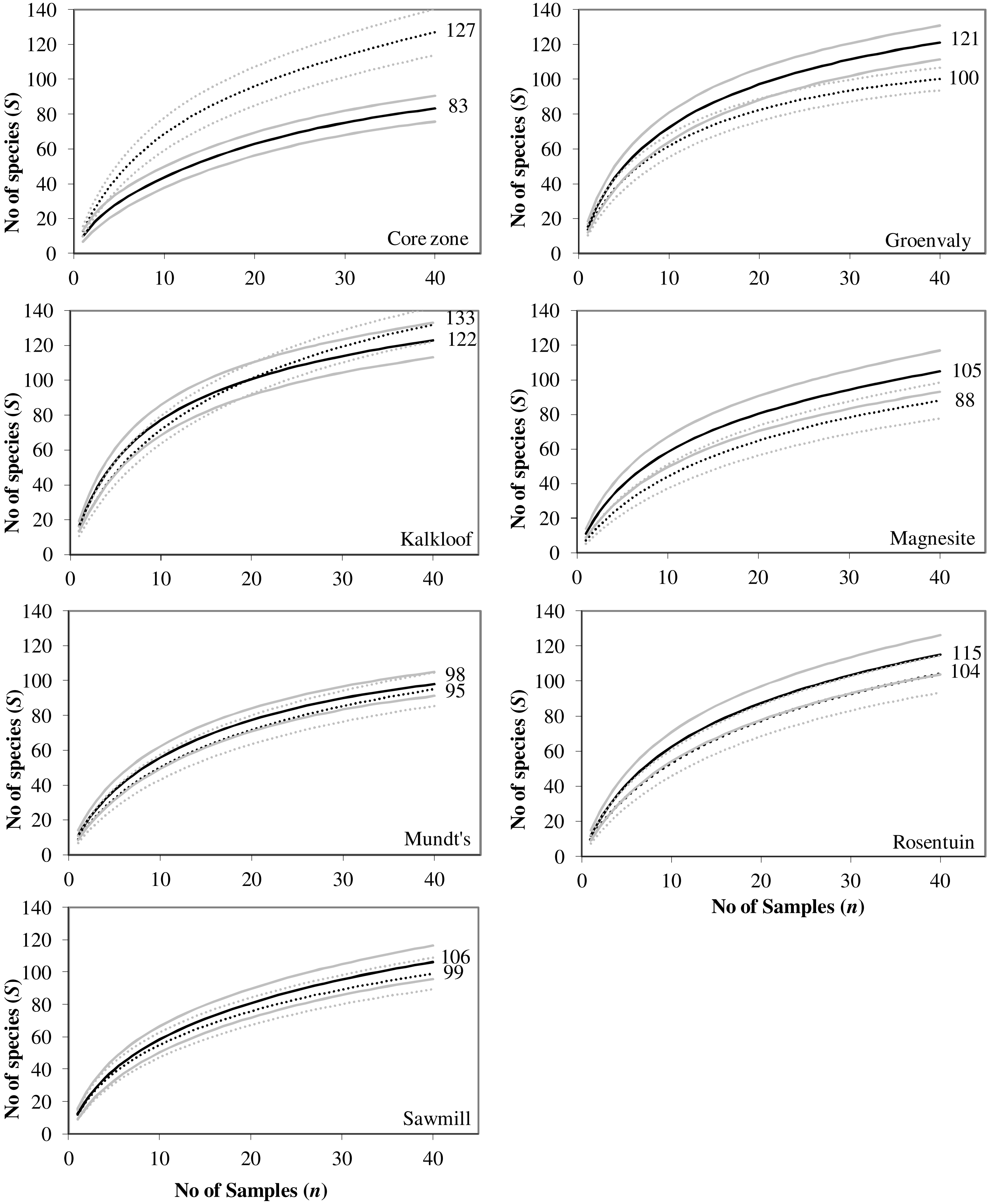

Figure 2. Observed species richness of selected serpentine sites (solid lines) and adjacent non-serpentine areas (broken lines) plotted against number of 1 m^2 samples. The overall value of S for the randomly pooled samples is labelled at the end of each curve and 95% confidence intervals are shown in grey. All graphs are scaled to the same x and y-axes values.

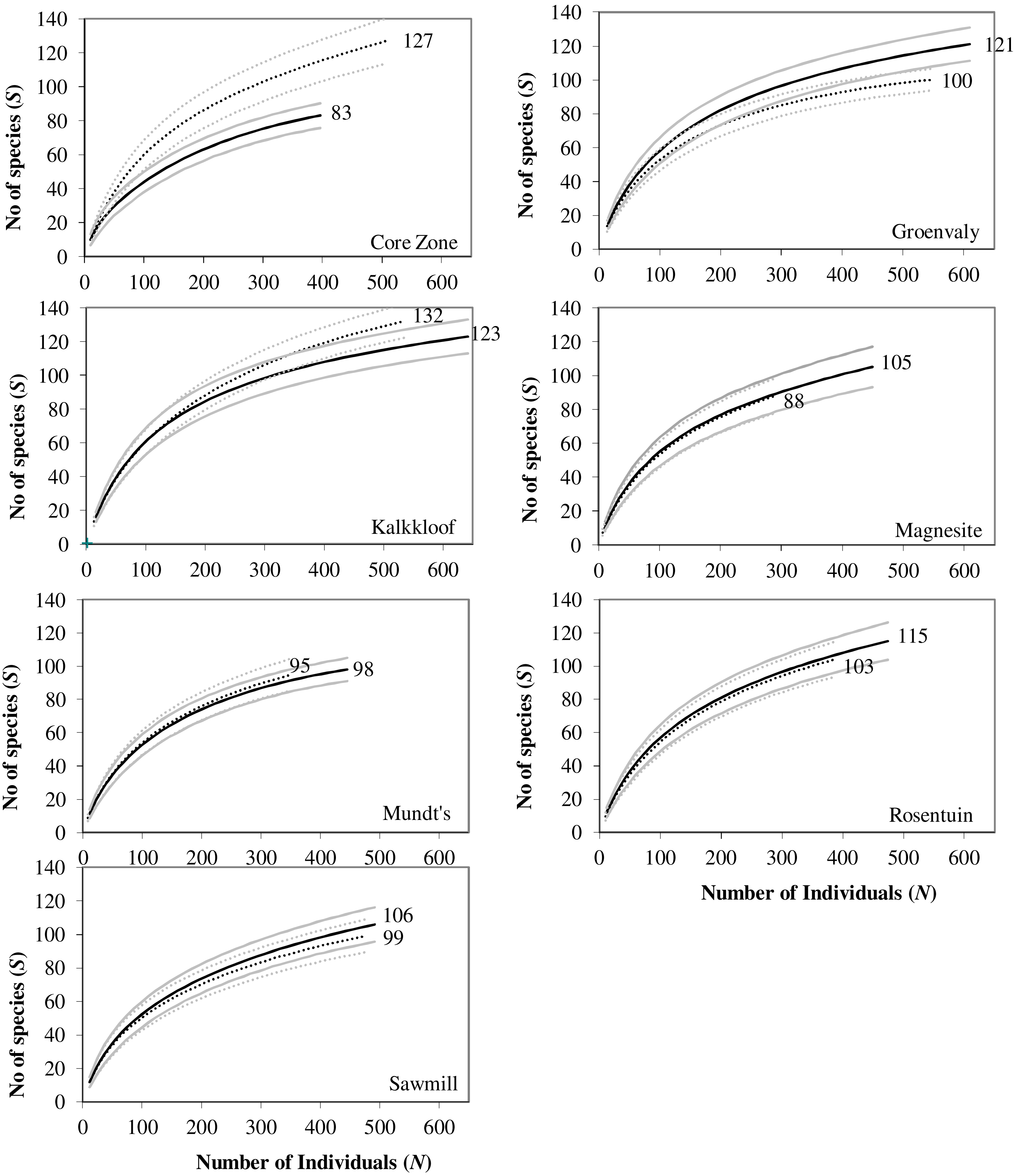

Figure 3. Observed species richness of selected serpentine sites (solid lines) and adjacent non-serpentine areas (broken lines) plotted against individual numbers (indicating sampling intensity). The overall value of S for the randomly pooled samples is labelled at the end of each curve and 95% confidence intervals are shown in grey. All graphs are scaled to the same x and y-axis values.

Table 2. Sørenson's coefficient of similarity for serpentine and non-serpentine areas for each site of the Barberton Greenstone Belt, expressed as percentages, correlated with the vegetation type within which the outcrop falls.

Site	% Similarity	Vegetation type (Low and Rebelo, 1996)	Vegetation type (Mucina and Rutherford, 2006)
ore-Zone	21.69	Sour Lowveld Bushveld	Legogote Sour Lowveld Bushveld
Groenvaly	26.42	Sour Lowveld Bushveld	Barberton Montane Grassland
Magnesite Canal	28.03	Mixed Lowveld Bushveld	Kaalrug Mountain Bushveld
Kalkkloof	28.18	North Eastern Mountain Grassland	KaNgwane Montane Grassland
Sawmill	28.93	Sour Lowveld Bushveld	Barberton Montane Grassland
Dunbar[1]	31.2	North Eastern Mountain Grassland	Swaziland Sour Bushveld
Mundt's Concession	33.03	Sour Lowveld Bushveld	Granite Lowveld
Rosentuin	33.33	North Eastern Mountain Grassland	Swaziland Sour Bushveld
Malolotja[2]	35.7	North Eastern Mountain Grassland	Barberton Montane Grassland
Forbes reef[2]	42.2	North Eastern Mountain Grassland	Barberton Montane Grassland
Motjane[2]	74.7	North Eastern Mountain Grassland	Barberton Montane Grassland

[1]Changwe and Balkwill (2003); [2]McCallum (2006).

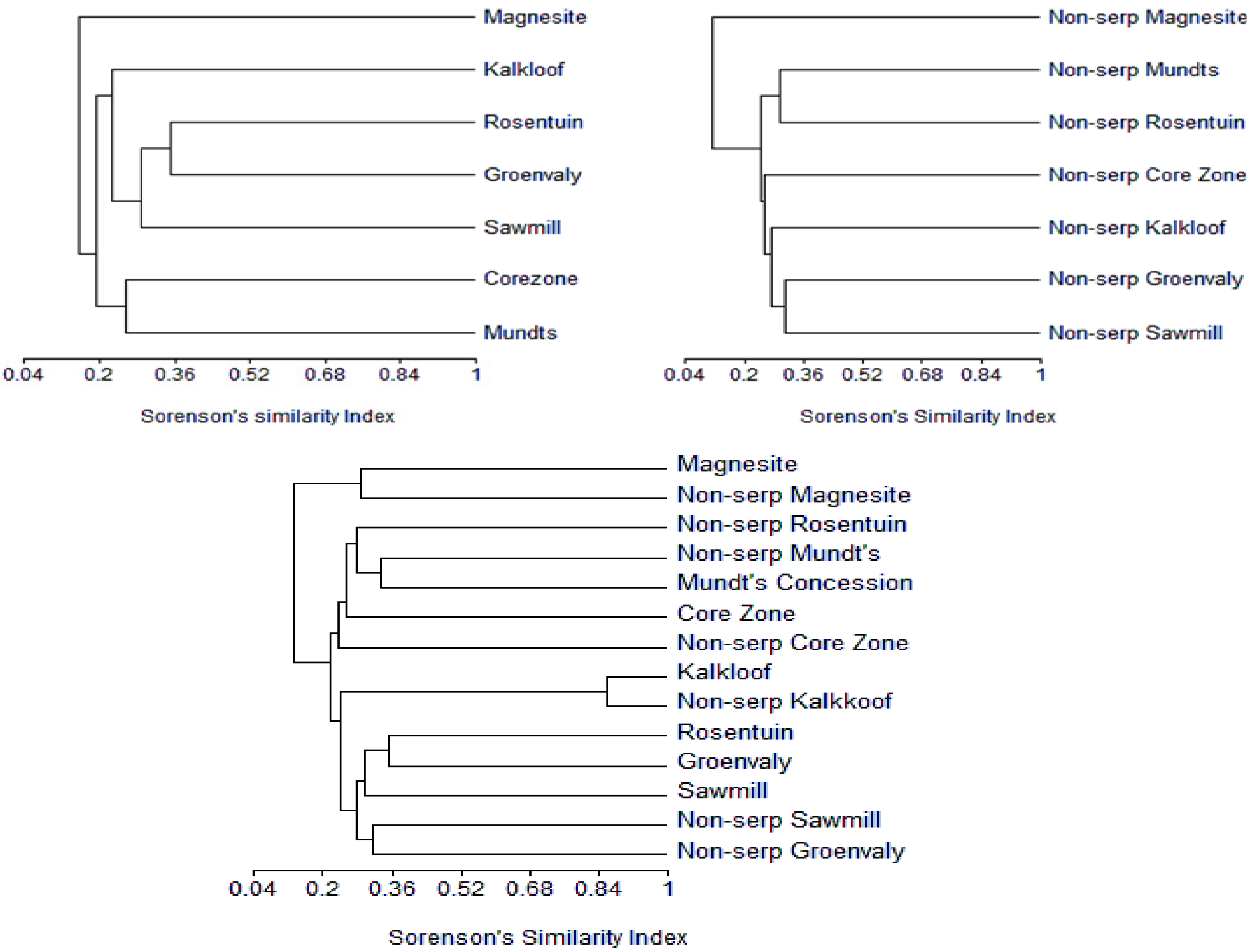

Figure 4 Dendrograms representing the Sørenson's coefficient measuring the similarity between sampled serpentine sites, the adjacent non-serpentine samples and a combination of both serpentine and non-serpentine samples. Dendrograms were drawn online using the UPGMA algorithm.

Table 3. Sørenson's coefficient of similarity measuring the degree of similarity between sampled serpentine sites, expressed as percentages.

Site	Core zone	Groenvaly	Kalkkloof	Magnesite Canal	Mundt's Concession	Rosentuin
Groenvaly	17.7	-				
Kalkkloof	18.3	25.4	-			
Magnesite	*14*	18.9	9.4	-		
Mundt's	25.4	27.5	15.7	21.1	-	
Rosentuin	17	**35**	24.9	15.9	22.5	-
Sawmill	16.2	34.1	17	*14*	21.1	23.3

indicates that Rosentuin and Groenvaly sites are most similar.

The high similarity between these two outcrops is possibly due to their proximity and that they seem to be part of the same group of outcrops (Figure 1). Although the Magnesite site has similar observed species richness to most of the other sites, it shares very few taxa with the other sites. This site is part of a spatially isolated group of outcrops that are found at a much lower altitude (Figure 1) than the other outcrops and are surrounded by a very different vegetation type (Table 2). The dendrogram representing the similarity between the non-serpentine samples (Figure 5b) also isolates the Magnesite sample from the other groups, but groups the other samples quite differently showing the Groenvaly sample most similar to the Sawmill sample.

When the data are combined, the dendrograms show that the serpentine sites that are geographically clustered are more similar in species composition to each other than to their adjacent non-serpentine vegetation and form two distinct groups of outcrops. The geographically isolated serpentine sites are seen to be more autochthanous. Of interest is that the Kalkloof serpentine and non-serpentine samples are the most similar to one another of any pair. This serpentine site has the lowest nickel and chromium levels of all other sites.

In many cases, observed species richness underestimates actual species richness as it is likely that not all species were sampled in the plots. Therefore, non-parametric methods, which estimate the lower bounds of predicted total species numbers, are used to estimate species richness (Colwell, 2005). The Chao and ICE estimators over-estimated species richness at the beginning of a sample (because they are strongly affected by the high number of singletons and rare species present in the first few samples) making them unstable initially and were therefore discounted. The second- and first-order Jack knife estimators (Jack 2 and Jack 1) consistently generate the highest estimates and the Bootstrap the lowest estimates for all samples (Figure 5). Jack 2 seems to approach an upper limit for the number of species likely to be found at each site and Bootstrap provides the lower-bound estimate of species richness. For six of the seven sites (excluding Mundt's Concession), the estimators tend to rise in parallel with the observed species accumulation curve and do not reach an asymptote, but show the same trends in terms of comparisons between serpentine and non-serpentine areas at each site.

The upper and lower bound predictors of expected species richness show the same trend, as the observed species richness, for the Groenvaly site with the non-serpentine curve having reached an asymptote but the serpentine curve still increasing slightly (Figure 5). This suggests that the actual difference in species richness may be even greater than measured here. The upper bound species richness estimator, Jack 2, representing the upper bound of species richness, predicts that differences between serpentine and non-serpentine are greater than that suggested by the observed species richness. It suggests significant difference ($P<0.5$) in five of the sites and biological difference ($P<0.02$) for the Rosentuin and Sawmill sites (Table 4).

Species diversity and evenness

The Simpson's, Shannon and Fisher's alpha diversity indices were used as they highlight different aspects of diversity in a plant community (Magurran, 2004). It was thus expected that the different indices used would possibly highlight differences between the serpentine and non-serpentine vegetation other than those shown by the species richness values. However, the species diversity indices calculated for each site show similar trends to those established by the species richness estimators that is, serpentine areas that show significantly higher species richness than non-serpentine areas also show significantly higher species diversity (Table 4).

The evenness (J') values are very similar for serpentine and non-serpentine areas, which suggests that the species richness component, of the diversity indices, contributes significantly to the differences between individual serpentine and non- serpentine areas and not their relative abundance based on the evenness (J') index. The Corezone site is one exception as the evenness (J') calculated for the non-serpentine area is significantly higher than that for the serpentine outcrop. This suggests that the non-serpentine vegetation here has more species that are equally abundant, that is, there is higher dominance of certain species on the serpentine outcrop. The evenness values for the other sites is contradicted by the differences in relative abundance

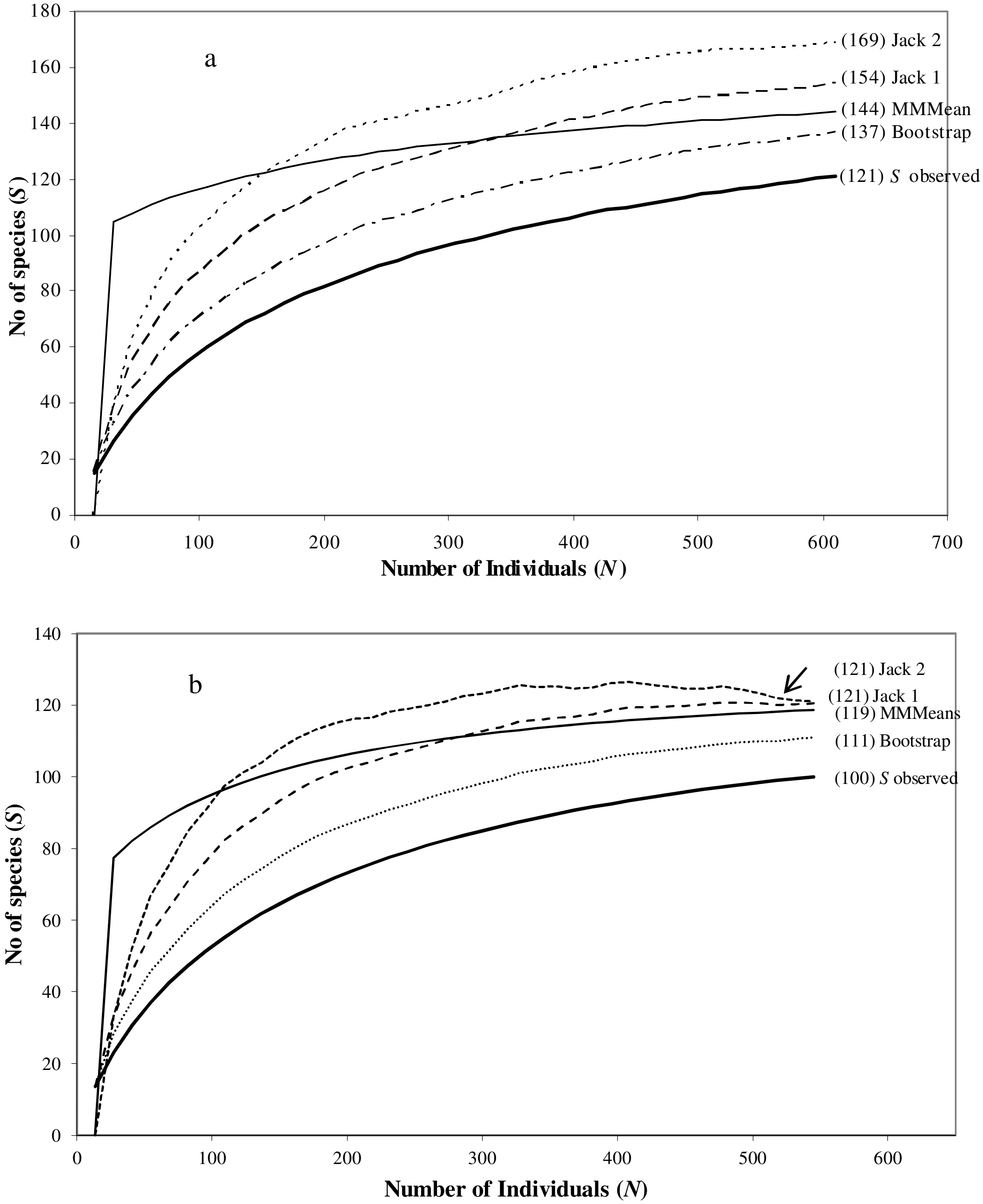

Figure 5. The performance of the non-parametric species richness estimators compared with the species accumulation curve (*S* observed) for the serpentine outcrop Groenvaly (a) and the adjacent non serpentine areas (b). The overall value of S for the randomly pooled samples is labelled at the end of each curve (in brackets).

exhibited by Figure 3, which suggest that six of the seven serpentine sites (excluding the Corezone site) have higher relative abundance than the adjacent non-serpentine areas.

The evidence (Table 4) shows that the serpentine outcrops with high species diversity also support a higher number of singletons. This supports the observation that assemblages with high species richness tend to have low evenness (Weiher and Keddy, 1999). These results suggest that the evenness index (J’) selected is not sensitive enough to differentiate between the different assemblages sampled.

Table 4. Richness estimators, diversity indices and evenness values calculated for the vegetation of each serpentine outcrop and its adjacent non-serpentine vegetation.

Parameter	Core Zone	Groen-valy	Kalk-kloof	Magne-site	Mundt's Concession	Rosen-tuin	Saw-mill
Serpentine outcrops							
No. of samples (*n*)	40	40	40	40	40	40	40
No. of individuals (*N*)	396	610	642	449	446	474	492
Observed species richness (*S*)	83	121	123	105	98	115	106
Species Richness estimators							
Jack 1	109.3	154.2	154.2	143.0	123.4	156.0	143.1
Jack 2	115.5	168.9	169.8	168.0	126.8	178.3	161.6
Bootstrap	96.1	136.6	137.5	121.5	111.1	133.4	122.8
Species diversity indices							
Simpson's (-ln λ)	3.4	4.1	4.3	4.0	3.8	4.0	3.9
Shannon (H')	3.8	4.4	4.4	4.2	4.1	4.3	4.1
Fisher's α	32.0	45.3	45.2	43.1	38.8	48.3	41.5
Evenness (J')	0.86	0.91	0.92	0.90	0.90	0.90	0.89
No. of singletons	27	34	32	39	26	42	38
Adjacent non-serpentine areas							
No. of samples (*n*)	40	40	40	40	40	40	40
No. of individuals (*N*)	508	545	535	290	352	386	475
Observed species richness (*S*)	127	100	132	88	95	104	99
Species richness estimators							
Jack 1	175.8	120.5	173.9	122.1	128.2	143.0	133.1
Jack 2	207.5	121.0	187.9	141.5	143.8	163.4	149.7
Bootstrap	148.0	111.0	152.2	103.4	110.3	121.6	114.5
Species diversity indices							
Simpson's (-ln λ)	4.15	3.84	4.17	3.78	3.97	3.84	3.81
Shannon (H')	4.40	4.14	4.43	4.06	4.15	4.16	4.07
Fisher's α	54.4	35.9	56.0	43.0	42.7	46.7	38.1
Evenness (J')	**0.91**	**0.90**	**0.91**	**0.91**	**0.91**	**0.90**	**0.89**
No. of singletons	**50**	**21**	**43**	**35**	**34**	**40**	**35**

Correlation of species richness with selected environmental factors

Multiple regression analyses suggest that there is no correlation (P>0.05) between species richness and each of the selected environmental factors that is, size of outcrop, latitude, distance to nearest outcrop and levels of chromium, nickel and the calcium to magnesium ratio for each site. The same lack of correlation with selected environmental factors was shown for species diversity estimated using the Shannon, Simpson's and Fisher's indices. There is a low correlation (P=0.05) between the serpentine diversity and altitude. As altitude increase so serpentine diversity increases, but as altitude increases the non-serpentine diversity decreases. The result that on the Barberton Greenstone Belt, the serpentine species diversity is not related distance to nearest outcrop is in contrast to the vegetation of the Tuscan serpentine outcrops where evidence suggested that taxonomic diversity decreases with increasing isolation of an outcrop and decreasing surface area (Selvi, 2007). For the Californian serpentine (Harrison et al., 2006) determined that regional richness is a strong predictor of local richness, demonstrating that the regional availability of species is one key determinant of local richness. The vegetation of the eastern escarpment of the Mpumulanga Province, within which the serpentine outcrops occur, is known to support high species richness (Thuiller et al.,

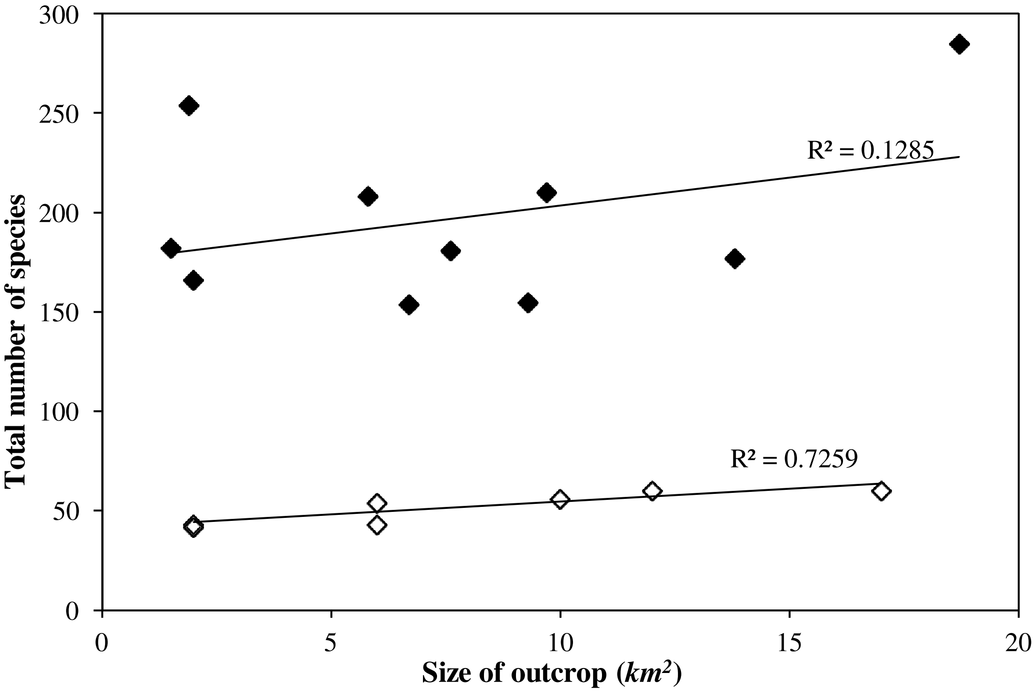

Figure 6. Scatter plot of the total number of species collected at a number of serpentine outcrops of the Barberton Greenstone Belt (♦) as compared to the number of plant species on similar sized outcrops from Tuscany (◊) in Italy (Selvi, 2007). R^2 values represent the goodness-of-fit for a linear regression calculated for each set of values. The Dunbar and Groenvaly sites are outliers with higher number of species than predicted by the line of best fit.

2006); thus it is predicted that this regional richness determines the richness of the serpentine vegetation. This is suggested by the small differences in serpentine and non-serpentine species richness and diversity. Further investigation is needed to establish the true determinants of species richness of the Barberton Greenstone Belt serpentine outcrops.

Comparisons of serpentine species richness across the world

There is huge variation in sampling strategy in the few reports of species richness of serpentine areas around the world. Werger et al. (1978) used plots of various sizes to determine the floristic diversity of the Great Dyke in Zimbabwe. Few studies of serpentine outcrops provide species richness values for individual outcrops. These factors make comparisons to the data presented here difficult. A direct comparison between the numbers of species recorded per outcrop of the Barberton Greenstone Belt and serpentine outcrops of similar size in Tuscany, Italy (Figure 6), show that the serpentines of the Barberton Greenstone Belt support many more species per outcrop than the serpentine in Tuscany (Selvi, 2007). A mean of 76 (range 26 to 205) species per serpentine was recorded for the outcrop of the Northern Apennines, Italy (Ferrari et al., 1992) which is considerably lower than the mean of 197 (range 155 to 285) species per outcrop recorded for the Barberton Greenstone Belt. The serpentine vegetation of Western North Carolina supports 27.9 species per 0.001 ha (Mansberg and Wentworth, 1984), which is higher than the serpentine of the Barberton Greenstone Belt with a mean of 23.9 species per 0.001 ha (Figure 7). The serpentine forest vegetation of the Wenatchee Mountains, Washington support 19.8 to 32 species per 0.1 ha (Del Moral, 1972), considerably lower than that of the Barberton Greenstone Belt (Figure 7).

The diversity of serpentine and non-serpentine vegetation on other continents has been compared by other authors, who have found varying patterns. The species diversity in the Siskiyou mountains is greater on serpentine than on quartz diorite or olivine gabbro except on xeric sites (Whittaker, 1960). Serpentine plant diversity (using Brillouin Index) was greater than non-serpentine diversity on xeric sites of the Wenatchee Mountains in Washington at higher elevations but not in very mesic conditions (Del Moral, 1972). On the Great Dyke in Zimbabwe, the species diversity on serpentine was found to be considerably lower than on granite or pyroxenite. In addition, it was found that on serpentine, species diversity and richness decreases with altitude (Werger et al., 1978).

In an effort to determine whether the vegetation of the serpentine outcrops has conservation value in terms of

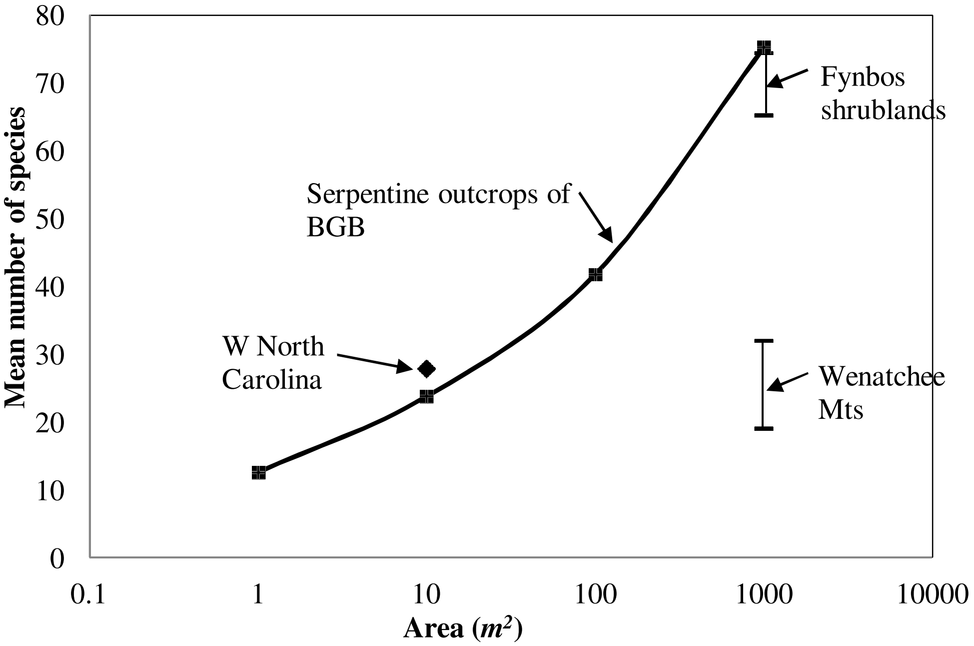

Figure 7. The mean number of species recorded in modified Whittaker plots (consisting of 1 m^2, 10 m^2, 100 m^2 and 1000 m^2 plots) for the serpentine outcrops of the Barberton Greenstone Belt (BGB) compared to the number of species recorded in the Cape Fynbos shrublands (Cowling, 1983), the serpentines of Western North Carolina (Mansberg and Wentworth, 1984) and the Wenatchee Mountains (Del Moral, 1972).

species richness, the number of species recorded per 0.1 ha plot on the serpentine of the Barberton Greenstone Belt was compared to those of the Fynbos shrubland vegetation of Cape Floral Kingdom as the Fynbos vegetation is considered to be extremely species-rich (Goldblatt, 1978). The number of species per 0.1 ha of the serpentine varies from 65.3 to 83.3 with a mean of 74.4, which is higher than the mean recorded for the Cape fynbos shrublands (66.4) (Cowling, 1983) (Figure 7). Although the two areas support decidedly different types of vegetation, the high species richness for the serpentine vegetation is an indication of potential conservation value.

Conclusion

This study represents the first attempt to comprehensively quantify the differences between the vegetation of serpentine outcrops and that of adjacent non-serpentine areas of the Barberton Greenstone Belt. We predicted that the serpentine outcrops of Barberton Greenstone Belt have been exposed sufficiently long to re-establish diversity following the geological changes associated with exposure. This study shows that the vegetation of the serpentine is not generally more species rich or diverse than the surrounding non-serpentine vegetation. However, one site showed significantly higher and a few sites slightly higher serpentine diversity, and it is possible that with more intensive sampling the species accumulation curves could show these differences to be significant. The species richness and diversity on the serpentine sites is not significantly lower than that of the surrounding vegetation, which supports Brown (1988)'s theory that the serpentine outcrops of the Barberton Greenstone Belt were exposed sufficiently long ago to allow the vegetation to re-establish diversity after the initial geological disturbance. The Core Zone site has significantly lower species richness and diversity, and it is possible that the isolation of this outcrop has resulted in its lower diversity.

The flora of the Barberton Greenstone Belt seems to be distinct from the surrounding vegetation. Quantifying this distinction, the Sørenson's index, comparing the species composition on and off serpentine for each site, suggests that although species richness is similar on and off serpentine, there is a considerable change in species composition across the serpentine to non-serpentine 'boundary'. This supports the view that serpentine outcrops support a unique and diverse flora. Further research is required to determine whether grassland species in this area are less able to colonise serpentine soils than bushveld species, as suggested by the correlation in Table 2. A further hypothesis that should be tested in the future is that sites that are geographically clustered are more similar to each other than to their adjacent non-serpentine vegetation.

Identification of the factors affecting diversity is essential for determining conservation priorities for these serpentine outcrops. There is considerable variation in the species richness and diversity calculated for each site sampled, with the sites in the southern parts of the Barberton Greenstone Belt having higher richness than the northern sites. However, the differences could not be correlated to latitude or any other climatic or edaphic factor. Further investigations, sampling the vegetation of more outcrops and considering additional environmental factors should reveal the factors determining diversity on the various outcrops and the extent of the altitudinal correlation with species diversity.

The serpentine sites of the Barberton Greenstone Belt show considerably higher species richness than that reported for the serpentines of Tuscany (Ferrari et al., 1992) and the serpentine forest vegetation of the Wenatchee Mountains, Washington (Del Moral, 1972) and slightly lower species richness than the serpentine vegetation of Western North Carolina (Mansberg and Wentworth, 1984). Further comparisons could not be made as additional comparable data could not be found and, therefore, no general statements can be made as to how the vegetation of the serpentine of the Barberton Greenstone Belt compares to other sites around the world in terms of species diversity.

The species richness and diversity values calculated here, together with levels of endemism still to be published, will form part of a larger study which will state a case for including serpentine vegetation in the planning for conservation of the vegetation of the eastern portion of Mpumalanga Province.

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Chapter 5

Beta and Gamma diversity of the serpentine vegetation of the Barberton Greenstone Belt

Introduction

Diversity is usually measured as within-habitat diversity (or alpha (α) diversity), which is considered to be the diversity of a defined assemblage or habitat (Magurran 2004) and refers to the number of species in a sample representing a community regarded as homogeneous (Whittaker 1977, Cowling *et al.* 1991). However, it is also possible to define diversity on a broader scale in terms of the biotic change or species replacement i.e. β (beta) diversity or between habitat diversity. Both α and β-diversity are components of γ (gamma) diversity, which represents the diversity of a landscape (Magurran 2004). Gamma diversity describes the species turnover among similar habitats along geographical gradients. The size of the regional species pool is a function of the interaction between alpha, beta and gamma diversities that are controlled by ecological and stochastic processes (Cody 1986).

Whittaker (1972) described β-diversity as a measure of change between samples along transects or along an environmental gradient i.e. species turnover. Other authors (Magurran 2004 and Vellend 2001) propose that the concept of β-diversity can also be applied to various spatial configurations of sampling units. It can also refer to the difference in species composition between local and regional assemblages (Koleff *et al.* 2003). The species that have arisen on California's serpentine outcrop add only modestly to the plant diversity of any one region (i.e. α diversity) but the narrowly distributed species add disproportionately to the among region (γ) component of diversity that is of such significance in producing California's botanical richness (Harrison *et al*. 1992).

High β and γ diversity indicate large numbers of taxa with high habitat specificity, many of which would be rare local endemics. The extinction of local endemic taxa that contribute to γ diversity, through habitat clearing and fragmentation, may lower all diversity components as resource use is adjusted within the community by density compensation (Cody 1986). This will produce more stable diversity patterns (Magurran 2004). Particular areas may have similar α diversity, however, if local endemics and rarities are significantly contributing to β and γ diversity, then it will be necessary to protect a network of sites that represent a broad range of geographic, geologic and climatic variation (Harrison *et al.* 2002). Thus, the quantification of β-diversity can be directly applied to questions of reserve design, the effects of fragmentation and estimation of global diversity (Cody 1986, Thomas 1990).

Since Whittaker (1960) devised his original measure of β-diversity, a large range of measures have been introduced, and there is no consensus about which ones are most appropriate for addressing particular ecological questions (Anderson *et al.* 2011). Theoretically, β-diversity measures the turnover of species composition among different communities or habitats (Vellend 2001). In practice, habitats and communities are nearly impossible to delineate, and so many of these measures have been applied at very different spatial scales (Anderson *et al.* 2011). The β-diversity in different groups of taxa across Britain was determined (Harrison *et al.* 1992), and a measure of beta diversity was used to compare paired plots of 500 m^2 on serpentine outcrops in California (Harrison *et al.* 2006). A comparison of absolute values for β-diversity calculated for different studies thus becomes problematic.

High habitat specificity of constituent organisms has been shown to be correlated with high β-diversity (Wilson and Shmida 1984, Cowling *et al.* 1991). These habitat specialists may be particularly rare and restricted if their preferred habitats are scarce. Habitat specificity has been shown to be the case in plant taxa restricted to serpentine soils in California (Cody 1986).

At first glance, the vegetation of serpentine sites of the Barberton Greenstone Belt may seem relatively homogeneous and outcrops seem similar to one another. However, the altitude of any one outcrop can vary by as much as 550 m. This change can cause additional variation in other edaphic factors including soil depth and nutrient availability, which are likely to cause changes in species composition along the elevation gradient. It is predicted that the larger the change in edaphic and other physical conditions the higher the change in species composition within a serpentine outcrop. Also, it will be determined whether rarer or locally endemic taxa contribute more significantly to species turnover within sites than the more common and widespread taxa.

In this study, β-diversity will be measured as the turnover of species in different habitats within serpentine sites. These data will be used to test the prediction that the β-diversity values of sites with the greatest altitudinal ranges and surface areas are higher than the diversity values of sites with smaller altitude differences and areas. This study also aims to determine whether there is a correlation between the levels of β-diversity (within serpentine site diversity) of the vegetation and the diversity of physical properties such as soil characteristics and topography.

Cody (1986) defines gamma (γ) diversity as the turnover rate with the distance between sites of similar habitat, or with expanding geographic areas. However, Magurran (2004) considers γ diversity an inventory diversity describing the diversity of a landscape and Cody's (1986) definition is closer to the delta (δ) diversity, as defined by Magurran (2004). In this study, Cody's (1986) definition of γ diversity will be adopted and will be measured as the turnover rate of species between various serpentine outcrops of the Barberton Greenstone Belt, which are separated by distance. The prediction that the turnover of taxa between sites with different altitudes and large latitudinal and longitudinal ranges is higher than that of sites with similar altitudes and close proximities will be tested using γ diversity data. In addition, turnover of taxa will be correlated with distance from the closest neighbouring site to determine whether turnover between sites is related to geographic isolation of sites. These predictions are based on the reasoning that the insular nature of serpentine outcrops will result in limited dispersal and biotic homogenization in evolutionary and ecological time (Harrison and Inyoue 2002). This should cause a greater change in biotic communities over a certain distance in comparison with the floras of more continuous habitats.

The difference in species composition between pairs of serpentine regions in California was higher in serpentine endemics than for plants as a whole and showed greater variation across geographic and environmental distance (Harrison *et al.* 2002). In addition, the γ diversity data will be used to determine whether the indicator and endemic taxa of the Barberton Greenstone Belt contribute more to the turnover between sites than the more common and widespread taxa.

The ability to assess and predict biodiversity is important both for identifying places of high conservation value and for monitoring the efficacy of resource management (Hewitt *et al.* 2005). Therefore, the results of this study together with data on endemism (Chapter 3) and species richness (Chapter 4) could have important implications for establishing conservation priorities for the serpentine sites.

Materials and Methods

β-diversity has historically been measured as species turnover along a habitat gradient. However, Vellend (2001) argues that β-diversity indices can also measure among-plot variability in species composition independently of the position of individual plots on spatial environmental gradients. To use the appropriate index and to make comparisons between investigations, this distinction must be recognised. In this study, β-diversity was measured for each of the selected serpentine outcrops of the Barberton Greenstone Belt (listed in Chapter 1) using the placing of Modified-Whittaker plots at bases, crests, north-facing and south-facing slopes as the environmental gradient.

Various turnover diversity measures were calculated using the presence – absence data collected from the Modified-Whittaker plots. Since it is not clear from the literature that any single measure is better than any other and since no two measures record the same quantities, β-diversity of the serpentine outcrops was calculated using a number of different turnover diversity measures. These measures were selected based on their ease and simplicity of calculation and interpretation and their frequency of use in similar studies. One of the simplest measures of β-diversity was devised by (Whittaker 1960):

$$\beta_w = {}^{S}/_{\bar{\alpha}}$$

where S is the total number of species recorded in the system (γ diversity) and $\bar{\alpha}$ is the average sample diversity, measured as the average number of species per sample. Whittaker subsequently (1972) recommended modifying the measure by subtracting 1.0 from the ratio i.e.

$$\beta_w = ({}^{S}/_{\bar{\alpha}}) - 1).$$

This has the effect of putting the result on the scale of 0 (minimum β-diversity) to 1 (maximum β-diversity) when β_W is calculated between pairs of samples. To calculate the within site turnover of the selected serpentine outcrops S is the total number of species recorded from the four Modified-Whittaker plots for a particular site and $\bar{\alpha}$ is the average number of species per plot. This measure determines the proportion by which the whole serpentine outcrop is richer than the average plot within it. This measure is recommended when samples are not arranged along a single gradient (Wilson and Shmida 1984) as is the case here. β_w was also used to determine the pairwise differences or turnover between serpentine outcrops.

Numerous modifications to this formula have been proposed that constitute variations on the theme. Cody (1975) proposed a measure of species turnover, which adds the number of new species encountered along a gradient to the number of species that are lost. A modification of Whittaker's measure was introduced to allow for comparisons between samples of different size and a second modification ensured that the measure was insensitive to species richness trends (Harrison *et al.* 1992). Wilson and Shmida's (1984) measure determines the rate of species loss (l) and gain (*g*) along a gradient.

$$\beta_T = \frac{[g(H) + \ l(H)]}{2\bar{\alpha}}$$

These measures were also used to calculate β and γ diversity of the serpentine outcrops to facilitate comparisons between this investigation and others.

Indices of complementarity and similarity were also used to determine the β and γ diversity of the selected serpentine outcrops. The term 'complementarity' describes the difference between sites in terms of the species they support (Magurran 2004). The more complementary two sites are, the higher their turnover diversity, i.e. the higher the need to have both plots to make up the full complement of species. These indices commonly combine three variables: *a,* the total number of species present in both samples; *b,* the number of species in sample 1 and *c*, the number of species in sample 2. The Jaccard similarity index:

$$C_J = \frac{a}{a + b + c}$$

and Sørensen's similarity measure:

$$C_S = \frac{2a}{2a + b + c}$$

were used. To describe the complementarity of sites

$$1 - C_J \qquad or \qquad 1 - \ C_S$$

would be used.

As a measure of β-diversity (within site diversity), four measures of turnover (β_W, β_T, C_J and C_S) were used to calculate the turnover, pair-wise, between four 1000 m^2 Modified–Whittaker plots placed on each outcrop (site) as described in Chapter 1. The matrices showing turnover between pairs of plots were then correlated with matrices showing differences in altitude, latitude and longitude between each plot. A Pearson correlation within Mantel test was conducted between each turnover and environmental characteristic matrix with P-values based on 10 000 randomizations of the turnover matrix. Each turnover matrix was also correlated with the equivalent turnover matrix for the adjacent non-serpentine area, to determine whether the diversity on serpentine is related to the diversity in the entire area.

βw does not measure the turnover of species directly but instead measures the proportion by which a region is richer than the average locality within it. This is in contrast to β_T, which measures the species replacement among localities within a region (Blackburn and Gaston 1996). To determine whether βw accurately represents species turnover between plots on individual sites and between serpentine outcrops, the calculated values for β_W, β_T, C_S and C_J were correlated with one another. The correlation between measures was either determined using linear regression or using a Mantel test of matrix correlation.

Three absolute measures of β-diversity i.e. β_W of Whittaker, modified Whittaker's, β_{H2} of Harrison *et al.* (1992), and β_T of Wilson and Shmida were calculated for each serpentine site from data collected in the plots. Each measure of β-diversity was correlated with surface area, altitudinal range, latitude, longitude and soil chemistry of each site. To compare the species turnover between two habitats, transects and plots were placed along parallel gradients. As the plots were placed on the selected serpentine sites and adjacent non-serpentine areas in similar positions, the turnover between serpentine and non-serpentine was also determined and correlated to environmental factors such as longitude, latitude, altitude and surface area of outcrops.

The matrices resulting from the calculation of between site turnover of the serpentine outcrops or γ diversity were correlated with the matrix of between site geographical distance using Pearson's correlation within the Mantel test calculated using the XLSTAT statistical package for Excel. In order to identify whether the plant species occurring on the serpentine outcrops have narrow or wide tolerances for various climatic and physical conditions, the presence of species on particular outcrops was correlated to mean altitude above sea level (m.a.s.l), mean annual precipitation, mean minimum and maximum temperatures, latitude and longitude of each outcrop. Climatic data were sourced and collated from a number of websites (http://www.worldweatheronline.com; http://en.climate-data.org and http://portal.sasa.org.za. The presence/ absence of plant species at each site was tabulated and arranged in matrices according to each environmental factor to determine the number of taxa with possibly restricted tolerances to these conditions. Checklists of plant taxa collected at Agnes Mine (AM), Kaapsehoop (KH) (Williamson 1994) and Dunbar (DB) (Changwe and Balkwill 2003) were included to broaden the analysis.

Results and Discussion

Beta diversity (Within site diversity)

β_W and β_T are calculated quite differently and quantify different aspects of turnover. β_W measures the proportion by which a region is richer than the average locality within it and β_T measures species replacement among localities in a region. It is thus predicted that there would not be a significantly large correlation between these measures. However, significantly high correlations between the different measures reveal that although β_W does not directly measure species turnover between plots and within each serpentine site, it is equally capable of measuring turnover on serpentine sites as are measures β_T, C_S, *and* C_J (Table 5.1 and 5.2). This strong correlation is expected when $[g(H) + l(H)]$ used to calculate β_T is proportional to S (the total number of species recorded) used to calculate β_W (Blackburn and Gaston 1996).

When β_W and β_T were calculated using the pooled data from each plot on a site the correlation between these two measures becomes weaker ($R = 0.905$, $P<0.01$) than correlations between measures calculated using the mean turnover between pairs of plots on each site (Table 5.1). This suggests that the pooled data have less resolution and may weaken the correlation. This is possibly due to the small sample size, demanding a larger statistical difference before those differences are recognised as significant.

Table 5.1: The coefficients (R) of the correlations between the different measures of β-diversity when calculated as the mean turnover between each plot within a site. N = 7 in all cases.

	C_J		β_W		β_T	
	R	Significance	R	Significance	R	Significance
C_S	0.999	P<0.001	1	P<0.001	0.990	P<0.001
C_J			0.999	P<0.001	0.990	P<0.001
β_W					0.990	P<0001

A comparison of the β-diversity calculated from the presence/absence data obtained from the 1000 m^2 Modified-Whittaker plots placed on serpentine outcrops and on neighbouring non-serpentine areas reveals that five of the seven sampled sites have lower β-diversity than the non-serpentine areas (Figure 5.1). However, these differences in β-diversity are only significant for two outcrops ($P<0.001$ for CZ and $P<0.01$ for MM) across all measures and methods used. Within site β-diversity was calculated by averaging the values calculated for pairwise comparisons of each plot placed at each site (Figure 5.1 and Table 5.2) and also by pooling the data from all the plots (Figure 5.2). The matrices with the values of the various β-diversity measures calculated for each pair of plots for each site are shown in Appendix E. The different methods of calculating β-diversity and each of the measures yield different numerical answers, but consistently highlight higher β-diversity between the plots placed in the non- serpentine vegetation at five of the sampled sites. The various measures yield different values due to the variation in scaling of $\bar{\alpha}$ (average species richness per plot). Higher β-diversity values were determined for the serpentine outcrops of the Klamath-Siskiyou Mountains in California (Whittaker 1960). In contrast, non-serpentine meadows have higher β-diversity than serpentine meadows in northern California (Harrison 1999). A possible reason for the lower β-diversity on non-serpentine meadows is that the total habitat area of the non-serpentine vegetation leads to a larger regional pool of species on non-serpentine (Harrison 1999).

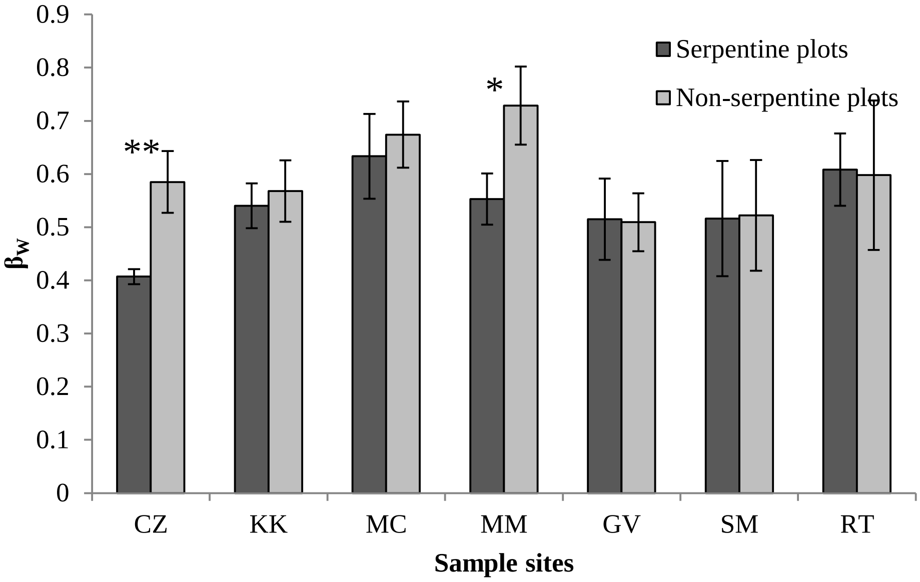

Figure 5.1: Comparing the Whittaker (1960) measure of β–diversity for serpentine sites with that of the adjacent non-serpentine sites, calculated by averaging the pair-wise comparisons of plots placed at each site. * indicates a significant difference between means at P<0.01 and ** indicates P<0.001

Table 5.2: Means of various β-diversity measures calculated using the averages of the pairwise comparisons of plots placed at various positions on each serpentine outcrop with the equivalent measure for adjacent non-serpentine areas.

	β_T		$1\text{-}C_J$		$1\text{-}C_S$	
Sites	Serpentine	Non-serpentine	Serpentine	Non-serpentine	Serpentine	Non-serpentine
CZ	0.703	0.792	0.578	0.737	0.407	0.585
KK	0.771	0.784	0.701	0.723	0.540	0.568
MC	0.817	0.837	0.773	0.804	0.633	0.674
MM	0.764	0.864	0.711	0.841	0.553	0.729
GV	0.760	0.753	0.677	0.674	0.515	0.509
SM	0.758	0.761	0.676	0.681	0.516	0.522
RT	0.809	0.799	0.755	0.740	0.608	0.598

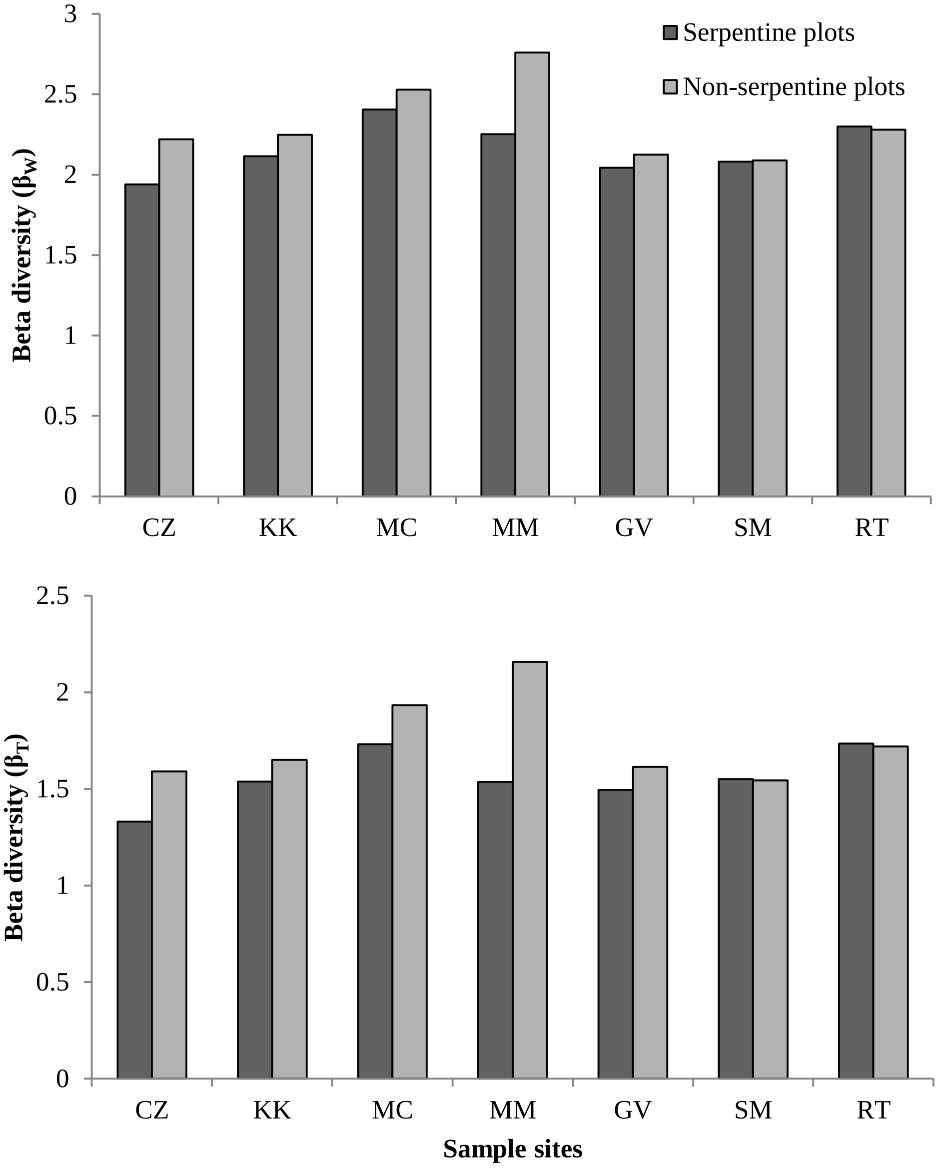

Figure 5.2: Comparison of β-diversity on each serpentine site with that of each adjacent non-serpentine area, using two different measures of β-diversity, calculated from the pooled data from all the plots at each site.

A large number of the species in the regional pool around the Barberton Greenstone Belt are excluded from the serpentine outcrops (Appendix B), reducing the species pool for the serpentine vegetation. Small localities cannot fully absorb the abundance of species that are produced at the regional scale by the high rates of speciation and/or low rates of extinction that are characteristic of highly productive regions (Harrison *et al.* 2006). This results in a lower value for *S* (i.e. total number of species recorded) for the non-serpentine species pool and thus a lower overall β-diversity using Whittaker's (1960)

$$\beta_W = \frac{s}{\bar{\alpha}}.$$

The values for β_W and β_T presented in Figure 5.1 do not fall between 0 and 1, as they were calculated for more than one plot within each site. Values between 0 and 1, with 0 representing minimum turnover and β-diversity and 1 representing maximum diversity, are achieved only for pairs of plots.

Determining whether the values for each site are relatively high or low is problematic as it is difficult to define finite limits of β_W and β_T mathematically. However, the values for β_{H2} fall between 0 and 100 and as the β_{H2} for each site falls below 50 (CZ = 31 to MC = 47). This suggests that β-diversity on the serpentine sites of the Barberton Greenstone Belt is relatively low and supports the visual observation of floristic homogeneity.

The values of β_W calculated for these sites correspond with those calculated for the serpentine outcrops of the Siskiyou Mountains ($\beta_W = 2.33$) (Whittaker 1960). There is merit in comparing the results from this study with those of Whittaker (1960) as similar sized plots were used to sample the vegetation. However, it is not meaningful to compare absolute values of β-diversity calculated for the Barberton Greenstone Belt with those of many other investigations as β-diversity has been measured at different spatial scales. Comparisons between values of β-diversity across different investigations are further complicated by differences in the sizes of the areas within which the sampled areas are embedded (Gaston and Williams 1996).

Harrison and Inouye (2002) predict that within site species richness (α diversity) will be lower on small isolated serpentine outcrops, but that β-diversity will be higher for these outcrops. The results for the serpentine outcrops of the Barberton Greenstone Belt seem to contradict that prediction. There is no correlation between the degree of isolation of a site (measured as distance to nearest neighbouring outcrop) and the β-diversity of the flora. One of the least isolated outcrops, Mundt's Concession (MC) (See Figure 2.1 of Chapter 2) has the highest β-diversity. However, other sites that are equally close to their nearest neighbours have comparatively low β-diversity e.g. Groenvaly (GV) and Sawmill (SM). The second most isolated outcrop, Core Zone (CZ) has the lowest β-diversity; however, the most isolated outcrop (KK) has comparatively high β-diversity. The differences in β-diversity are more likely due to the numbers of species occurring at each plot as there is a strong correlation between levels of β-diversity and the average number of species recorded in each plot ($P < 0.02$) (Table 5.3).

No significant correlation was found between levels of β-diversity at the sampled sites and α diversity (Chapter 4) at each site (Table 5.3). The CZ site has relatively low β-diversity and α diversity, while GV has low within site β-diversity but relatively high α diversity (Chapter 4). These data contradict the prediction that within site species richness (α diversity) will be lower on small isolated serpentine outcrops (Harrison and Inouye 2002). Also, no significant correlations were found between various environmental factors measured at each site (Table 5.3) including size and position of each site. A weak correlation ($P < 0.2$) was found for levels of β-diversity and elevation, which suggests that β-diversity decreases with increasing elevation. These results confirm the findings of Harrison and Inouye (2002) and Whittaker (1960), who found a more rapid turnover in species composition along an elevational gradient in serpentine compared to the non-serpentine. This is possibly due to the interaction between rainfall and soil factors such as the rain resulting in the leaching of some of the toxic heavy metals to the lower altitude areas and creating a soil toxicity with elevation changes. A further weak correlation ($P< 0.2$) was found for β_T and surface area of outcrops; however this was not seen for the other measures of β-diversity. Although the correlations between β-diversity and elevation and surface are not considered to be statistically significant, the P values could be high due to small sample sizes and may indicate a true biological difference.

The differences in the degree of correlation between β_W and β_T and the various factors listed in Table 5.3, are attributed to each measure using different components of the flora to determine turnover. β_W is a ratio of the total number of species recorded in the plots (S) to the average number per plot ($\bar{\alpha}$), while β_T is determined using the sum of the gain and loss of species between plots.

Table 5.3: R-values and the statistical significance resulting from linear correlations of the different β-diversity measures with various biological and environmental factors, measured at each outcrop (ns = not significant at $P \geq 0.05$).

Feature	Whittaker (β_W)		Wilson & Shmida (β_T)	
	R	Significance	R	Significance
Observed species richness	0.476	ns	0.263	ns
Mean no of species per plot	0.868	$P<0.02$	0.778	$P<0.05$
Mean altitude (m.a.s.l.)	-0.604	ns	-0.173	ns
Altitudinal range	0.430	ns	0.457	ns
Mean Longitude	0.477	ns	0.145	ns
Mean Latitude	0.298	ns	0.134	ns
Surface area (km^2)	0.364	ns	0.607	ns
Soil [Ni]	0.179	ns	0.292	ns
Range in measured [Ni]	0.228	ns	0.253	ns
Soil [Cr]	0.361	ns	0.173	ns
Range in measured [Cr]	0.226	ns	0.170	ns
Ca: Mg	0.302	ns	0.221	ns
Ca: Mg range	0.303	ns	0.207	ns

The results suggest that the levels of β-diversity of the serpentine outcrops of the Barberton Greenstone Belt are not determined by habitat diversity, as measured by range in elevation, longitude and latitude, or by variation in soil chemistry at each site. The mean turnover of species from the base to crest plots for all serpentine sites is significantly ($P < 0.05$) lower than the mean turnover between the adjacent plots placed on non-serpentine areas when compared using paired T-tests. This further suggests that elevation as a factor causing increasing β-diversity in non-serpentine areas has a reduced causal effect on β-diversity on serpentine soils. The different components of the serpentine vegetation (i.e. herbs and grasses vs. trees and shrubs) contribute to the overall turnover diversity (β_W) at each site in various ways (Figure 5.3). The turnover of trees and shrubs is greater on three serpentine sites (i.e. KK, GV and SM) than on the adjacent non-serpentine areas. Although the overall turnover is similar when considering the whole flora (Figure 5.1). There are fewer trees and shrubs at these serpentine outcrops than on neighbouring non-serpentine areas (Table 5.4). However, the higher turnover diversity values suggest that these trees are distributed more patchily through the serpentine areas. Both types of vegetation show similar turnover between plots on the MM and CZ serpentine sites. Although there are significantly fewer trees on the MC serpentine site (Table 5.4) the turnover diversity of the trees and shrubs is similar to that calculated for the adjacent non-serpentine areas (Figure 5.3).

The turnover diversity (β_W) of the trees and shrubs is strongly correlated ($P<0.05$) with the elevation of individual sites, while the turnover of the herbaceous species is not correlated with elevation (Figure 5.4). The weak correlation between overall β_W and elevation of the serpentine sites (Table 5.3) is possibly due to the small numbers of trees and shrubs occurring on the serpentine sites.

In areas with high β-diversity, some species may have evolved to occupy different positions along habitat gradients. Populations of these species would then occupy restricted ranges of the total gradient (Whittaker 1972). β-diversity values calculated for the three largest plant families represented on serpentine sites show that taxa in the Poaceae family show significantly lower turnover within five of the seven serpentine sites (KK, MC, MM, GV and SM) than plants in the Asteraceae and Fabaceae (Figure 5.4). As the species in the Poaceae have the highest abundance and richness, their numbers would significantly contribute to the lower overall β-diversity calculated for each site and the herbaceous plants. Although the overall turnover within serpentine outcrops was found not to be correlated with soil chemistry, the turnover of the species in the Poaceae was significantly correlated with chromium concentrations and calcium to magnesium ratios ($P<0.02$).

The low β-diversity values calculated for the serpentine flora suggest that a large proportion of the species (such as those in the Poaceae) have more continuous distributions. Also, the habitat gradients presented by the serpentine outcrops are not large enough to cause a greater turnover of species along these gradients. Greater sampling of the flora and soils on each outcrop should reveal whether the turnover patterns reported here are a true characteristic of the serpentine flora or due to the small sample sizes.

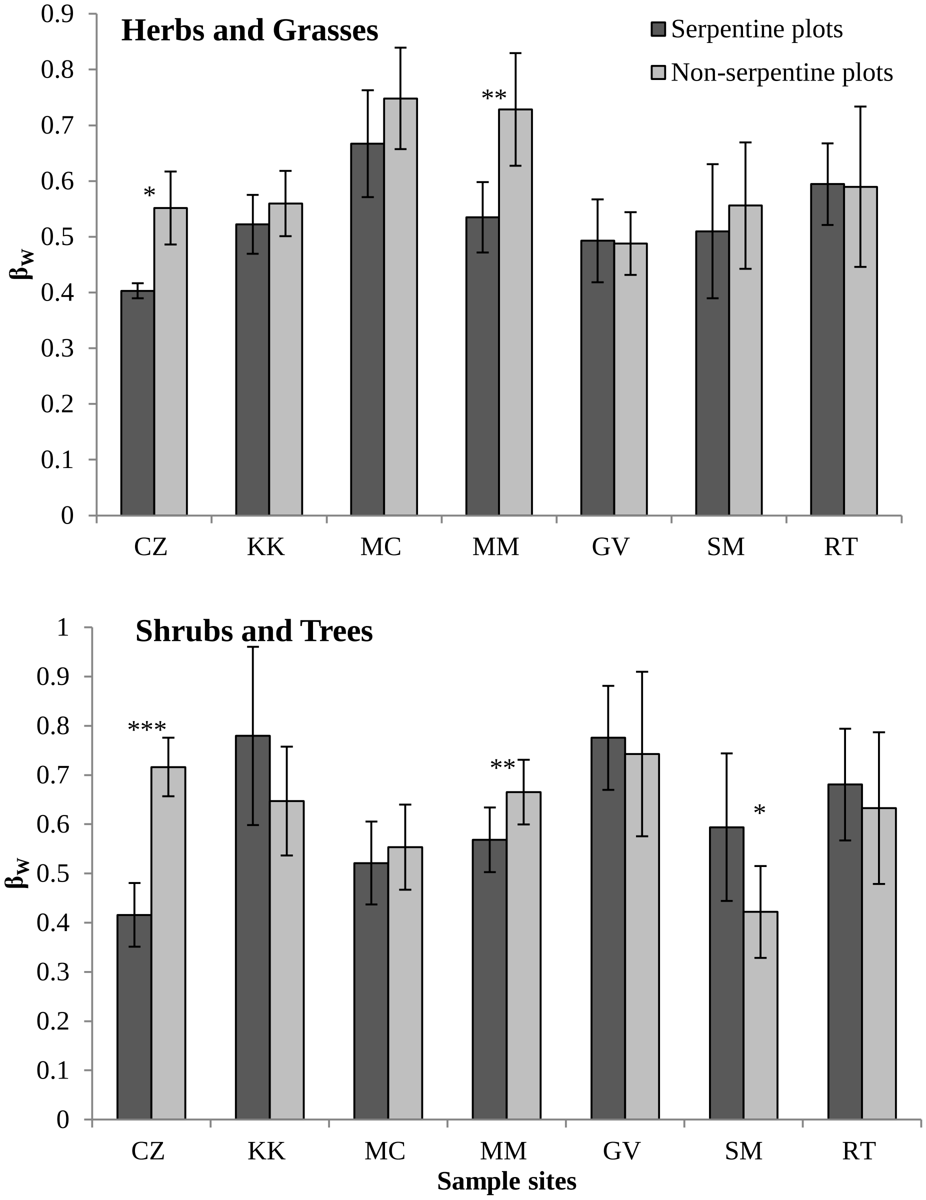

Figure 5.3: Comparison of β-diversity values determined for serpentine and adjacent non-serpentine plots, with the vegetation separated into herbaceous and woody components, calculated by averaging the pairwise comparisons of plots placed at each site. * indicates significant difference between means at P<0.01, ** indicates P<0.005 and *** indicates P<0.001**.**

Table 5.4: Comparing the proportions, as percentages, of trees and shrubs on each serpentine site with those of the adjacent non-serpentine areas using z-tests (ns indicates differences that were not significant at P≥0.05).

Site	Numbers of herbs and grasses		Numbers of trees and shrubs		% of trees and shrubs		Significance
	Serpentine	Non-serpentine	Serpentine	Non-serpentine	Serpentine	Non-serpentine	
CZ	106	144	37	46	25.8	24.2	ns
KK	161	165	16	21	9.0	11.3	ns
MC	122	91	43	58	26.1	38.9	P<0.01
MM	145	137	19	18	11.6	11.6	ns
GV	126	117	32	54	20.3	31.6	P<0.01
SM	138	128	17	35	11.0	21.5	P<0.01
RT	137	119	28	34	17.0	22.2	ns

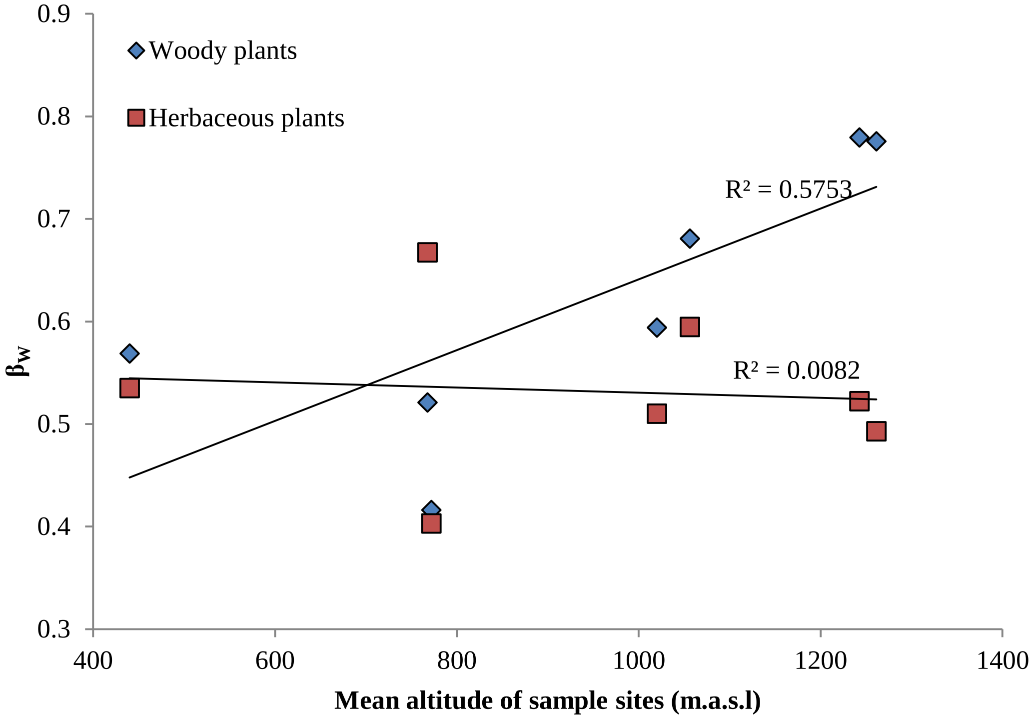

Figure 5.4: The correlation of the β diversity of the woody and herbaceous species at each serpentine site with elevation of each site.

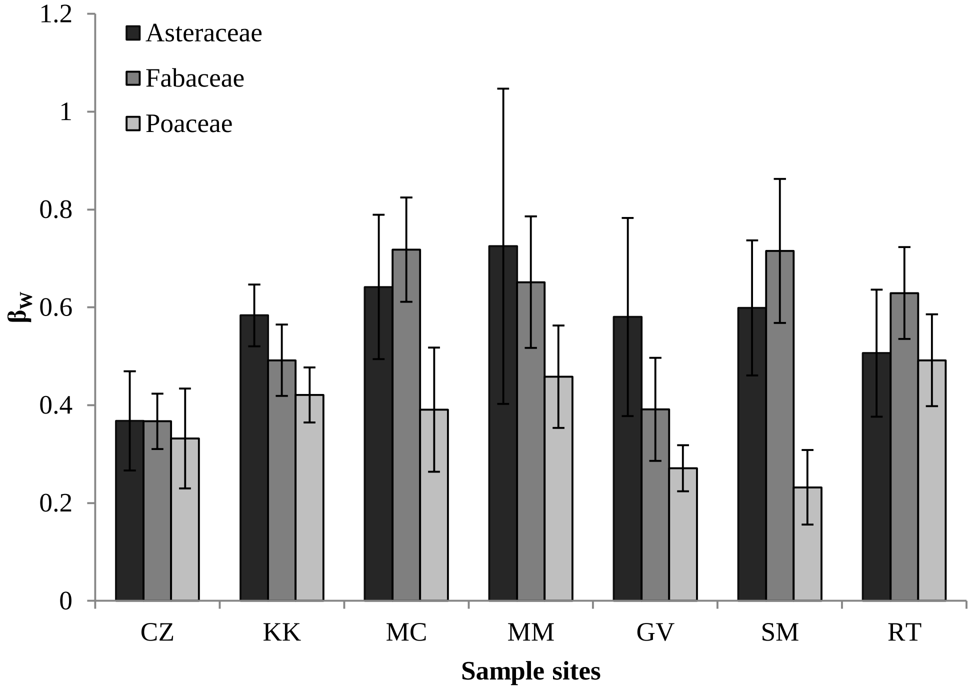

Figure 5.5: A comparison of the β-diversity (β_W) values determined for the three largest families represented on each of the serpentine outcrops.

In addition, most of the non-serpentine areas around the serpentine sites have been disturbed by human activities such as agriculture (CZ, MC and MM), forestry (GV and SM) and the presence of alien plants such as *Lantana camara* L., *Pinus* spp. and *Eucalyptus* sp. These disturbances could cause increasing patchiness in the distribution of plant species growing in the non-serpentine areas and may result in an increase in turnover or β-diversity between plots on these sites (Harrison 1997). In contrast, the serpentine outcrops of the Barberton Greenstone Belt are relatively undisturbed due to the toxicity of the soil and being unsuitable for agriculture and forestry, resulting in decreased patchiness and turnover between plots.

It has been revealed that, for different indices, β-diversity can decrease with increasing sampling effort. This results from the level of effort determining that many species that are shared by two communities may not be sampled from one or both by pure chance. Differences between them are therefore artificially inflated (Colwell and Coddington 1994 and Chao *et al.* 2005). This happens even when comparing samples that come from the same community, which should ideally exhibit no differences. On the other hand, the opposite situation is also possible. If the compared communities share a few very abundant species, and if there are many rare species that are exclusive to each community, estimated β-diversity may increase with increased sampling effort. In such cases, undersampling will tend to reveal only the shared, more abundant species and β-diversity will, therefore, be underestimated (Cardoso *et al.* 2009). Almost all β-diversity measures are based on presence/ absence data and do not consider frequency data. Most researchers derive new measures for the purpose of addressing such issues as the changes in species composition along spatial or environmental gradients (Koleff *et al.* 2003).

The analysis of α diversity completed in Chapter 4 revealed that about 50% of the species recorded in the plots placed on each serpentine site were recorded only once or twice (i.e. singletons and doubletons). 39 Thirty-nine % of the KK flora and 58 % of CZ flora, respectively, are singletons and doubletons, and percentages of the other sites fall between these values. This suggests that with a larger number of samples taken at each site the estimated β-diversity presented here could increase, as more of the rare species will be recorded with more plots used.

Turnover diversity across edaphic boundary

As with the within site β-diversity, different measures give different ranges of values but show similar patterns of relative values (Figure 5.6). Whittaker's (1960) β_W represents a turnover value, and Sørensen's C_S and Jaccard's C_J (Magurran 2004) represent the similarity between the flora of each serpentine site and its surrounding non-serpentine area. The similarity values as calculated for C_S are higher than those calculated for C_J. This is due to the variable *a* (the total number of species present in both samples) being doubled in the formula for C_S giving the number of species shared by two samples a greater representation in the similarity value, consequently representing a lower turnover or β-diversity.

The levels of turnover, calculated using Whittaker's (1960) β_W, between floras of individual serpentine sites and their surrounding non-serpentine sites range from 0.667 to 0.765 (Figure 5.1). There is a weak negative correlation between β_w calculated within each site and the β_w calculated for turnover across the serpentine / non-serpentine edaphic boundary ($R = 0.69$, $P < 0.1$). This suggests that those sites with low β-diversity show high turnover in species across the serpentine / non-serpentine edaphic boundary. For example, although the CoreZone (CZ) site has low β-diversity or within site diversity (Figure 5.1) there is a high turnover of species ($\beta_W = 0.765$) from the serpentine site to the surrounding non-serpentine soils in the area (Figure 5.6). Conversely, Mundt's Concession (MC) serpentine site supports a relatively high within site β-diversity and lower levels of species turnover from the serpentine soils to the surrounding non-serpentine soils ($\beta_W = 0.667$).

Most serpentine soils have unusually high levels of heavy metals such as nickel, chromium, cobalt, iron, etc.; along with high ratios of Mg:Ca (Brooks and Yang 1984) and low concentrations of plant nutrients such as nitrogen, phosphorus and potassium (Brooks 1987). It would, therefore, be expected that the plant species turnover from serpentine to non-serpentine would be greatest from serpentine sites with the highest levels of toxic metals. However, no significant correlation was found between the species turnover (β_T) or similarity (C_S and C_J) across the edaphic boundary and soil nickel and chromium concentrations ($P > 0.6$ and $P > 0.5$, respectively) as well as the soil calcium to magnesium ratio ($P = 0.8$). A weak correlation was found between β_W across the edaphic boundary and soil Ca: Mg ($P < 0.2$). This weak correlation suggests a true

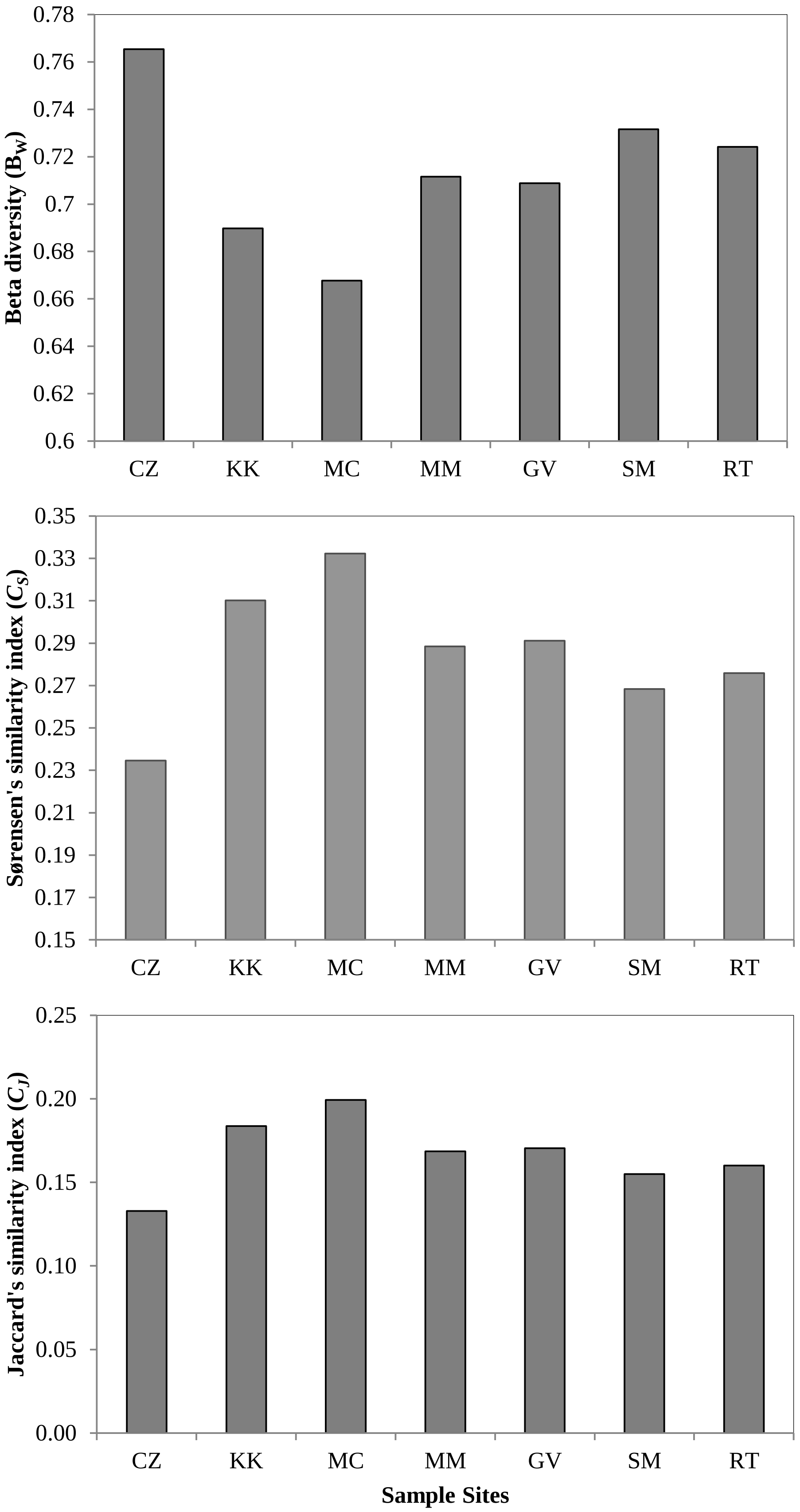

Figure 5.6: β diversity values calculated using β_W, C_S and C_J, for each site measured as the turnover of species across the serpentine / non-serpentine edaphic boundary.

biological effect and the high P value is possibly due to the small sample sizes. These correlations suggest that the toxic levels of magnesium in serpentine soils could be a greater contributor to the contrast in floras between serpentine and non-serpentine, than the concentrations of nickel and chromium in the soil.

The data presented in the graph (Figure 5.6) would suggest that sites with low numbers of recorded species, such as CZ, have a higher turnover of plant species across the serpentine edaphic boundary. However, there is no significant correlation between total numbers of species recorded in sample plots and the levels of species turnover between serpentine and adjacent non-serpentine areas. There is a weak negative correlation ($P<0.1$) between within serpentine site β_W and turnover between serpentine and non-serpentine areas, suggesting that the higher the turnover of species composition within a serpentine site the lower the turnover of species across the serpentine and non-serpentine edaphic boundary.

Even though the similarity between serpentine and non-serpentine floras is relatively high at the Mundt's Concession (MC) and Kalkkloof (KK) sites and low at the Core Zone (CZ), the values for C_S only range from 0.23 and 0.33. This is equivalent to a less than 35% similarity between the serpentine and non-serpentine floras, supporting the conclusions from previous chapters of the relative uniqueness of the serpentine flora.

Gamma diversity (Between site diversity)

Magurran (2004) defines gamma diversity as inventory diversity within a landscape. The term gamma diversity according to Cody (1986) is used to describe the difference in turnover between serpentine sites. Turnover between serpentine sites, either measured as β_W or as the similarity between sites (C_S), shows that sites that are closer together are more similar to each other. The floras of the Groenvaly (GV) and Rosentuin (RT) sites show the lowest turnover ($\beta_W = 0.650$) (Table 5.5) or highest similarity (35%) (Table 5.6). The sites with the greatest distance between them i.e. Magnesite Mine (MM) and Kalkkloof (KK) are the least similar to each other (9.4%).

The values of species turnover and species similarity at each sampled site were found to be strongly correlated with the geographic distance between the sites (Figure 5.7). The data show that the mean turnover ($\beta_W = 0.800$) between serpentine sites is significantly

Table 5.5: Matrix of turnover diversity using Whittaker's Beta diversity measure (β_W), for pairs of serpentine sites. The highest turnover between sites is indicated in bold, and the lowest is underlined.

	KK	**MC**	**MM**	**GV**	**SM**	**RT**
CZ	0.817	0.746	0.811	0.823	0.838	0.830
KK	-	0.843	**0.906**	0.746	0.830	0.752
MC		-	0.789	0.725	0.898	0.775
MM			-	0.860	0.887	0.842
GV				-	0.660	<u>0.650</u>
SM					-	0.767

Table 5.6: Matrix of Sørensen's similarity index calculated for pairs of sampled serpentine sites. The highest similarity between sites is indicated in bold, and the lowest is underlined.

	KK	**MC**	**MM**	**GV**	**SM**	**RT**
CZ	18.3	25.4	14	17.7	16.2	17
KK	-	15.7	<u>9.4</u>	25.5	14	24.9
MC		-	21.1	27.5	21.1	22.5
MM			-	18.9	14	15.9
GV				-	34.1	**35**
SM					-	23.3

higher than the mean turnover ($\beta_W = 0.714$) measured between each site and its surrounding non-serpentine area ($P < 0.001$). The vegetation on each serpentine outcrop is thus more similar to its surrounding non-serpentine vegetation than to other serpentine sites. This dissimilarity increases with increasing distance between serpentine sites.

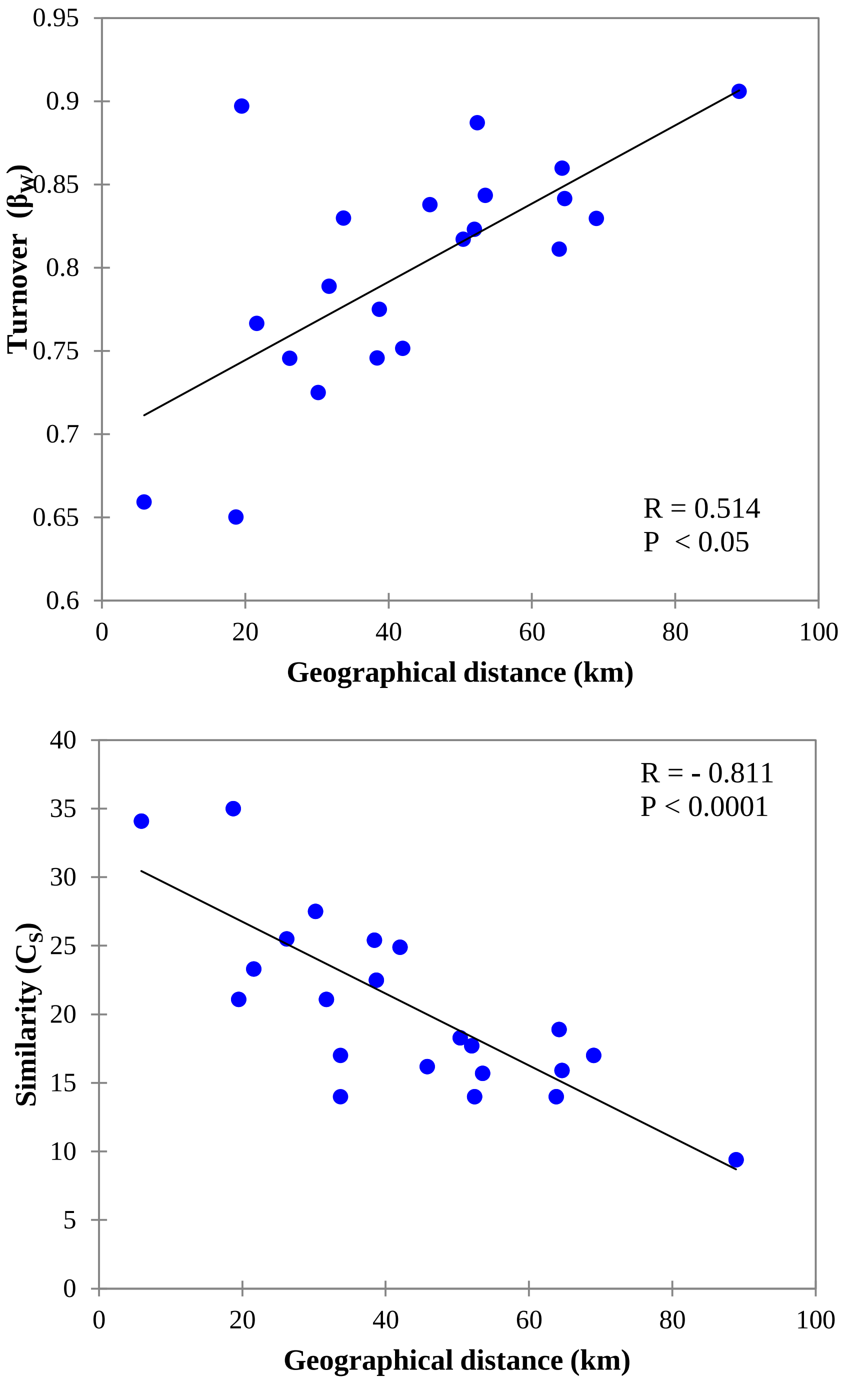

Figure 5.7: Graphical representations of the Pearson Correlations between matrices of turnover (β_W) and similarity between serpentine sites (C_J) and geographical distance between the sites.

In an investigation to determine whether the geographical distance and β-diversity correlation of a variety of organisms across Britain are related to dispersal ability, it was concluded that dispersal ability is not a major factor determining turnover with distance (Harrison *et al.* 1992). However, β-diversity is a function of the organisms' niche breadths combined with the spatial structure of their environment (Harrison *et al.* 1992). The low similarities between serpentine sites suggest that the ranges of plants are quite discontinuous across the Barberton Greenstone Belt. This high β-diversity is possibly due to many of the taxa having narrow tolerances for varying habitat factors. The distributions of plants on serpentine exhibit a heightened sensitivity to climatic variation, leading to a greater change in community composition for a given degree of change in average rainfall, in comparison with floras on other soils (Harrison *et al.* 2002).

Analysis of the checklists of 10 serpentine outcrops reveals that 59% of the taxa recorded occur only on a single outcrop. Of the remaining taxa, 31.5% show a relatively narrow tolerance to altitude (Figure 5.8), 24% to rainfall, 29% to minimum temperatures, 23% to maximum temperatures, 33% to latitude and 24% to longitude of the outcrop. Additional graphs of species tolerances to rainfall and minimum temperatures are shown in Appendix F. More than 50% of these taxa show a relatively narrow tolerance to two or more environmental conditions. This is expected as many environmental variables, including physical variables, act in combination to determine species distributions (Brown 1984). The large numbers of taxa that occur at only a single site and those with apparent narrow environmental tolerances, contribute to the high turnover between the serpentine outcrops of the Barberton Greenstone Belt.

Conclusion

Within site β-diversity has been shown to be relatively low, compared to that of surrounding non-serpentine areas. Differences in β-diversity have been shown to have some correlation with changes in elevation between plots, which confirms findings on serpentine floras in California. However, the elevation correlation is specific to the woody component of the vegetation. Correlations between soil chemistry and the β-diversity of species within the Poaceae were found. These results suggest that the factors determining β-diversity within sites are specific to various components of the .

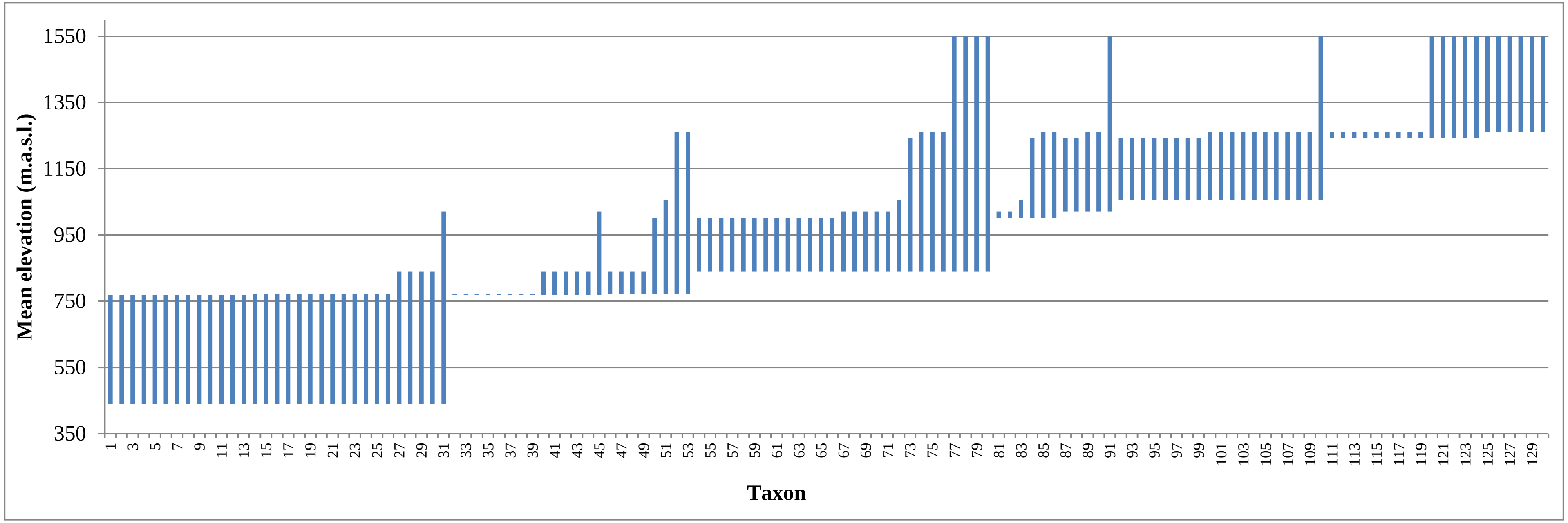

Figure 5.8: Graph showing the altitudes at which the plant taxa with apparent narrow altitude tolerances have been recorded. The numbered plant taxa are listed in Appendix F.

vegetation and could also be different for each serpentine outcrop of the Barberton Greenstone Belt. There is a high turnover of species composition across the serpentine /non-serpentine edaphic boundary, but this has been found to be unrelated to nickel and chromium concentration as well as calcium to magnesium ratios.

The aim of this study was to compare patterns of β-diversity within serpentine sites and between the serpentine outcrops and the surrounding non-serpentine areas to assess these sites for conservation importance. It was also intended to compare the patterns of β-diversity with those of other areas around the world. However, it was difficult to compare the values for the different types of turnover diversity with other studies as analysis of the literature revealed that this study is unique by investigating the β-diversity patterns of the serpentine flora in this way. Within site β-diversity was determined for the serpentine outcrops of the Siskiyou Mountains and compared to the vegetation on surrounding gabbro (Whittaker 1960) and the relationship between productivity and the β-diversity of the herbaceous vegetation of the serpentine outcrops have been investigated (Harrison *et al.* 1992, 2006). However, there is no other in-depth investigation of the β-diversity patterns within serpentine outcrops and between outcrops.

The flora of each serpentine outcrop is relatively unique with low levels of species turnover occurring between the individual serpentine sites sampled. This turnover is strongly related to the geographical distance between sites. This study has provided much fundamental data and hypotheses that now need to be further developed and tested using data from additional outcrops. Many of the outcrops of the Barberton Greenstone Belt have yet to be described and investigated in the same way. A further hypothesis requiring corroboration is that serpentine sites, which are geographically clustered, share more species than those that are more isolated.

The hypothesis that a larger proportion of the serpentine vegetation has narrow tolerances to environmental conditions than the flora of the non-serpentine vegetation, supported by the data presented here, can be further tested as the floras of more ultramafic outcrops of the Barberton Greenstone Belt are documented and investigated.

It has been demonstrated that the conservation of the endemic-rich flora of the Californian serpentines requires a network of sites to capture its among-region or spatial component of diversity. High plant species turnover values between sites recorded for the serpentine sites of the Barberton Greenstone Belt suggest that any conservation plan will need to include a network of sites (Harrison and Inouye 2002). The conservation plan for the serpentine outcrops of the Barberton Greenstone Belt will be developed further in Chapter 7.

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Chapter 6

Does the higher taxon richness of serpentine outcrops act as a surrogate for plant species richness?

Introduction

The development of conservation plans for the vegetation adapted to grow on ultramafic soils was identified as a research priority by delegates of The First International Conference on Serpentine Ecology held in 1991 (Kruckeberg 1992). However, more than 20 years later, over 65% of the serpentine vegetation of the outcrops of the Barberton Greenstone Belt is not yet formally protected. Chapter Three highlighted the need for conservation of the serpentine outcrops as many are at risk due to anthropogenic factors, which could put some of the rarer species at risk of extinction. It is possibly unrealistic to aim to protect and manage all of the more than 30 large outcrops that make up the Barberton Greenstone Belt. Thus, conservation activities must be prioritized so that scarce funds and resources are used efficiently and effectively to prevent long-term loss and degradation of biodiversity and ecological systems (Wilson *et al.* 2009). Currently, the conservation of the unique vegetation of the serpentine outcrops of the Barberton Greenstone Belt is being hindered by a lack of resources and by incomplete or non-existent lists of species and endemic taxa for each outcrop.

Outcrops with high species diversity, with large numbers of endemic taxa and that are relatively unique in their species composition should be prioritised for conservation action. The estimated species diversity of the seven serpentine outcrops sampled and analysed in Chapter 4 has not reached an asymptote. Therefore, even after intensive collection and documenting of the flora the actual number of species occurring on each serpentine site has not yet been accurately determined. It has been suggested that one possibility in which the biodiversity of different areas may be compared is by using measures based on the number of higher taxa present in each (Williams and Gaston 1994). These authors report that family richness is a good predictor of species richness for a variety of groups and regions. If higher taxa show patterns similar to those of species, they can be used in rapid biodiversity assessments to rank a large number of sites according to their relative biodiversity value based on higher taxon richness (Heino and Soininen 2007). Using higher taxon richness to predict species richness would be substantially cheaper and easier to identify specimens from a survey to the level of higher taxon than to species level. Thus, using higher taxa richness as a surrogate may make field surveys and data processing more efficient and cost effective (Balmford *et al*. 1996a). This is based on three underlying assumptions: there are fewer families and genera than species, the sampling effort required to achieve a representative sample is lower for families and genera compared with species and the distribution of species within families and genera is relatively homogeneous (Mandelik *et al*. 2007).

Other surrogates for species richness include environmental indicators (Faith *et al.* 2004), taxonomic indicators (Duelli and Obrist 2003) and subsets of taxa expected to reflect wider patterns of diversity (Pearman and Weber 2007). However, using higher taxa as surrogates for species has the advantage of studying the focal taxa directly, avoiding the use of indirect measures (Mandelik *et al.* 2007) and using higher taxa has been found to be more effective than environmental data (Rodrigues and Brooks 2007). Much of the research on the use of higher taxa as surrogates of species has focused on wide spatial scales i.e. regional (e.g. 100 x 100 km; Balmford *et al.* 1996a) to global scales. At these scales, genus richness has been found to be a better predictor of species richness than family richness (Balmford *et al.* 1996a, Villaseñor *et al.* 2005). The large units used in many analyses are very much larger than those assessed in conservation planning. Therefore, direct inferences from these studies may not be valid as they were conducted at regional and global scales and relied on the assumption that community structuring processes act similarly at all scales (Mandelik *et al.* 2007). This is an important limitation. Any small-scale differences in the speciosity of higher taxa could be masked in comparisons of extremely large units, yet might introduce substantial noise in species to higher taxon relations across sites the size of protected areas (Balmford *et al.* 1996a). At large spatial scales, concurrence between patterns of richness, rarity, and composition of species and higher taxa is generally high (Mandelik *et al*. 2007). Only a few researchers (Balmford *et al.* 1996b, Mandelik *et al.* 2007), however, have examined this relationship at the local scale, which is frequently the relevant scale in land-use conflicts. There are no studies that correlate the patterns of richness at species level with those of higher taxa for floras of ultramafic outcrops.

This chapter aims to determine whether higher taxa (i.e. family and genus) diversity can be used as a reliable surrogate for species diversity of the serpentine outcrops of the

Barberton Greenstone Belt. Using higher taxa as surrogates could speed up the process of prioritising areas for conservation. It is predicted that genera would be good predictors of species richness on the serpentine sites of the Barberton Greenstone Belt, but that families will be less reliable predictors of species richness.

Materials and Methods

The same sample set that was used to calculate species diversity in Chapter 4 was used to assess the diversity at family and genus level. This ensured that all data sets have the same sampling intensity. Numbers of families and genera were recorded in each of the ten 1 m^2 plots from the four Modified-Whittaker plots (Stohlgren *et al.* 1995) placed at each sample site. The presence-absence data for genera and families were analysed using the software EstimateS (version 9.1.0, Colwell 2013) to calculate the richness for each taxon level. To facilitate comparisons with species richness, the genus and family richness were determined using the same parameters as those selected for the calculation of species richness in Chapter 4. Unidentified specimens were included in the analysis if they were positively placed into a family, but excluded if not so. The output data produced by EstimateS allowed for the quantification of the expected number of families and genera at each site and were used to produce cumulative richness curves.

In addition, the number of families and genera were determined from the flora inventories compiled for each site. Data from additional sites i.e. Agnes Mine, Kaapsehoop (Williamson 1994) and Dunbar (Changwe and Balkwill 2003) were included to extend the data set.

The ability of the various measures of higher-taxon richness to predict species richness was determined by plotting the number of higher taxa against the number of species from each site. The slopes of the graphs were assessed using standard regression techniques. It was assumed that no adjustment for the size of sample sites was needed as species diversity has been shown not to be correlated with outcrop area (Chapter 4).

Results and Discussion

The numbers of taxa predicted by the cumulative richness curves are lower than those determined by the flora inventories for each site. The 1 m^2 plots within the Modified-

Whittaker plots represent only a subset sample of the whole flora. However, the slopes of the cumulative richness curves allow comparison of the richness at different hierarchical levels for the serpentine flora and the surrounding non-serpentine areas.

The ability of genus richness curves to predict species richness varies from one serpentine site to another. The cumulative richness curves obtained using the presence-absence data from the sample plots show that the richness curves for genera follow the species richness curves (Figure 6.1). These richness curves for genera could be used to predict the differences in species richness between serpentine sites and their neighbouring non-serpentine areas for four of the sample sites (i.e. CZ, KK, MM and GV). However, the genus richness curves for MC, SM and RT do not seem to be able to predict this difference in species richness.

The slopes of cumulative richness curves (Figure 6.2) were similar for genus and family level and approach zero at a lower sampling effort than the species curves. At this level of sampling the species richness curves for most sites (with the possible exception of KK) have not yet reached an asymptote. For most sample sites the family-level curves level off at about 20 samples, while the genus-level curves level off from 30 samples. The maximum taxon richness for families and genera will be reached with much lower sampling effort than that of species richness when sampling the flora using the 1 m^2 plots from the Modified-Whittaker plots, but this will result in an underestimation of the species richness if families or genera were used as surrogates. In particular, the cumulative richness curves suggest that the species richness will increase with increased sampling, without an associated increase in genus and family richness. When the ratio of species numbers to the numbers of higher taxa becomes high, the latter becomes a poor surrogate for the species numbers (Gaston 2000). This occurs especially when the number of higher taxa asymptotes rapidly and at relatively low species numbers, as shown in Figures 6.1 and 6.2. The ratios of species number to family number are significantly higher than the ratio of species to genus (Table 6.1), further suggesting that family richness is an unsuitable surrogate for species richness.

The distribution of species between genera recorded from the sample plots for each serpentine site is significantly different from those as determined from the flora inventories. For the flora inventories compiled from each outcrop, 66 – 73% of the

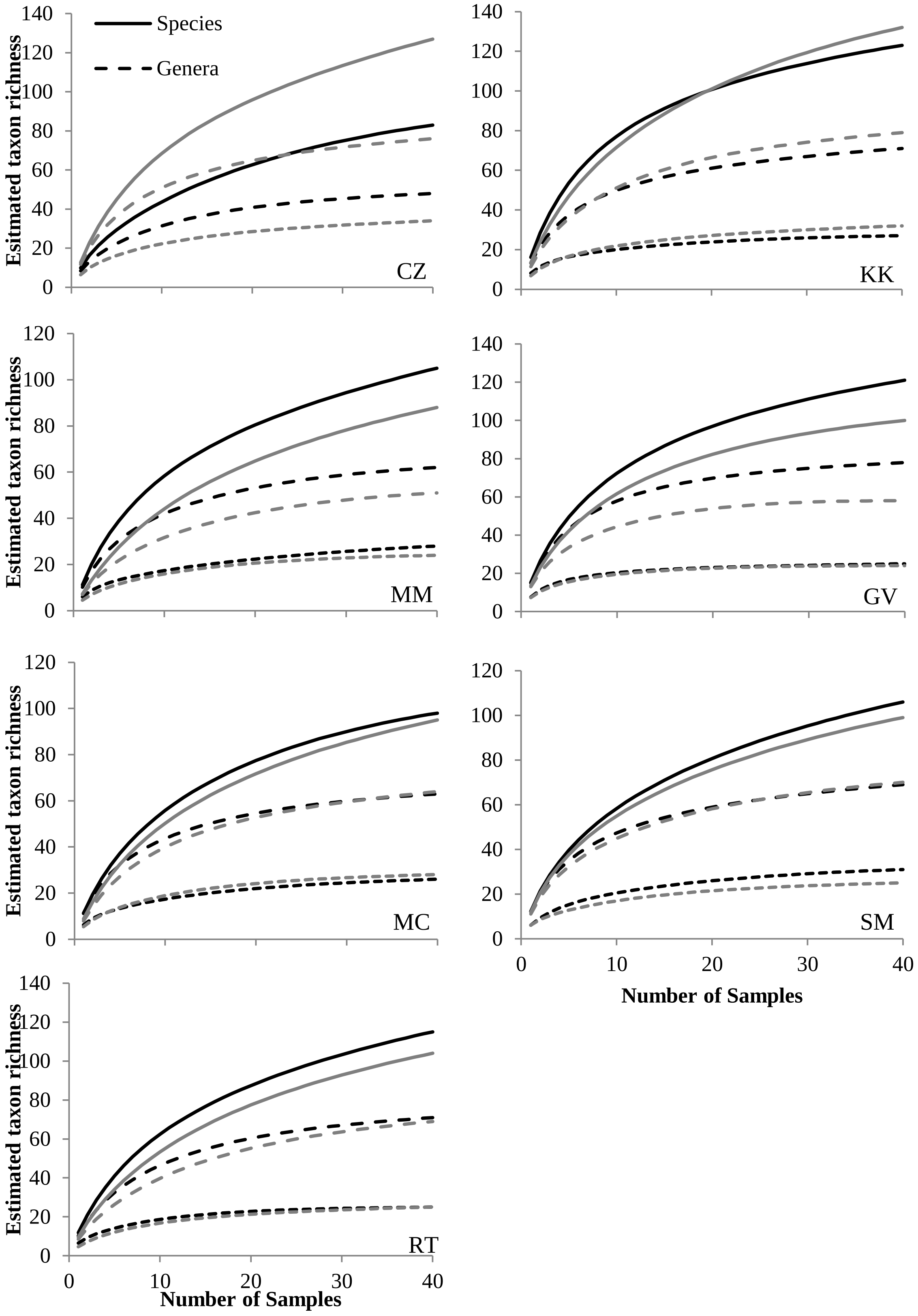

Figure 6.1: Cumulative richness curves estimated for each serpentine site (black lines) and its neighbouring non-serpentine area (grey lines) for species, genera and families.

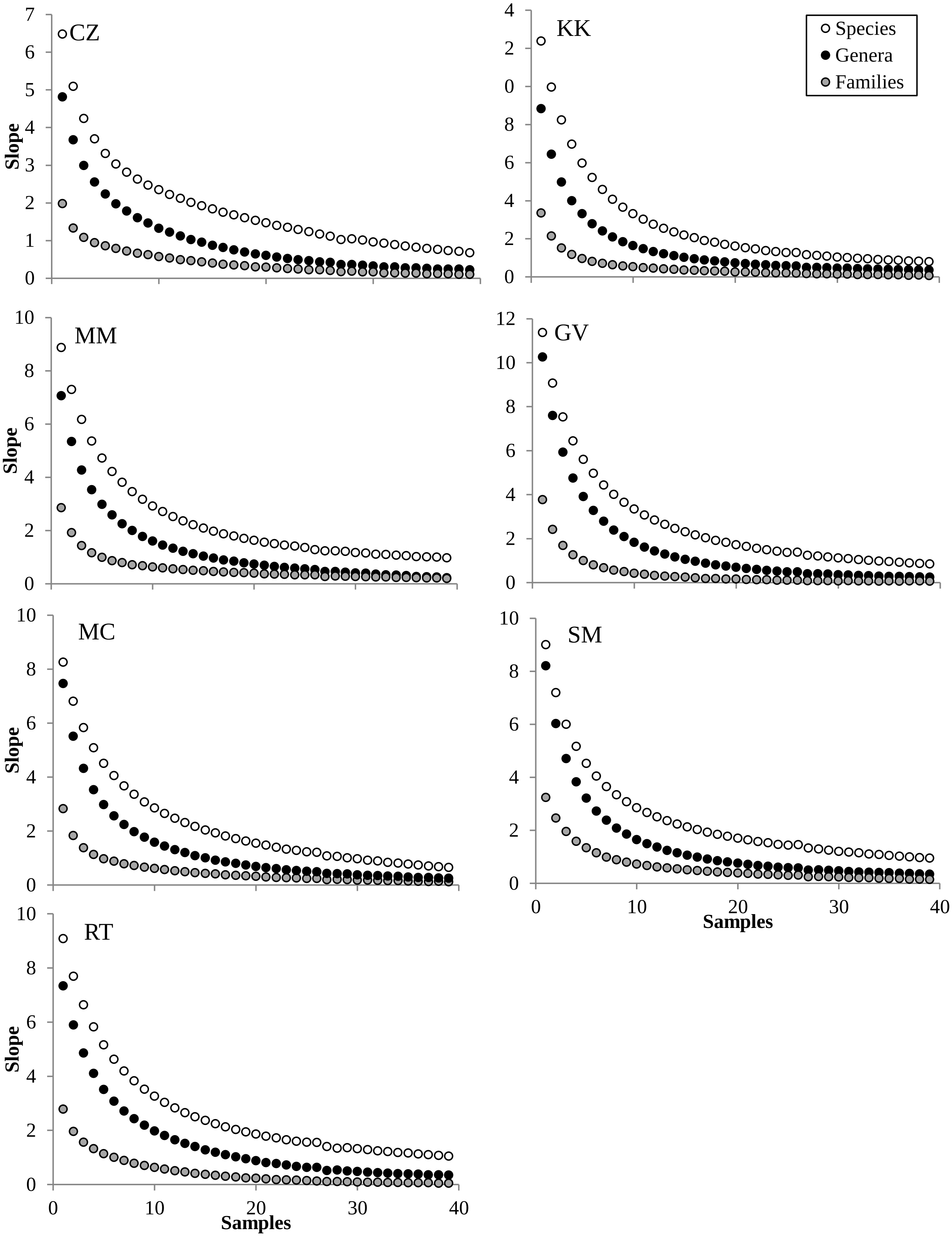

Figure 6.2: Slopes of cumulative richness curves for each serpentine site.

Table 6.1: Numbers of taxa estimated from samples and those determined from flora inventories of each serpentine site.

Site	Richness predicted from accumulation curves			Species: Genus	Species: Family	Richness obtained from flora inventories			Species: Genus	Species: Family
	Species	Genera	Families			Species	Genera	Families		
CZ	83	48	22	1.73	3.77	162	117	46	1.38	3.52
KK	123	71	27	1.73	4.56	182	120	41	1.52	4.43
MM	105	62	28	1.69	3.75	179	122	48	1.47	3.73
GV	121	78	25	1.55	4.84	285	185	61	1.54	4.67
MC	98	63	26	1.56	3.77	206	142	47	1.45	4.38
SM	106	69	31	1.54	3.42	151	111	42	1.36	3.6
RT	115	71	25	1.62	4.60	184	126	45	1.46	4.09
KH*	-	-	-	-	-	181	137	54	1.32	3.35
AM*	-	-	-	-	-	210	152	57	1.38	3.68
DB**	-	-	-	-	-	254	172	63	1.48	4.03

*(Williamson 2004)

**(Changwe and Balkwill 2003)

genera were monotypic and 92 – 96% of genera had one to three species. The taxon lists compiled from the Modified-Whittaker plots recorded 68 – 78% monotypic genera and 95 – 100% of the genera had one to three species (Figure 6.3).

The significantly large difference (P <0.001) in the number of genera with one to three species recorded by the sampling and the flora inventories has resulted in the differences in the species: genus ratios (Table 6.1). In addition, the inventories have recorded a greater number of genera with more than three species than the plots (Figure 6.3). The greater effort used in collecting specimens for complete inventories has probably resulted in a more accurate depiction of the distributions of species within genera. In addition, the difference between species to genus ratios determined from the data from the flora inventories and the plots may occur because the 1 m^2 plots are not randomly distributed but are clustered within a Modified-Whittaker plot and with relatively large distances between clusters.

The species to family ratios (Table 6.1) are very similar and are not significantly different (P=0.4) for the two different sampling methods. In addition, the species to family ratios show greater variation between sites for both sampling methods than the species to genera ratios. The flora inventories record 69 – 79% of the families having one to three species, while 71 – 83% of the families recorded within the plots have one to three species. The flora inventories record 36 – 56% of the families as monotypic while 36 – 53% of the families recorded within the plots are monotypic.

Correlations between species and genus richness were significant for both richness determined by flora inventories (R = 0.976, P<0.001) (Figure 6.4) and for richness as determined by accumulation curves (R = 0.933, P<0.001) (Figure 6.5). The correlations between species and family richness for the numbers of taxa from flora inventories was also significant (R = 0.835, P<0.001) (Figure 6.4). These results show that at a local scale, the generic richness of serpentine vegetation is positively related to its species richness. These strong correlations suggest that genus richness is a suitable surrogate for determining differences in species richness between serpentine outcrops and ranking serpentine sites by increasing species richness. This could be done using either sampling or inventories as a method of determining taxon numbers.

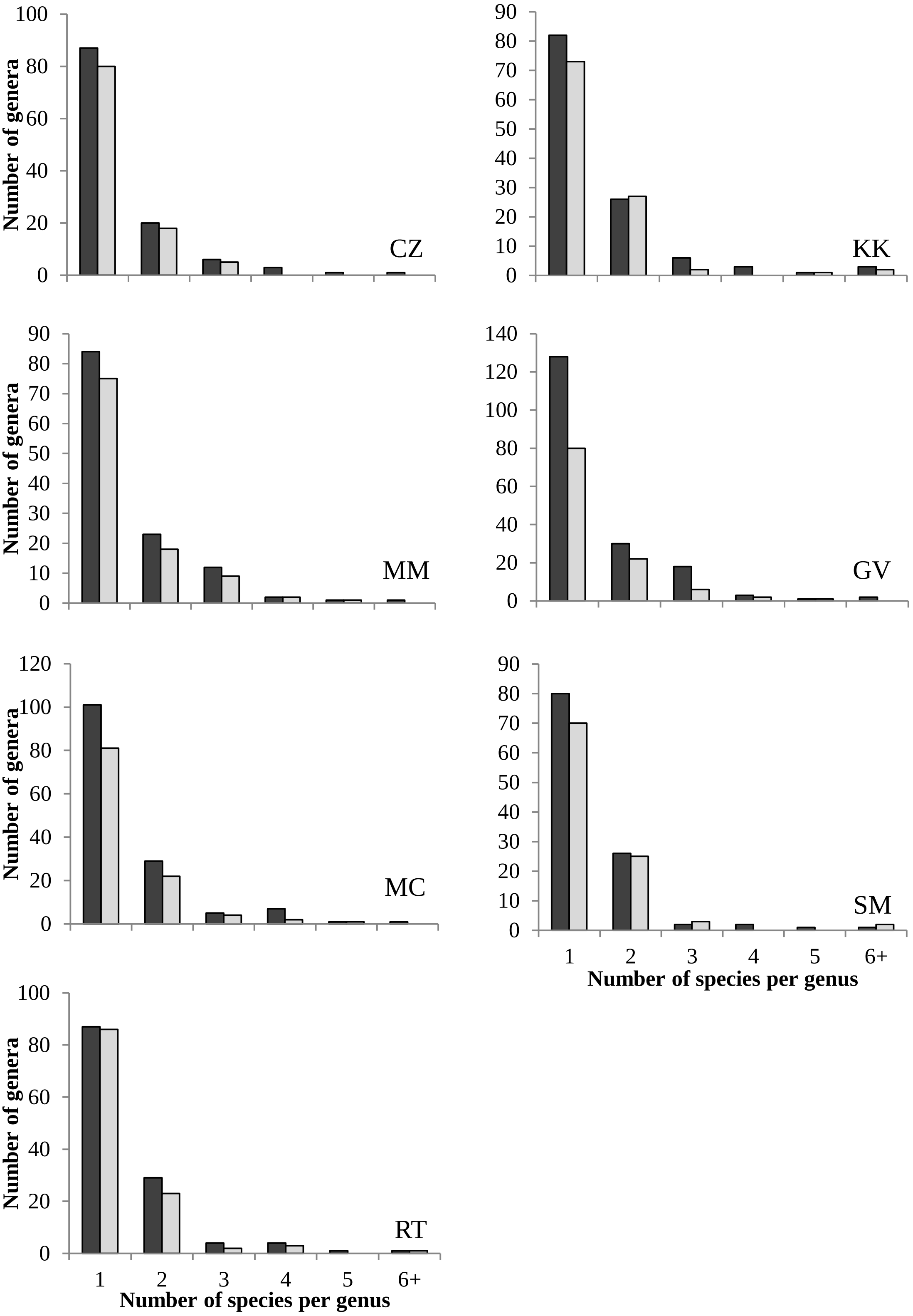

Figure 6.3: Graphs showing the comparison of the distribution of species between genera recorded by the flora inventories (black bars) of each serpentine outcrop and the Modified-Whittaker plots placed at each site (grey bars).

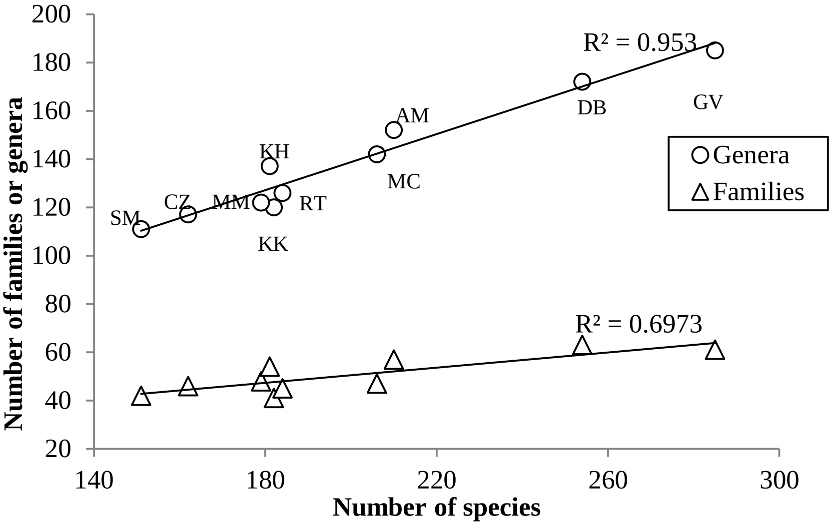

Figure 6.4: Graphs showing a correlation of the number of species against number of families (△) and genera (○) determined from the flora inventories of individual serpentine outcrops. Data points from each serpentine outcrop are labelled with the abbreviation of the outcrop name.

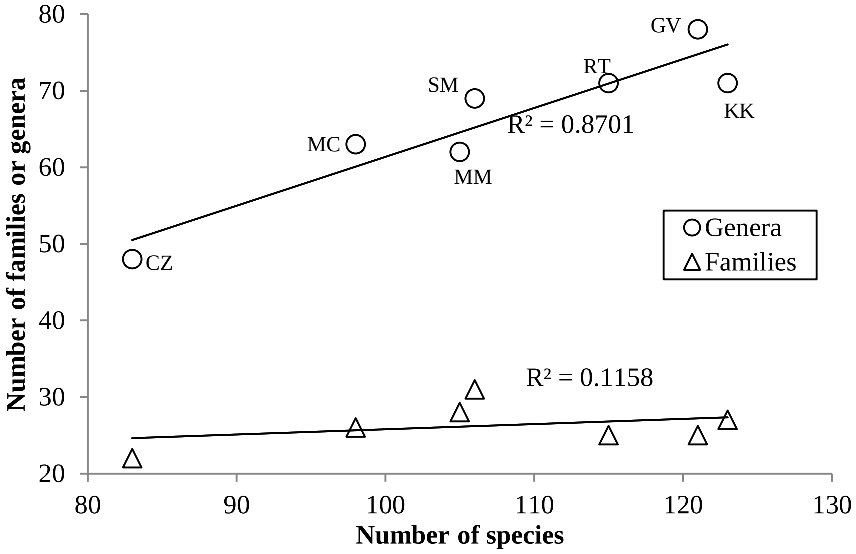

Figure 6.5: Graph showing the correlation between numbers of species and numbers of families (△) and genera (○) using data from the subplots of the Modified- Whittaker plots. Data points from each serpentine outcrop are labelled with the abbreviation of the outcrop name.

The strong correlation between family and species richness for the numbers of taxa determined by inventories of the floras (Figure 6.5) suggest that this method could also reliably be used to predict the species richness of the serpentine sites. However, the very low correlation between the richness of families and species ($R = 0.340$, $P<0.01$) (Figure 6.5), confirms that family richness is not a reliable surrogate for species richness when the flora is sampled using smaller plots. It has been suggested that evolutionary radiation within insular biota, which include the floras of serpentine outcrops, would lead to very high ratios of numbers of species to numbers of higher taxa (Gaston 2000). These high ratios would indicate a lower effectiveness of using higher taxa, especially above the genus level, as surrogates for species richness.

Similar results were reported in a study of diversity patterns of three commonly used surrogate taxa: vascular plants, beetles and moths in the Jerusalem Mountains, Israel. Cumulative richness curves of species and genera showed similar patterns, levelling off at equivalent sampling efforts. The data show that genus-level assessments were a reliable surrogate for local patterns of species richness, rarity, and composition, but family-level assessments performed poorly (Mandelik *et al.* 2007). The species-higher taxon links at the local level of individual serpentine outcrops reported here are in fact almost identical in strength to those recorded in equivalent analyses of angiosperm diversity at a regional scale (Balmford *et al.* 1996a).

As the correlation between genus richness and species richness is stronger when using the flora inventory data as opposed to the data from the 1m^2 plots, using genus numbers from flora inventories for each serpentine outcrop would be a more reliable means of predicting the species richness value of individual outcrops for conservation purposes. However, the strong correlations between genus and species richness determined from the 1m^2 plots within the Modified-Whittaker plots suggest that, if time and resources are limited, sampling the serpentine flora in this way could still be used to determine genus richness as a substitute for species richness. Sampling a subset of the serpentine flora would save time, money and effort when compared to the cost and effort required for the compilation of complete genus lists. Making a full flora inventory, even with identifications to genus level only, of each serpentine site would require more field work than sampling a subset of the flora using plots. This would be valuable for ranking serpentine outcrops with increasing species richness to aid in choosing sites of higher diversity for conservation purposes. However, it would not be reliable to use this method as a means of predicting the species richness value of individual areas. Despite the significant relations between species richness and genus and family richness, the precision with which absolute species richness can be predicted from higher-taxon richness is low.

Conclusion

The different serpentine sites of the Barberton Greenstone Belt vary considerably in their overall plant richness at all hierarchical levels. The analysis of the higher-taxon approach for the serpentine outcrops suggests that genera have some potential for facilitating the ranking of outcrops in terms of biological richness to select sites for conservation planning. The numbers of plant species were found to be closely related to genus richness and weakly to family richness. These results mirror patterns seen at a regional scale of analysis (Balmford *et al.* 1996).

Using genus richness as a surrogate for species richness could save substantial time and money. Although the time spent completing the collections for measuring genus richness as opposed to species richness would be similar in the field, identifying specimens to genus level can often be completed in the field. Identifying specimens to species level may necessitate additional time spent on identification work in an herbarium (Balmford *et al.* 1996b). Targeting woody plant families and genera rather than species while cataloguing biological diversity in the forests of Sri Lanka reduced survey costs by 60% and 85%, respectively (Balmford *et al.* 1996b). These researchers determined that using genus rather than species data had virtually no effect on the representation of species in priority sites.

Using samples of the vegetation from plots such as Modified-Whittaker plots to survey diversity at coarse taxonomic scales would not reduce overall sampling effort. In addition, sampling the flora by collecting data from subplots reduced the accuracy of the richness analyses substantially and therefore complete inventories would be needed.

The outcrops analysed in this study are all relatively far apart geographically. Further analyses are required on outcrops nearer to those studied here to determine if outcrops within close proximity show similar levels of richness at both species and genus level. If sites are relatively near to one another (as may be expected if they are alternative candidates for protected area development), variance in site richness will probably be lower (Balmford *et al.* 1996a).

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Chapter 7

Conservation of the serpentine areas of the Barberton Greenstone Belt

Introduction

Globally, there is growing emphasis on biodiversity conservation. This is as a response to increasing degradation and consumption of the world's natural resources. In South Africa, loss and degradation of natural habitat are the biggest pressures on biodiversity. They are heightened by the strong and expanding economy and the drive for job creation through growth. The resultant pressure on our globally recognised biodiversity is marked and widespread (Ferrar and Lötter 2007). It is therefore necessary to prioritize the conservation of biodiversity in the selection of conservation areas (Lötter *et al.* 2014).

The province of Mpumalanga has a high diversity of plant species. It comprises only 3% of Southern Africa's surface area, yet supports 21% of its species diversity (Emery *et al.* 2002). However, a history of mining, plantations of *Eucalyptus* spp. and *Pinus* spp., extensive agriculture, urban development and invasion of alien plants has increasingly put the natural habitats under pressure. Mpumalanga has 117 protected areas, which include state lands, private and provincial nature reserves and national parks. In South Africa protected areas are defined as parts of the landscape that are formally protected by law in terms of the Protected Areas Act, and managed primarily for the purpose of biodiversity conservation (Lötter *et al.* 2014). Protected areas comprise 19.7% of the Province, but only 7.7% if the Kruger National Park contribution is excluded (Lötter *et al.* 2014). Most of these protected areas were not established with the aim of conserving a representative sample of biodiversity. Many of the protected areas were created on lands of low biodiversity value in scenic or recreationally attractive areas or for the protection of a few charismatic animal species (Ferrar and Lötter 2007).

A Biodiversity Conservation Plan for the Mpumalanga province was produced in 2007 (Ferrar and Lötter 2007) and was updated in 2014 (Lötter *et al.* 2014) to guide conservation and land-use decisions in the future. These plans focus on the conservation of biodiversity of the Province but do not include the conservation of endemism as part of the plan. The link between endemism and conservation was expressed by Takhtajan

(1986) in this way: “The choice of territory to be protected can be made by means of very diverse criteria, ranging from the extremely practical to the purely aesthetic. But no matter what the path leading to the choice of a protected territory, it is necessary everywhere, in every part of the world and with every environment, to protect from the outset the flora of those territories which represent the richest storehouse of the world’s unique genetic material—namely, the endemic forms of life.”

Although serpentine soils occur all over the world, there is only one report of actions being taken to conserve these unusual areas. The conservation of serpentine sites of California was discussed in 1986 at a conference on Rare and Endangered Plants (Reeves 1992), and a systematic conservation plan has been completed for these sites (Thorne *et al.* 2011). A‘Resolution’ was passed by the delegates of The First International Conference on Serpentine Ecology held in 1991. This resolution supports the conservation of the vegetation of serpentine areas worldwide (Kruckeberg 1992). This resulted in a number of papers on conservation and management of serpentine endemics and their habitats (Wolf 2001, Mattner *et al.* 2002, Whiting *et al*. 2004, Selvi 2007, Thorne *et al.* 2011 and García-Barrisuso *et al.* 2012). Some of these also reported on a large number of rare and endemic plants that were threatened by mining activities, industry and recreational traffic. However, implementation of preservation protocols suggested in these reports has lagged, and there are a large number of sites that need to be conserved and are still left unprotected. Although there are also reports of mines that are revegetating mine spoils with serpentine species and a few other areas which have been protected (Kruckeberg 1992).

Despite considerable conservation action in the eastern parts of the Mpumalanga Province, there has been no deliberate selection of serpentine outcrops for conservation. Many of the serpentine outcrops and their floras are threatened by a number of activities (Williamson and Balkwill 2006). The vegetation type found on the serpentine outcrops i.e. Barberton serpentine sourveld as described by Mucina and Rutherford (2006) is classified as vulnerable (Lötter *et al.* 2014). The vulnerable classification is selected for a vegetation type by determining that ecosystem functioning will be compromised if habitat loss continues.

Serpentine outcrops around the world have been shown to support distinctive floras and communities that are markedly set apart from those on adjacent non-serpentine substrates (Kruckeberg 1992). The floras of the serpentine outcrops of the Barberton Greenstone Belt also show high plant diversity and species richness (Chapters 2 and 4) and support many rare and endemic species and especially hyperaccumulators of heavy metals (Chapter 3). Many of the endemic plant species have particularly small geographic ranges and are thus deemed worthy of conservation due to their rarity. These narrow endemics are already endangered by their limited ranges and are thus more liable to extinction due to climate change (Boyd *et al.* 2009).

The metal tolerant species occurring on serpentine outcrops need to be preserved as they are the optimal choice for site restoration at mine closure and for the rehabilitation of metal-contaminated land (reclamation and rehabilitation) (Whiting *et al.* 2004). Species that are hyperaccumulators of nickel or other minerals can be used for the bio-recovery or phytoextraction of minerals from soils. However, rare and endemic taxa alone cannot be conserved because these taxa, together with common and widespread species, exist within ecosystems or communities (Vane-Wright *et al.* 1991). Therefore, once an area has been identified as a priority area, the whole flora of this area needs adequate protection as a functional ecosystem.

There are a large number of different conservation priority-setting approaches described in the scientific literature (Rebelo 1994, Lombard *et al.* 1999, Rodrigues and Gaston 2002, Brooks *et al.* 2006, Wilson *et al.* 2009). Each of these approaches aims to identify where, how and based on what measure we should act to conserve biodiversity efficiently, based on the assumption that costs are limited and not everything can be conserved (Wilson *et al.* 2009). Much of describing conservation priority-setting deals with the identification of new protected areas or networks of protected areas. This is referred to as systematic conservation planning (Margules and Pressey 2000).

The aim of this chapter is to review the current conservation status of the serpentine outcrops of the Barberton Greenstone Belt. In addition, sites with high conservation priority will be identified. It has been demonstrated that the conservation of the endemic-rich flora of the Californian serpentine requires a network of sites to capture its among-region or spatial component of diversity. Small isolated sites showed increased β-diversity (i.e. between site diversity and decreased within- outcrop diversity)

(Harrison 2002) as a result of more frequent extinction and lower colonization rates (MacArthur and Wilson 1967). It is predicted that due to the high between site diversity, an assemblage of serpentine outcrops of the Barberton Greenstone Belt would need to be protected to conserve the maximum regional diversity of these outcrops.

Materials and Method

All the serpentine outcrops shown on a geological map (Ward 2000) were digitised by ArcView 3.2. The resultant shapefile was overlaid onto the Mpumalanga Biodiversity Sector Plan (Lötter *et al.* 2014) web map (http://conservation3.arcgisonline.com/Apps/MBSP) to determine the extent that each serpentine outcrop is found within a protected area. This map was also used to determine which sites are found within Critical Biodiversity Areas (CBAs) and Ecological Support Areas (ESAs) as defined by Lötter *et al.* (2014). Additional information regarding threats to the serpentine outcrops was noted during the visits to each site.

Two approaches were taken to determine the priority sites for conservation of the serpentine outcrop. Initially, the preservation of the maximum number of plant species occurring on the serpentine outcrop was the goal while selecting priority sites for conservation. With the second approach, the preservation of the serpentine endemics became the primary focus of the selection procedure. Common species will be caught up in the conservation actions for the endemic species. Considering only species diversity of the serpentine sites as the conservation goal may not include all the endemic species as it has been shown on other serpentine outcrops that areas with low diversity can have a high number of endemics that could be rare and unique (Kruckeberg and Rabinowitz 1985). Rare or threatened species, which most of the serpentine endemic species are, are considered to have higher conservation value that a common species, as they contribute more to regional biodiversity than the ubiquitous species (Duelli and Obrist 2003).

It was decided not to use one of the various computer software packages such as Marxan and C-Plan, for the selection of priority serpentine outcrops as this data set (i.e. number of outcrops under consideration and the species richness and a number of endemics at each site) is small enough to be analysed manually. The planning unit

(Wilson *et al.* 2009) selected was the entire serpentine outcrop as each outcrop has a well-defined edaphic boundary and is separated geographically from other serpentine outcrops. The conservation target was first to include each plant species occurring on serpentine outcrops at least once in the priority sites. In the second instance, the conservation target was to represent each serpentine endemic in the priority areas at least once. This represents a minimum set coverage approach, in which the objective is to find a solution that achieves all conservation targets at minimum costs (Moilanen *et al.* 2009).

To select the priority serpentine sites to meet the biodiversity goal a simple iterative procedure as described in the Marxan online tutorial (http://www.uq.edu.au/marxan/tutorial/module1.html) was used. This involved selecting the most species-rich site first. The site with the next highest species richness of the remaining species was then selected using the principle of complementarity (Moilanen *et al.* 2009) to ensure that each species occurring on a serpentine site was selected at least once. An iterative selection procedure algorithm designed specifically for the prioritization of areas based on endemic species was also used (Rebelo 1994). This method prioritizes sites based on the number of endemic species as well as the rarity of endemic species found at each site.

Results and Discussion

Current conservation status of the serpentine outcrops of the Barberton Greenstone Belt

Currently, ten of the 23 large (> 1km^2) serpentine sites in the Barberton Greenstone Belt are wholly or partially (>50% of the surface area) contained with protected areas (Figure 7.1). Although 30 of the 79 (38%) of the smaller (< 1km^2) sites are enclosed within protected areas, this represents only 4.8% of the total area of all the serpentine outcrops (Table 7.1). Ten of these small sites are found within Songimvelo Game Reserve (SGR), and a further eleven are found within the Barberton Nature Reserve (BNR). Both reserves are managed by the Mpumalanga Tourism and Parks Authority (MTPA). However, the rest of the small sites, especially those in the band between MC and KH, are not within protected areas (Figure 7.1). Similarly, only 29% of the area of the larger sites is found within protected areas. The largest outcrop is partially protected within the Nkomazi Wilderness Nature Reserve, which is privately owned and managed (Table 7.2). The Barberton Nature Reserve protects a number of the larger outcrops,

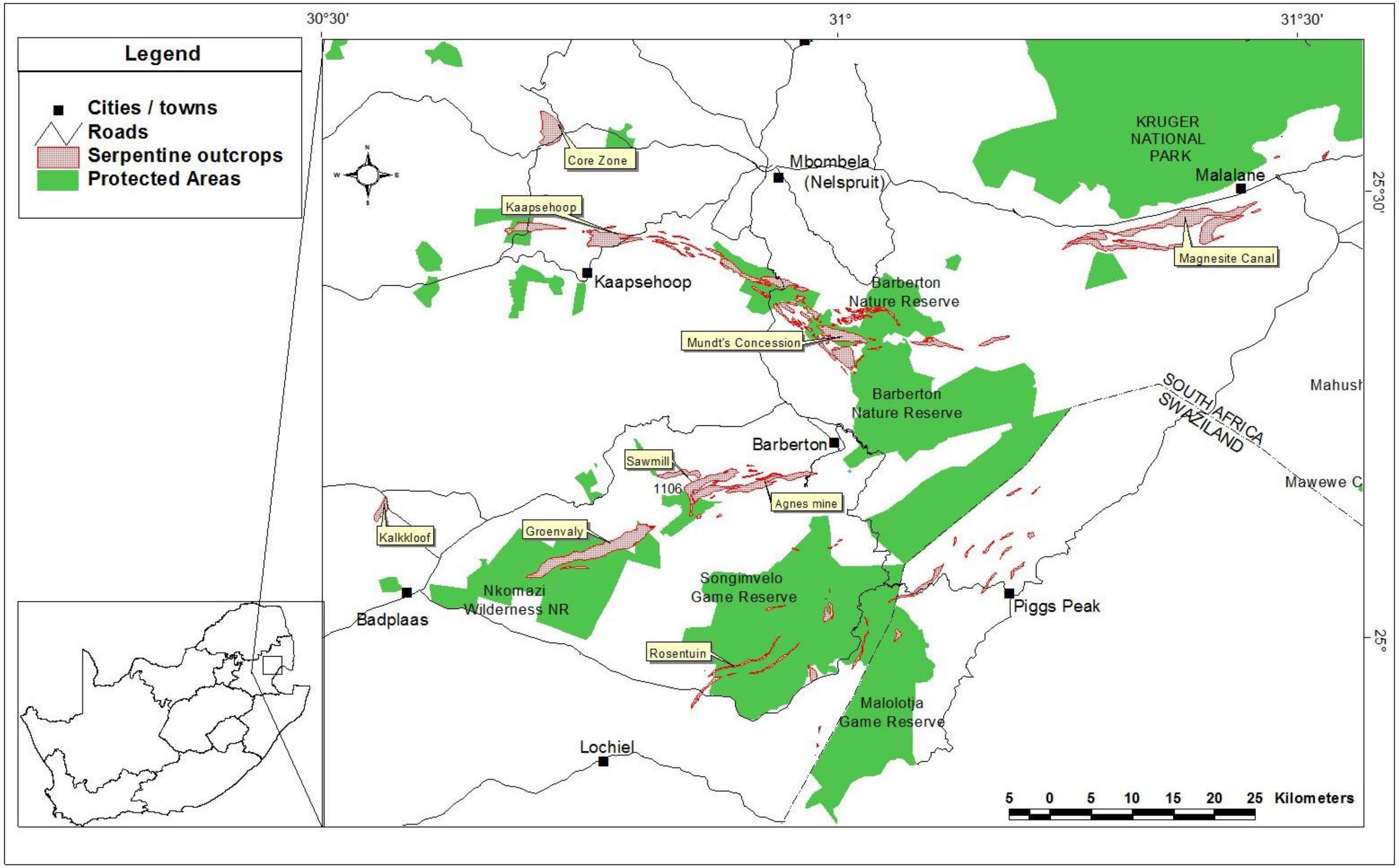

Figure 7.1: Map showing serpentine outcrops overlain with existing protected lands.

Table 7.1: Analysis of the area of the serpentine outcrops of the Barberton Greenstone Belt in Mpumalanga that are found within Protected Areas (PA).

	Number of outcrops	Number of outcrops more than 50% protected	Total area (km^2)	Area protected (km$^{2)}$	% of total serpentine area protected
Small outcrops (< 1 km^2)	79	30	16.7	5.8	4.8
Large outcrops (> 1 km^2)	23	8	106.8	31.5	25.4
All outcrops	102	33	123.6	37.3	30.2

including MC, in the central part of the Barberton Greenstone Belt. However, many of the large southern outcrops remain unprotected (Figure 7.1).

Of the serpentine outcrops that have been studied in detail, three with large numbers of endemic species i.e. AM, SM and KH are inadequately protected (Table 7.2). Each of these sites harbours a species that is endemic to that outcrop only. *Brachystelma dyeri* is endemic to the AM outcrop; *Cyphia bolusi* has thus far only been found on the KH site and *Graderia* sp. nov. is only known from the SM site (Table 3.1).

Thirteen serpentine outcrops are completely or partially (>50%) are protected within the Songimvelo Game Reserve, which is managed by MTPA. This represents 4.2% of the total area of the serpentine outcrops. However, one of these is almost completely planted to *Pinus* sp. and there is very little natural vegetation remaining. Two sites near the Msauli Mines are heavily disturbed, with little effort being made to revegetate mine tailings with serpentine species. The Barberton Nature Reserve, also managed by the MTPA, protects 22 outcrops, representing 11% of the total serpentine area. Although small portions of the AM site are within the Cythna Letty and Thorncroft Nature Reserves, the site is under considerable threat by *Eucalyptus* sp. plantations that have

Table 7.2: Summary of conservation status of selected serpentine outcrops (in decreasing order of species richness) of the Barberton Greenstone Belt in Mpumalanga Province.

Serpentine outcrop	No of endemic species	Approximate % of outcrop within a Protected area	Name of Protected area	Type of Protected area
AM *	11	5	Cythna Letty & Thorncroft Nature Reserves	MTPA
GV	10	75	Nkomazi Wilderness Nature Reserve	Private Nature Reserve
MC	9	100	Barberton Nature Reserve	MTPA
SM	8	5	Queens River Reserve	MTPA
DB **	7	100	Songimvelo Game Reserve	MTPA
KH *	6	0	-	-
RT	5	95	Songimvelo Game Reserve	MTPA
MM	1	0	-	-
KK	0	0	-	-
CZ	0	0	-	-

* Williamson (1994)

** Changwe and Balkwill (2003)

already been planted on parts of the site. Three other sites are under the authority of the South African Government, Department of Agriculture, Forestry and Fisheries (Balkwill *et al.* 1995) but have no protection status. Large parts of Kaapsehoop and sites on the farm Groenvalei are already planted to *Pinus* spp. Sites in the Queens River Valley are planted to *Eucalyptus* spp. The more northern sites are often partially under agriculture or disturbed due to grazing or building of roads and invasion of exotic plants. One site, (MM), in the Kaapmuiden area, is partially protected by a conservancy set up by local farmers, but this area is heavily disturbed by a large magnesite mine.

Priority outcrops for the conservation of the Barberton Greenstone Belt

Ranking the sites by species richness selects all the sites that have high species richness and endemic species. Due to the high turnover in species composition between serpentine outcrops (Chapter 5) the first four sites selected as priority only covered about 65% of all the plant species recorded. Selecting seven priority outcrops out of the ten studied does not meet the conservation goal of each plant species being represented by the selected sites at least once (Table 7.3). Using complementarity when applying the iterative procedure of selecting priority sites for species richness is preferable to ranking the sites based on species alone. The complimentary method selects the MM site with 3rd highest conservation priority. This outcrop has a relatively low species richness (Chapter 4), however, it has a very low similarity to all the other sites and thus a relatively unique species composition amongst the other serpentine outcrops. Therefore, to conserve its unique species composition it is selected early in the iterative procedure.

It was predicted (Chapter 6) that genus richness would be a suitable substitute for species richness to rank sites in order of priority for conservation. The same priority sites were selected when using genus richness in the iterative procedure instead of species richness (Table 7.3). This method has promoted AM above KH in priority but GV, DB and MM are still selected as the top three sites. This confirms that genus richness is a suitable substitute for conservation priority setting when the preservation of species richness is the conservation goal.

One of the constraints of ranking the sites according to species richness and considering the complementarity principle is that the serpentine outcrop RT is excluded from the top priority list (Table 7.3). Five endemic species are found on RT. These include a single known population of *Graderia* sp. nov., which is thought to be serpentine endemic, but further investigation is required to determine its status.

Three outcrops i.e. CZ, KK and MM, are not given a high conservation priority when the conservation goal is the protection of endemics (Table 7.3). No endemic plant species have been recorded from both CZ and KK and the endemic *Macledium zeyheri* subsp. *thyrsifolium* first collected from MM has subsequently also been collected at the MC site. However, excluding MM from the priority outcrop list in this way, only allows for the protection of 78% of the species richness on serpentine outcrops. Selecting all

Table 7.3: Outcrops with high conservation priority as selected using species richness and endemism as conservation goals.

Conservation goal	Serpentine outcrop selected after each iteration							% of plant taxa included in selection
	1st	2nd	3rd	4th	5th	6th	7th	
Species richness	GV	DB	MC	AM	RT	KH	MM	89
Species richness applying Complementarity Principle	GV	DB	MM	KH	KK	AM	MC	90
Genus richness applying Complementarity Principle	GV	DB	MM	AM	KH	KK	MC	95
Endemism and rarity	MC	GV	KH	DB	AM	SM	RT	100 (endemics)

the remaining sites as priority areas would include all the endemic species and multiple populations of the widespread endemic species. Each of the sites selected, except MC, would be considered to be irreplaceable as each outcrop has at least one species that occurs only on that particular site (Chapter 3). MC is considered to be irreplaceable as it is the only selected site that would preserve a population of *M. zeyheri* subsp. *thyrsifolium*. In addition to these sites, a small (10 ha) serpentine outcrop south of Songimvelo Game Reserve is the only site of an endemic species, *Dioscorea strydomiana* (Figure 3.10). This outcrop should thus also be considered irreplaceable and be given high conservation priority.

The selection of MC, GV, KH, DB, AM, SM and RT as priority serpentine outcrops for conservation satisfied the need for connectivity between populations of the endemic species. These sites are relatively close to each other or provide landscape corridors between distant outcrops. This would allow for gene flow between populations of endemics on different outcrops.

It has been demonstrated that the conservation of the endemic-rich flora of the Californian serpentine requires a network of sites to capture its among-region or spatial component of diversity. Small isolated sites showed increased β-diversity, (i.e. between site diversity and decreased within- outcrop diversity (Harrison 2002), as a result of more frequent extinction and lower colonization rates (MacArthur and Wilson 1967). Due to the high plant species turnover between sites recorded for the serpentine sites of the Barberton Greenstone Belt, it is suggested that any conservation plan will need to include a network of sites. At present, protected area only preserve MC, DB and RT and about 75% of the area of GV of the selected priority outcrops (Table 7.2). The rest of the sites remain unprotected. The inclusion of the additional selected sites AM, KH and SM, would create the necessary network of reserves to conserve adequately all the serpentine endemic species.

In a study investigating the reproductive fitness of two species, *Calystegia collina* and *Helianthus exilis* endemic to serpentine outcrops in California, results showed that the reproductive success of some endemic species was significantly higher on large outcrops than on small outcrops (Wolf 2001). These findings suggest that clusters of local populations should be protected for the long term conservation of endemic, selfincompatible plant species. Reproduction of *C. collina* on small outcrops was greatest in areas where several of these outcrops were located within 200m of one another. To conserve a representative complement of genetic diversity in endemic species the conservation of many populations is required as genotypes tend to be unique among populations. Therefore, larger outcrops or clusters of outcrops need to be protected.

Considerations for the conservation of serpentine outcrops in the future

This study has focused on ten of the well analysed large serpentine outcrops. The floras and species richness of more of the serpentine outcrops need to be documented and analysed in order to determine whether other areas also have high conservation priority. It is hoped that once these areas have been documented and analysed in this way the program Marxan (Ball *et al.* 2009), a reserve design algorithm that identifies spatial solutions to user-determined objectives, could be used by conservation agencies to determine the most cost-effective reserve design for the area within which the serpentine areas fall and to guide the inclusion of more serpentine areas into current reserves.

Management of protected serpentine outcrops is necessary to protect the plants, especially the endemic species, from threatening influences such as invasive alien species, overgrazing and illegal collecting of rare and medicinal plants. Many of the plants growing on serpentine outcrops would also be useful for reintroduction and restoration programs for other serpentine areas.

Conclusions

Of the serpentine outcrops considered in this study, only a little over 30% of the surface area is currently found in protected areas. Many of the other sites are threatened by mining activities, invasion of alien species, plantations of *Pinus* and *Eucalyptus* spp. and illegal collections of rare plants. Seven of these sites are considered to be irreplaceable as there are plant species endemic to those particular outcrops. Only four of these outcrops are currently adequately protected within various nature reserves managed by the Mpumalanga Tourism and Parks Authority or by private entities. The remaining sites, AM, KH, SM and MM need to be included in protected areas in order to ensure the survival of their rare and endangered endemic species.

This analysis has focused specifically on plant conservation, but there is very little if any information regarding animals especially invertebrate richness and their need for conservation on these outcrops. Further investigation should be directed at determining whether plant species richness of the serpentine outcrops predicts the richness of other taxonomic groups such as invertebrates. This would improve the case for conservation of these sites. It has been suggested that the practical selection of conservation areas based on data for well-known taxonomic groups can cautiously proceed in the meantime, under the assumption that these areas capture species in less well-known taxa within the same realm (Whiting *et al.* 2004).

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Chapter 8

General Conclusion

To date about 750 serpentine-tolerant plant taxa have been recorded from these selected sites. The species represent 319 genera and 94 families (Chapter 2). Comparisons with the surrounding non-serpentine vegetation highlighted the distinctiveness of the serpentine vegetation in terms of the composition of monocotyledons and dicotyledons and the representation of different families e.g. the increased representation of the Asteraceae on serpentine. This distinctiveness is further highlighted by the exclusion of a large number of plant species from the serpentine outcrops that are commonly found in the surrounding vegetation. Conversely, there are a number of plant species common on serpentine soils but are less common elsewhere, which have been identified as indicators of serpentine soils. The flora of individual serpentine outcrops shows higher affinity with each other than with the surrounding non-serpentine vegetation. However, the relatively large differences between the species composition of each site indicate the uniqueness of the vegetation of each serpentine outcrop.

A hypothesis predicted that the serpentine vegetation would be more species-rich and diverse than the surrounding non-serpentine areas. One site, GV, showed significantly higher species diversity and a few sites, MC, MM, RT and SM, showed slightly (but not significantly) higher species diversity. It is predicted that with increased sampling these differences could become significant as the graphs of serpentine species richness (Chapter 4) had not reached an asymptote in each case. One site, CZ, shows significantly lower species diversity, which could be as a result of its geographic isolation from all the other outcrops. Considerably higher species richness was recorded for the serpentine sites of the Barberton Greenstone Belt when compared to other sites around the world for which comparable data exist.

Estimated richness at the genus level mirrors the patterns of species richness at each serpentine site. Serpentine genus richness is significantly higher at MM and GV and significantly lower at CZ than non-serpentine richness (Chapter 6). Family richness at all sites is similar in serpentine and non-serpentine vegetation. These results show that genera have the potential for facilitating the ranking of outcrops for the setting of conservation priorities. There is a strong correlation between the ranking of outcrops for conservation and using species richness as well as genus richness as a surrogate for species richness (Chapter 7).

The serpentine floras include 33 plant taxa that are restricted to i.e. endemic to serpentine outcrops (Chapter 3). This included five taxa that have been recorded from non-serpentine soils but that are other types of ferromagnesium ultramafic soils. These endemic taxa represent 33% of the endemic or near endemic taxa of the Barberton Centre of Endemism. The families Asteraceae and Anacardiaceae have a significantly higher number of endemic species than expected, particularly in the genera *Berkheya* and *Ozoroa*. Other genera such as *Ocimum* also show higher than expected representation amongst the endemic taxa. No statistical correlation was found between the size and position of the serpentine outcrops and the level of endemism. These results do not support the second hypothesis (Chapter 1) that predicts that endemism would increase with an increase in altitude range, size and habitat diversity of each site.

The composition of endemic species on each serpentine outcrop adds to their distinctiveness. Eight of the known endemics have only been found on a single outcrop and each of these occurs on a different outcrop (Chapter 1). Each of the sites with one or two of these very restricted endemics has high conservation priority as they are considered irreplaceable (Chapter 7). Eight serpentine outcrops (including DB, AM and KH and the Swaziland sites) have more than five endemic species, which gives each of these sites conservation priority. Outcrops that are near one another show greater similarity in endemic species composition while sites that are isolated geographically are less similar.

In answer to the key question (Chapter 1) regarding the biological profile of the endemic taxa, there was no statistical difference found between some characteristics of the serpentine and non-serpentine vegetation. However, there are more species with annual shoots arising from a perennial rootstock than expected. There is also a low over-representation of small shrubs and an under-representation of trees and climbers amongst the serpentine endemic when compared to the surrounding non-serpentine vegetation. A large proportion of the endemic taxa exhibits long-range dispersal mechanisms, which supports the selection of a large number of outcrops for conservation as these would provide necessary connections between populations of endemic taxa.

It was predicted (Chapter 1) that the serpentine endemic taxa would be classified as palaeoendemics. However, distribution information for the endemic taxa and their possible congeners suggests that six of the endemics are neoendemics due to their restricted distributions and low taxonomic ranking. The remaining taxa have widespread distributions and are geographically separated from their congeners and are thus, probably palaeoendemics. *Berkheya rehmannii* var. *rogersiana* is considered to be neither palaeoendemic nor neoendemic but an intermediate.

Bykov's index of endemicity predicts that areas with larger area will have a greater number of endemics. This study shows that despite their small areas, the serpentine outcrops support a higher than expected degree of endemism (Chapter 3). Results show that for the serpentine outcrops of the Barberton Greenstone Belt, levels of endemism are positively correlated with soil chemistry.

The vegetation of the serpentine outcrops is relatively homogeneous as there is a low rate of species turnover (ß – diversity) between sites on the same outcrop. This level of turnover between samples is lower on serpentine sites than on non-serpentine areas. It was found that levels of ß – diversity for the whole flora are not determined by habitat diversity or by variation in soil chemistry at each site. However, correlations between ß – diversity and elevation changes are seen within the woody component of the flora and correlations with soil chemistry are seen in within the family Poaceae. Turnover across the edaphic boundary (between serpentine samples and non-serpentine samples) was found to be relatively high. There is a less than 35% similarity between serpentine and non-serpentine floras. No statistical correlation was found between the levels of species turnover across the edaphic boundary and the species richness of the site.

The hypothesis that the turnover of taxa between sites would be greater for outcrops that are further apart geographically is supported by the results presented in Chapter 5. It was found that the outcrops in close proximity geographically have relatively high similarity with the greatest similarity between GV, SM and RT. However, none of the sites show more than 35% similarity to any of the other sites. The MM site showed the least similarity to any other site and is also the greatest distance from the other sites.

Using species richness or endemism to select priority conservation areas for the serpentine outcrop has identified five of the seven selected study areas and three additional sites as priority (Chapter 7). Both methods identify the largest site, GV, as either the top or second most important site for conservation. The sites GV, MC and DB, which have high conservation priority, are already found within protected areas. However, other priority areas such as AM, KH and MM are currently unprotected. To preserve 100% of the serpentine tolerant species, all sites would need to be selected due to the unique species composition present on each of the outcrops. All sites with endemic species are considered irreplaceable as they each have one or two endemic taxa found only on those outcrops.

The floras of only ten of the 23 large serpentine outcrop of the Barberton Greenstone Belt have been described and catalogued. Therefore, a great deal more exploration of the other outcrops is justified and necessary. It is predicted that further exploration could identify further taxa that are endemic to serpentine and other ultramafic outcrops. Scientists all over the world are asking questions such as why are there so many endemics on serpentine soils and what evolutionary processes have led to the unique flora found on serpentine outcrops. These questions cannot be answered for the Barberton Greenstone Belt without a thorough description of the flora of all the outcrops in terms of endemism and diversity.

Very little is known about the diversity of other groups of organisms on the serpentine outcrops (e.g. invertebrates, lichens) and these represent further research needed to describe these areas fully. It is also not known how the serpentine-tolerant plants of the Barberton Greenstone Belt would react to disturbance and whether they can be used to restore and revegetate disturbed serpentine sites.

This study presents a detailed analysis of the biogeography of seven selected serpentine outcrops of the Barberton Greenstone Belt in Mpumalanga, South Africa. These serpentine outcrops have been characterised in terms of species richness, species diversity, endemism and diversity at higher taxonomic levels. All of the data have indicated that the serpentine outcrops of the Barberton Greenstone Belt have a high conservation priority.

Appendix A

Checklist of the flora on selected serpentine outcrops in the Barberton Greenstone Belt in Mpumalanga, South Africa. Families are arranged alphabetically, with species arranged alphabetically within each family. Names of sites are supplied in full in Table 1. Collectors include: K. Balkwill (KB), M-J. Balkwill (MJB), M-J. Cadman (MJC), S.D. Williamson (SDW), M. Stalmans (MS), A. Paton (AP), T. Herron (TH), C. Kidger (CK), S. Smith (SS), D. Goyder (DG), C. Payet (CP)

Taxon	**CZ**	**GV**	**KK**	**MM**	**MC**	**RT**	**SM**
Ferns and Fern allies:							
ANEMIACEAE							
Mohria nudiuscula J.P.Roux (SDW 860)					X		
SINOPTERIDACEAE							
Cheilanthes hirta Sw. var. *hirta* (SDW 608 & 659)		X			X	X	
Cheilanthes involuta (Sw.) Schelpe & N.C.Anthony var. *obscura* (N.C.Anthony) N.C.Anthony (SDW 635 & 497)	X				X		
Cheilanthes sp.			X	X			
Cheilanthes viridis (Forssk.) Sw. var. *viridis* (SDW 808)					X		
Pellaea calomelanos (Sw.) Link var. *calomelanos*			X	X	X	X	
Pellaea sp.		X		X		X	X
Dicotyledons:							
ACANTHACEAE							
Asystasia subbiflora C.B. Clarke (KB & MJC 2615; KB, SDW & SS 9873)		X					X
Barleria lancifolia T.Anderson subsp. *lancifolia*				X			

Taxon	CZ	GV	KK	MM	MC	RT	SM
Barleria meyeriana Nees				X			
Barleria obtusa Nees (SDW 874)					X		
Barleria ovata E.Mey. ex Nees (KB & MJB 8175; SDW 512)	X	X		X			X
Blepharis sp				X	X		
Blepharis subvolubilis C.B.Clarke				X			
Chaetacanthus costatus Nees		X	X			X	
Chaetacanthus sp. near *C. burchellii* Nees (SDW 770 & 855)					X		
Chaetacanthus sp. (KB & MJC 2618)							X
Chaetacanthus sp. nov. (KB, MJB & CK 8117; KB, SDW & SS 10072)		X					
Chaetacanthus sp. nov. (SDW 826; MS 2900)					X	X	
Crabbea acaulis N.E.Br. (KB, MJB & CK 8182)	X	X	X	X		X	X
Crabbea galpinii C.B.Clarke (SDW 614 & 888)					X		
Crabbea hirsuta Harv. (KB, MJB, DG, AP & SDW 10868; SDW 350)	X				X		
Crossandra greenstockii S.Moore (SDW 827; KB 10186)			X		X		
Dyschoriste rogersii S.Moore				X			
Hypoestes forskaolii (Vahl) R.Br. form A (KB 9046)		X					
Isoglossa sp.	X						
Isoglossa sp. nov. 1 (SDW 674)					X		
Justicia anagalloides (Nees) T.Anderson (KB, MJB & CK 8151; KB, SDW & SS 9919 & 10139)	X	X	X		X	X	X
Justicia betonica L. (SDW 739; KB, MJB & SDW 6677a; MS 3008)					X		X

Taxon	CZ	GV	KK	MM	MC	RT	SM
Justicia cheiranthifolia (Nees) C.B.Clarke (SDW 730)							X
Justicia flava (Vahl) Vahl				X			
Justicia protracta (Nees) T.Anderson				X			
Lepidagathis scabra C.B.Clarke (SDW 859)				X	X		
Monechma sp. aff. *M. debile* (Forssk.) Nees				X			
Rhinacanthus xerophylis A.Meeuse				X			
Ruellia cordata Thunb. Taxon B form 1 (KB, SDW & SS 9688)	X						
Ruellia cordata Thunb. (SDW 767)		X	X	X	X		
Ruellia patula Jacq.		X			X	X	
Ruellia stenophylla C.B.Clarke			X				
Sclerochiton triacanthus A.Meeuse (SDW 863a)					X		
Thunbergia atriplicifolia E. Mey. ex Nees (KB, MJB & SDW 6695a; KB, SDW & SS 10160 & 9942; SDW 649 & 768)	X	X	X		X	X	X
Thunbergia galpinii Lindau (KB, MJB & SDW 6696a)						X	X
Thunbergia natalensis Hook.		X	X				
AMARANTHACEAE							
Aerva leucura Moq. (SDW 955)				X	X		
ANACARDIACEAE							
Lannea discolour (Sond.) Engl.					X		
Lannea edulis (Sond.) Engl. var. edulis (KB, SDW & SS 10144; SDW 556)	X		X				
Lannea sp.						X	
Ozoroa barbertonensis Retief (KB & SDW 10200)							X

Taxon	**CZ**	**GV**	**KK**	**MM**	**MC**	**RT**	**SM**
Ozoroa sp near *O. engleri* R.Fern. & A.Fern				X			
Ozoroa sp. probably *O. albicans* R.Fern & A.Fern (SDW 621)					X		
Ozoroa sp. (KB, MJB, DG, AP & SDW 10087)					X		
Ozoroa sp. (KB, SDW & SS 9730)	X						
Ozoroa sp. D. (SDW 508)	X						
Ozoroa sphaerocarpa R.Fern & A.Fern (KB 9059; KB, SDW & SS 9838)		X					
Sclerocarya birrea (A.Rich.) Hochst. subsp. *caffra* (Sond.) Kokwaro	X			X			
Searsia dentata (Thunb.) F.A.Barkley		X				X	
Searsia discolor (E.Mey. ex Sond.) Moffett unusual form (KB & MJB 8186)		X					
Searsia discolor (E.Mey. ex Sond.) Moffett (KB & MJB 8186)		X	X				
Searsia lancea (L.f.) F.A.Barkley (SDW 432)	X						
Searsia leptodictya (Diels) T.S.Yi, A.J.Mill. & J.Wen	X						
Searsia leptodictya (Diels) T.S.Yi, A.J.Mill. & J.Wen forma leptodictya				X			
Searsia pentheri (Zahlbr.) Moffett (SDW 430)	X				X		
Searsia pondoensis (Schönland) Moffett (SDW 838)			X		X	X	
Searsia pygmaea (Moffett) Moffett (KB & MJC 2611; SDW 569; SDW 623; SDW 863b)		X			X		X
Searsia pyroides (Burch.) Moffett var. *gracilis* (Engl.) Moffett (SDW 516)	X						
Searsia rehmanniana (Engl.) Moffett var. *rehmanniana* (KB 9055)		X				X	

Taxon	CZ	GV	KK	MM	MC	RT	SM
Searsia rogersii (Schönland) Moffett (KB, MJB & CK 8176; KB, SDW & SS 10050; KB, MJB, DG, AP & SDW 10870; SDW 409; 597; 625 & 821)		X			X	X	X
Searsia sp.						X	
Searsia transvaalensis (Engl.) Moffett						X	
ANNONACEAE							
Annona senegalensis Pers. subsp. *senegalensis* (SDW 360)	X				X		
APIACEAE							
Alepidea peduncularis A.Rich. (SDW 601 & 656; KB & MJC 2613)		X				X	X
Alepidea setifera (N.E.Br.)		X				X	X
Centella asiatica (L.) Urb. (SDW 560)			X				
Heteromorpha pubescens Burtt Davy (KB, MJB & SDW 6690a; KB, SDW & SS 10077)		X					X
Pimpinella transvaalensis H.Wolff		X				X	X
APOCYNACEAE							
Asclepias aurea (Schltr.) Schltr. (KB, MJB & CK 8146)		X					
Asclepias brevipes (Schltr.) Schltr. (SDW 1007)							X
Asclepias sp. 1				X		X	
Asclepias sp. 2					X		
Asclepias sp. aff. *A. velutina* (Schltr.) Schltr. (SDW 681)				X			
Aspidoglossum araneiferum (Schltr.) Kupicha (MS 2945; SDW 662; KB 12022)		X				X	

Taxon	CZ	GV	KK	MM	MC	RT	SM
Aspidoglossum sp. nov. near *A. biflorum* E.Mey. (SDW 655)						X	
Brachystelma macropetalum (Schltr.) N.E.Br.	X						
Brachystelma sp.		X					
Carissa bispinosa (L.) Desf. ex Brenen (KB, SDW & SS 10062)		X					
Ceropegia sp.				X			
Pachycarpus sp.						X	
Parapodium costatum E.Mey. (SDW 573)		X					
Raphionacme galpinii Schltr. (KB, SDW & SS 10066 & 9723; KB 10123, SDW 585, 1005 & 835)	X	X	X		X	X	X
Raphionacme procumbens Schltr. (SDW 503; 680a & 558)	X		X	X	X		
Raphionacme sp.			X	X		X	X
Schizoglossum sp.						X	
Sisyranthus randii S.Moore (KB, MJB & SDW 6672a)							X
Stenostelma corniculatum (E.Mey.) Bullock (SDW 637)					X		
Xysmalobium acerateoides (Schltr.) N.E.Br. (KB, SDW & SS 10068)		X					
ARALIACEAE							
Cussonia spicata Thunb.		X				X	
ASTERACEAE							
*Anthemis sp. (KB, SDW & SS 10101)		X					
Aster bakerianus Burtt Davy ex C.A.Sm.			X				X
Aster harveyanus Kuntze (KB, MJB & CK 8121; KB, SDW & SS 9883; MS 2252; SDW 830)		X			X	X	

Taxon	**CZ**	**GV**	**KK**	**MM**	**MC**	**RT**	**SM**
Aster lydenburgensis Lippert (KB, SDW & SS 9951)		X					X
Athrixia phylicoides DC. (KB 9053; SDW 428 & 487)	X	X	X		X	X	X
Athrixia sp. nov. (KB 9052; SDW 587)		X					
Athrixia subsimplex Brenan subsp. nov. (KB, SDW & SS 10107)		X					
Berkheya coddii Roessler (KB & MJB 8188; SDW 406; SDW & CP 290; KB, MJB & SDW 6680a; MS 2359)		X				X	X
Berkheya echinacea (Harv.) O.Hoffm. ex Burtt Davy							X
Berkheya insignis (Harv.) Thell. (KB, SDW & SS 9820)			X	X			
Berkheya nivea N.E.Br. (KB, MJB, DG, AP & SDW 10869; MS 3005; SDW 613 & 815)					X		
Berkheya rehmanii Thell. var. *rehmannii* (KB, MJB, DG, AP & SDW 10864; SDW 847)					X		
Berkheya rehmanii Thell. var. *rogersiana* Thell. (KB, MJB & CK 8149; KB, SDW & SS 9874 & 9944; SDW 732)		X				X	X
Berkheya seminifera Harv. & Sond.			X				
Berkheya setifera DC. (KB, SDW & SS 10114)		X					
Berkheya sp. aff. *B. insignis* (Harv.) Thell. (KB, MJB & SDW 6693a)							X
Berkheya sp. nov. (KB 11834 (bufl)			X			X	
Berkheya sp. probably *B. bipinnatifida* (Harv.) Roessler subsp. *echinopsoides* (Baker) Roessler (MS 3020)					X		
Berkheya zeyheri Oliv. & Hiern					X		
Callilepis laureola DC. (KB, SDW & SS 9731; SDW 773)	X	X			X		X

Taxon	**CZ**	**GV**	**KK**	**MM**	**MC**	**RT**	**SM**
Callilepis leptophylla Harv. (KB, SDW & SS 10108)		X					
Chrysanthemoides monilifera (L.) T. Norl. subsp. *canescens* (DC.) T.Norl. (SDW 646)						X	
Conyza podocephala DC.			X				
**Crepis hypochoeridea* (DC.) Thell. (KB, SDW & SS 10173 & 9947)		X	X				
Dicoma anomala Sond. subsp. *anomala* (SDW 578 & 596)		X	X		X		X
Dicoma anomala Sond. subsp. *gerrardii* (Harv. ex F.C.Wilson) S.Ortíz & Rodr.Oubiña (SDW 953)					X	X	
Dicoma macrocephala DC.						X	
Dicoma swazilandica S.Ortiz (KB & MJB 8178)		X					
Dimorphotheca jucunda E.Phillips (KB, MJB & CK 8120; KB, SDW & SS 10100 & 9948)		X					
Euryops laxus (Harv.) Burtt Davy (KB, SDW & SS 10103)		X	X				
Euryops sp.				X			
Felicia muricata (Thunb.) Nees subsp. *muricata* (KB, SDW & SS 10182 & 9875)		X	X				X
Gazania krebsiana Less. subsp. *serrulata* (DC.) Roessler (SDW 494; SDW & AS 759; KB, SDW & SS 9876)		X	X			X	
Gazania krebsiana Less. subsp. *krebsiana* (SDW 735)							X
Geigeria burkei Harv. subsp. *burkei* var. *burkei*			X			X	X
Geigeria burkei Harv. subsp. *burkei* var. *hirtella* Merxm. (SDW 407;615; 851 & 352)	X	X			X		

Taxon	CZ	GV	KK	MM	MC	RT	SM
Gerbera ambigua (Cass.) Sch. Bip. (KB, SDW & SS 9885 & 9739; KB, MJB & CK 8122)	X	X					
Gerbera jamesonii Bolus ex Adlam (KB, SDW & SS 9877)	X	X		X			
Gerbera piloselloides (L.) Cass. (KB, SDW & SS 10174 & 10115; SDW 491)		X	X			X	
Gerbera sp.	X		X	X		X	X
Gerbera viridifolia (DC.) Sch. Bip. (KB, SDW & SS 10165 & 10175)			X				
Gymnanthemum corymbosum (Thunb.) H.Rob. (KB & MJB 8184)		X					
Haplocarpha scaposa Harv. (KB, SDW & SS 9884; 9954; 10168 & 9737; SDW 736 & 836)	X	X	X		X		X
Helichrysum acutatum DC.			X				
Helichrysum allioides Less.			X				
Helichrysum athrixiifolium (O.Kuntze) Moeser (SDW 511)	X						
Helichrysum cerastioides DC. var. *cerastioides* (KB, SDW & SS 9825)				X			
Helichrysum harveyanum Wild (SDW 839)					X		
Helichrysum kraussii Sch. Bip. (KB, SDW & SS 10169; SDW 496)			X			X	
Helichrysum mixtum (Kuntze) Moeser var. *grandiceps* Hilliard (SDW 651)						X	
Helichrysum nudifolium (L.) Less. var. *nudifolium* (KB, SDW & SS 10171 & 10096; SDW 642 & 353)	X	X	X		X	X	
Helichrysum nudifolium (L.) Less. var *oxyphyllum* (DC.) Beentjie (SDW 819 & 858; KB, SDW & SS 9821 & 9735	X	X		X			X
Helichrysum nudifolium (L.) Less. var. *pilosellum* (L.f.) Beentjie (KB, MJB &		X					X

Taxon	CZ	GV	KK	MM	MC	RT	SM
CK 8134; KB, SDW & SS 10097; 10098 & 10105; SDW 737)							
Helichrysum pallidum DC.		X					
Helichrysum rudolfi Hilliard							X
Helichrysum sp 1			X		X		X
Helichrysum sp 2			X	X			
Helichrysum sp 3			X			X	
Helichrysum sp. probably *H. oreophilum* Klatt (SDW 599)		X					
Helichrysum sp. near *H. thapsus*	X						
Helichrysum sp. near *H. aureonitens* Sch.Bip.						X	
Helichrysum thapsus (Kuntze) Moeser (KB, MJB & CK 8181)		X					
Hilliardiella aristata (DC.) H.Rob (KB, SDW & SS 9879, 9949& 9740; SDW 762)	X	X	X			X	X
Hilliardiella hirsuta (DC.) H.Rob (KB, MJB & CK 8142; KB, SDW & SS 9953; KB, MJB & SDW 6673a)	X	X	X				X
Hilliardiella oligocephala (DC.) H.Rob (KB, SDW & SS 9952; SDW 794)		X			X		
Inezia integrifolia (Klatt) E.Phillips (KB, SDW & SS 10106)		X				X	X
Inezia sp. nov. (KB 9048)		X					
Inulanthera calva (Hutch.) Källersjö Barberton form (KB & MJB 8170)		X					
Inulanthera calva (Hutch.) Källersjö (KB 9049)		X					
Lactuca inermis Forssk. (KB, SDW & SS 10180)			X				
Lactuca sp.		X					X
Lactuca sp. (KB, SDW & SS 10176)			X				

Taxon	**CZ**	**GV**	**KK**	**MM**	**MC**	**RT**	**SM**
Launaea rarifolia (Oliv. & Hiern) Boulos var. *rarifolia* (KB, SDW & SS 10178 & 10113)		X	X				
Macledium zeyheri (Sond.) S. Ortiz subsp. *thyrsiflorum* (Klatt) Netnou (KB, SDW & SS 9819)				X			
Macledium zeyheri (Sond.) S.Ortíz subsp. *zeyheri* (SDW 408 & 577)	X	X	X	X			X
Nidorella auriculata DC. (KB, SDW & SS 9881 & 9740c; SDW 420 & 355)	X	X			X		
Nidorella resedifolia DC. subsp. *resedifolia* (SDW 795)					X		
Nidorella sp			X				
Pegolettia lanceolata Harv. (KB, MJB & CK 8129; KB, SDW & SS 10099)		X					
Pseudognaphalium luteo-album (L.) Hilliard & B.L. Burtt (KB, SDW & SS 9956)		X					
Pseudognaphalium oligandrum (DC.) Hilliard & B.L. Burtt (SDW 647)						X	
Schistostephium crataegifolium (DC.) Fenzl ex Harv. (SDW 600 & 863)		X	X		X		X
Schistostephium heptalobum (DC.) Oliv. & Hiern (SDW 609)		X	X			X	
Senecio albanensis DC. possibly *S. albanensis* DC. var. *doroniciflorus* (DC.) Harv. (KB 9045)		X					
Senecio anomalochrous Hilliard (KB, MJB & SDW 6675a; KB, SDW & SS 9888)		X					X
Senecio bupleuroides DC. (KB, MJB & CK 8118)		X					
Senecio coronatus (Thunb.) Harv. (KB, SDW & SS 10172; 10181; 9880 & 9738; SDW 837a; SDW & TH 885)	X	X	X		X		X
Senecio erubescens Aiton var. *dichotomus* DC. (KB, SDW & SS 9945)		X					

Taxon	CZ	GV	KK	MM	MC	RT	SM
Senecio erubescens Aiton var. *erubescens* (KB, MJB & CK 8156)		X					
Senecio glaberrimus DC. (KB, MJB & CK 8119; KB, SDW & SS 10109; SDW 629)		X					
Senecio glanduloso-pilosus Volkens & Muschl. (KB, SDW & SS 10167, 10170 & 10104)		X	X				
Senecio latifolius DC.						X	
Senecio lydenburgensis Hutch. & Burtt Davy (KB & MJB 8169; KB, SDW & SS 9943; SDW 652)		X				X	
Senecio madagascariensis Poir. (KB, SDW & SS 9822)				X			
Senecio microglossus DC. (KB, SDW & SS 9732 & 10112, SDW 351 & 653)	X	X	X			X	
Senecio oxyriifolius DC. subsp. *oxyrrifolius* (KB, MJB & CK 8123; KB, SDW & SS 9890)		X					X
Senecio panduriformis Hilliard (KB, MJB & CK 8167)		X					
Senecio pterophorus DC. (KB, SDW & SS 10102)		X					
Senecio scitus Hutch. & Burtt Davy (KB, SDW & SS 9887)		X					
Senecio sp. 1			X				
Senecio sp. 2				X			
Senecio sp. aff. *S. coronatus* (Thunb.) Harv. (KB, SDW & SS 9950)		X					
Senecio sp. near *S. harveianus* MacOwan (KB, SDW & SS 9734)	X						
Senecio sp. near *S. latifolius* DC.			X				X
Senecio sp. near *S. venosus* Harv.	X		X				X
Senecio venosus Harv. (KB, SDW & SS 9818 & 9733; SDW 804)	X	X	X	X	X	X	

Taxon	**CZ**	**GV**	**KK**	**MM**	**MC**	**RT**	**SM**
Sonchus dregeanus DC. (KB, SDW & SS 10110 & 9889)		X					
Sonchus integrifolius Harv. var. *integrifolius* (KB, SDW & SS 9946)		X					
* *Sonchus oleraceous* L.						X	
Tarchonanthus camphoratus L.				X			
Vernonia galpinii Klatt (KB, SDW & SS 10179)			X				
Vernonia sp. near *V. fastigata* Oliv. & Hiern (KB, SDW & SS 9882)		X					
Vernonia sutherlandii Harv. (KB, MJB & CK 8128; KB, SDW & SS 9736, 9891 & 9823; SDW 798)	X	X	X	X	X		X
BIGNONIACEAE							
Tecoma capensis (Thunb.) Lindl. (A. van Oudtshoorn s.n.)						X	
BORAGINACEAE							
Cynoglossum hispidum Thunb. (KB, MJB & CK 8144)		X					
BURSERACEAE							
Commiphora neglecta I.Verd.				X			
Commiphora schimperi (O.Berg) Engl.	X						
Commiphora sp.	X						
CAMPANULACEAE							
Wahlenbergia undulata (L.f.) A. DC. (SDW 598 & 644)		X				X	
Wahlenbergia virgata Engl. (KB, SDW & SS 10029 ; KB, MJB & CK 8137)		X					

Taxon	CZ	GV	KK	MM	MC	RT	SM
CAPPARACEAE							
Boscia albitrunca (Burch.) Gilg & Gilg-Ben.				X			
Capparis sp.				X			
Cleome angustifolia Forssk. subsp. *petersiana* (Klotzsch ex Sond.) Kers				X			
Maerua parvifolia Pax (KB, SDW & SS 9797)				X			
Maerua sp.				X			
CARYOPHYLLACEAE							
Pollichia campestris Aiton (KB & MJB 8179)		X					
CELASTRACEAE							
Catha edulis (Vahl) Forssk. ex Endl. (KB, SDW & SS 10043)		X					
Gymnosporia buxifolia (L.) Szyszyl. (MS 2802)						X	
Gymnosporia glaucophylla Jordaan				X			
Gymnosporia heterophylla (Eckl. & Zeyh.) N.K.B. Robson (KB & MJC 2614)	X	X		X	X	X	X
Gymnosporia sp. nov.					X		
Gymnosporia senegalensis (Lam.) Loes (SDW 436)	X						
Maytenus undata (Thunb.) Blakelock (MS 2263)	X			X		X	
Pterocelastrus rostratus Walp. (KB, SDW & SS 10046)		X					
CELTIDACEAE							
Celtis africana Burm.f.	X						

Taxon	CZ	GV	KK	MM	MC	RT	SM
CHRYSOBALANACEAE							
Parinari capensis Harv. subsp. *capensis* (KB, SDW & SS 10131)			X				
COMBRETACEAE							
Combretum apiculatum Sond. subsp. *apiculatum* (KB, SDW & SS 9680)	X			X	X		
Combretum hereroense Schinz subsp. *hereroense* (SDW & TH 891)				X	X		X
Combretum molle R.Br.ex G.Don.	X	X			X		
Combretum zeyheri Sond. (SDW & TH 892)	X			X	X		
CONVOLVULACEAE							
Convolvulus farinosus L. (KB, SDW & SS 9704 & 9793)	X			X			
Convolvulus natalensis Bernh. ex Krauss (SDW 734)							X
Convolvulus sagittatus Thunb. (SDW 766)					X	X	
Convolvulus sp.				X			
Convolvulus sp. near *C. natalensis* Bernh. ex Krauss	X						
Evolvulus alsinoides (L.) L.				X		X	
Ipomoea crassipes Hook.			X				
Ipomoea dichroa Choisy				X			
Ipomoea oblongata E.Mey. ex Choisy (SDW 634 & 772)	X	X			X		X
Ipomoea obscura (L.) Ker Gawl. var. *obscura* (SDW 771)	X			X	X		
Ipomoea ommanneyi Rendle			X				
Ipomoea papilio Hallier f. (KB, SDW & SS 9682 & SDW 504)	X						
Ipomoea sp. 1		X					
Ipomoea sp. 2		X					

Taxon	CZ	GV	KK	MM	MC	RT	SM
Merremia tridentata (L.) Hallier f. subsp. *angustifolia* (Jacq.) Ooststr.						X	
Seddera suffruticosa (Schinz) Hallier f. (KB, SDW & SS 9809)				X			
Xenostegia tridentata (L.) D.F.Austin & Staples. subsp. *angustifolia* (Jacq.) Lejoly & Lisowski (SDW 616)				X	X		
CRASSULACEAE							
Crassula sp.							X
Crassula sp. nov. near *C. inandensis* Schönland & Baker f. or C. sarmentosa Harv. (KB, MJB, DG, AP & SDW 10875; SDW 829)					X		
Crassula vaginata Eckl. & Zeyh. subsp. *vaginata* (KB 11838; KB, SDW & SS 10061; SDW 630)		X					
Kalanchoe rotundifolia (Haw.) Haw. (KB 9060; KB, MJB & CK 8158)		X					
Kalanchoe sp.				X			
CUCURBITACEAE							
Kedrostis sp.	X						
DIPSACACEAE							
Cephalaria pungens Szabó or *C. zeyheriana* Szabó			X				
Scabiosa columbaria L. (KB, MJB & CK 8145; KB, SDW & SS 10056 & SS 9835; KB 10127; SDW 844 & 498; SDW & AS 760)		X	X		X	X	
EBENACEAE							
Diospyros galpini (Hiern) De Winter							X

Taxon	CZ	GV	KK	MM	MC	RT	SM
Diospyros lycioides Desf. subsp. *sericea* (Bernh.) De Winter	X		X		X	X	
Diospyros mespiliformis Hochst. ex A.DC.				X			
Diospyros whyteana (Hiern) F.White (KB, SDW & SS 10041; SDW 490)		X				X	X
Euclea crispa (Thunb.) Gürke subsp. *crispa* (SDW 488)	X	X			X	X	
Euclea daphnoides Hiern				X			
Euclea natalensis A.DC. subsp. *natalensis*		X	X		X		
ERICACEAE							
Erica drakensbergensis Guthrie & Bolus (KB, SDW & SS 10135)			X				
ERYTHROXYLACEAE							
Erythroxylum delagoense Schinz (KB, SDW & SS 10048)		X		X		X	
EUPHORBIACEAE							
Acalypha angustata Sond. (KB, MJB & CK 8140 & 8141)		X					X
Acalypha caperonioides Baill. (KB, SDW & SS 10117; SDW 799)	X		X		X		
Acalypha glandulifolia Buchinger ex Meisn. (KB, SDW & SS 10059, 10060 & 9831)		X					X
Acalypha peduncularis E.Mey. ex Meisn. (KB, MJB & CK 8152; KB, SDW & SS 9927)		X					
Acalypha punctata Meisn. var. *punctata* (KB, SDW & SS 9928)		X			X		
Acalypha sonderiana Müll.Arg.	X						
Acalypha sp.				X			

Taxon	**CZ**	**GV**	**KK**	**MM**	**MC**	**RT**	**SM**
Acalypha sp. near *A. wilmsii*			X				
Acalypha villicaulis Hochst. (SDW 510 & 809)	X				X	X	
Acalypha wilmsii Pax ex Prain & Hutch.		X					
Clutia affinis Sond. (KB, MJB & CK 8183)		X					
Clutia monticola S.Moore var. *monticola* (KB, MJB & CK 8154; KB, SDW & SS 10039 & 9854)		X					
Clutia pulchella L. var. *obtusata* Sond. (SDW 639 & 797)					X		
Clutia sp.	X					X	X
Clutia virgata Pax & K. Hoffm. (KB, SDW & SS 10051; SDW 658)		X				X	
Dalechampia capensis A.Spreng.				X			
Dalechampia galpinii Pax (SDW 800)					X	X	
Euphorbia cooperi N.E.Br. ex A.Berger				X			
Euphorbia gueinzii Boiss. var. *gueinzii* (KB, SDW & SS 10155 & 9713)	X		X				
Euphorbia neopolycnemoides Pax. & K.Hoffm.				X			
Euphorbia pseudotuberosa L. (KB SDW & SS 10150)		X	X				
Euphorbia schinzii Pax (KB, SDW & SS 9826)		X		X			
Euphorbia sp.				X	X		
Euphorbia sp. near *E. pseudotuberosa* Pax							X
Euphorbia striata Thunb. var. *striata* (KB, SDW & SS 9834 & 9921)		X					
Jatropha latifolia Pax var. *angustata* Prain (MS 2253)						X	
Jatropha latifolia Pax var. *latifolia* (SDW 841; KB SDW & SS 10122)			X		X		

Taxon	CZ	GV	KK	MM	MC	RT	SM
Jatropha latifolia Pax var. *swazica* Prain (KB, SDW & SS 9862)		X					
Jatropha sp.				X	X		
Tragia wahlenbergiana Prain (MS 2363 & 2364)						X	
FABACEAE							
Acacia ataxacantha DC.	X						
Acacia caffra (Thunb.) Willd				X	X		
Acacia davyii N.E.Br. (SDW 837)	X				X		
Acacia forbesii (Mim.) Benth.						X	
Acacia karroo Hayne		X		X		X	
Acacia nigrescens Oliv.				X			
Acacia nilotica (L.) Willd. subsp. *kraussiana* (Benth.) Brenan (SDW 854)					X		
Acacia sp.							X
Acacia swazica Burtt Davy				X			
Alysicarpus zeyheri Harv.			X				
Argyrolobium robustum T.J.Edwards (KB, MJB & CK 8139; KB, SDW & SS 9851; SDW 1003 & 654)		X	X			X	X
Argyrolobium rupestre (E.Mey.) Walp. (KB, SDW & SS 10163a & 10025; KB & SDW 10196)		X					X
Argyrolobium sp. 1	X						
Argyrolobium sp. 2			X				
Argyrolobium tomentosum (Andrews) Druce						X	
Argyrolobium wilmsii Harms (KB, SDW & SS 10052; MS 2407)		X				X	
Bauhinia galpinii N.E.Br.	X			X	X		
Bolusanthus speciosus (Bolus) Harms				X			

Taxon	**CZ**	**GV**	**KK**	**MM**	**MC**	**RT**	**SM**
Chamaecrista comosa E.Mey. var. *capricornia* (Steyaert) Lock		X	X	X		X	
Crotalaria recta Steud. ex A.Rich. (KB & MJB 8180)		X					
Dalbergia armata E.Mey. (KB, SDW & SS 10057)		X					
Dichrostachys cinerea (L.) Wight & Arn. subsp. *africana* Brenan & Brummitt	X			X	X		
Elephantorrhiza elephantina (Burch.) Skeels							X
Elephantorrhiza sp.	X				X		
Eriosema cordatum E.Mey. (KB, SDW & SS 10142 & 9913)	X	X	X			X	
Eriosema distinctum N.E.Br. (KB, MJB & CK 8148)		X					
Eriosema ellipticifolium Schinz		X					X
Eriosema psoralioides (Lam.) G.Don (KB & MJC 2619)							X
Eriosema sp.			X			X	
Flemingia grahamiana Wight & Arn. (SDW 425)	X						
Indigofera arrecta Hochst. ex A.Rich.					X		
Indigofera crebra N.E.Br. (KB, SDW & SS 9894)		X					
Indigofera hedyantha Eckl. & Zeyh. (KB, SDW & SS 10030)		X					
Indigofera hilaris Eckl. & Zeyh. var. *hilaris* (KB, SDW & SS 10031 & 10136; SDW 756)		X	X		X	X	X
Indigofera oxalidea Welw. ex Baker (KB, SDW & SS 10033)		X					
Indigofera rostrata Bolus (SDW 1002)							X
Indigofera sp. 1	X			X	X		
Indigofera sp. 2			X		X		

Taxon	**CZ**	**GV**	**KK**	**MM**	**MC**	**RT**	**SM**
Indigofera sp. near *I. sanguinea* N.E.Br.						X	
Indigofera spicata Forssk.			X				
Indigofera vicioides Jaub. & Spach var. *rogersii* (R.E.Fr.) J.B.Gillett				X			
Leobordea carinata (E.Mey.) B.-E.van Wyk & Boatwr. (KB, SDW & SS 9799; KB, MJB & SDW 6686a; SDW 672, 507 & 419)	X	X		X	X		X
Leobordea divaricata Eckl. & Zeyh		X	X			X	
Leobordea eriantha (Benth.) B.-E.van Wyk. & Boatw. (KB, MJB & CK 8171; KB, SDW & SS 9896; SDW 588 & 756a)		X	X			X	
Lotononis laxa Eckl. & Zeyh (SDW 834)					X		
Ormocarpum trichocarpum (Taub.) Engl.				X			
Otholobium polystictum (Benth. ex Harv.) Stirton (KB & MJB 8172)		X					
Pearsonia aristata (Schinz) Dummer							X
Pearsonia cajanifolia (Harv.) Polhill subsp. *cryptantha* (Baker) Polhill (KB & MJB 8174; KB, MJB & CK 8174)		X					
Pearsonia sessilifolia (Harv.) Dummer subsp. *marginata* (Schinz) Polhill (KB, SDW & SS 10071; SDW 793; 358 & 754)	X	X	X		X	X	
Pearsonia sessilifolia (Harv.) Dummer subsp. *sessilifolia* (KB, SDW & SS 10038; KB, MJB & SDW 6682a)		X					X
Pearsonia sp.	X					X	
Peltophorum africanum Sond. (SDW 848)	X			X	X		
Philenoptera violacea (Klotzsch) Schrire				X			
Pseudarthria hookeri Wight & Arn.			X			X	
Rhynchosia clivorum S.Moore						X	
Rhynchosia galpinii Baker f.					X		

Taxon	CZ	GV	KK	MM	MC	RT	SM
Rhynchosia minima (L.) DC. var. *minima* (KB, SDW & SS 9790; SDW 849)				X	X	X	
Rhynchosia monophylla Schltr. (KB, SDW & SS 10130; SDW 552)			X				
Rhynchosia nervosa Benth. & Harv. var. *nervosa* (KB, SDW & SS 10141)			X				
Rhynchosia nitens Benth. ex Harv.					X		X
Rhynchosia reptabunda N.E.Br. (KB, SDW & SS 9911)		X					
Rhynchosia sordida (E.Mey.) Schinz (KB, SDW & SS 10065)		X					
Rhynchosia sp				X		X	
Rhynchosia sp. c.f. *R. minima* (L.) DC. (SDW 670)					X		
Rhynchosia totta (Thunb.) DC. var. *totta* (KB, SDW & SS 10159; SDW 636)		X	X	X	X	X	X
Rhynchosia venulosa (Hiern) K.Schum. (KB, SDW & SS 9696)	X						
Schotia brachypetala Sond.				X			
Senegalia loetteri (SDW 954)					X		
Senna italica Mill. subsp. *arachoides* (Burch.) Lock (SDW 805)				X	X		
Senna petersiana (Bolle) Lock	X						
Stylosanthes fruticosa (Retz.) Alston (SDW 776; 834 & 348)	X		X		X		
Sutherlandia sp.							X
Tephrosia elongata E.Mey. var. *elongata* (SDW 411)		X	X	X	X	X	
Tephrosia longipes Meisn. subsp. *longipes*					X	X	
Tephrosia macropoda (E.Mey.) Harv. var. *macropoda* (SDW 565)	X	X		X		X	X
Tephrosia multijuga R.G.N.Young (SDW 831 & 857)				X	X		

Taxon	CZ	GV	KK	MM	MC	RT	SM
Tephrosia polystichya E.Mey. var. *hirta* Harv.			X				
Tephrosia retusa Burtt Davy (SDW 354)	X						
Tephrosia semiglabra Sond. (SDW 669 & 661)	X	X		X	X	X	
Tephrosia sp	X		X	X			
Vigna unguiculata (L.) Walp. subsp. *stenophylla* (Harv.) Maréchal, Mascherpa & Stainier						X	
Vigna vexillata (L.) A.Rich. var. *angustifolia* (Schum. & Thonn.) Baker (SDW 861)					X		
Vigna vexillata (L.) A.Rich var. *vexillata*	X						
Zornia capensis Pers. subsp. *capensis* (KB, SDW & SS 10032)	X	X			X	X	
Zornia linearis E.Mey.			X				
GENTIANACEAE							
Enicostema axillare (Lam.) A.Raynal subsp. *axillare* (SDW 777)					X		
GERANIACEAE							
Monsonia attenuata Harv. (SDW 632)		X					
Pelargonium luridum (Andr.) Sweet (KB, SDW & SS 9893)		X	X				
GREYIACEAE							
Greyia radlkoferi Szyszyl. (KB, SDW & SS 10045)		X					
HETEROPYXIDACEAE							
Heteropyxis natalensis Harv.	X						

Taxon	CZ	GV	KK	MM	MC	RT	SM
HYPERICACEAE							
Hypericum aethiopicum Thunb. subsp. *sonderi* (Bredell) N. Robson (KB, SDW & SS 10162 & 9926; KB, MJB & CK 8130)		X	X			X	X
LAMIACEAE							
**Salvia tiliifolia* Vahl (KB 9082)						X	
Acrotome hispida Benth.			X				
Acrotome thorncroftii Skan (KB, MJB, DG, AP & SDW 10867; SDW 774)					X		
Clerodendrum suffruticosum Gürke var. *suffruticosum*					X		
Leonotis sp.							X
Leucas glabrata (Vahl) Sm. var. *linearis* Codd (KB, SDW & SS 9796)				X			
Ocimum obovatum E.Mey. ex Benth. subsp. *obovatum* var. *obovatum* (KB, SDW & SS 10157)		X	X				
Ocimum serratum (Schlechter) A.J.Paton (KB, MJB, DG, AP & SDW 10861; SDW 818)	X				X		
Ocimum sp. near *O. obovatum* E.Mey. ex Benth. (KB & MJC 2616; KB, MJB & SDW 6670a & 6674a)			X				X
Ocimum sp. nov. (SDW 751)						X	
Ocimum tubiforme (R.Good) A.J.Paton (KB, MJB, DG, AP & SDW 10862 & 10863)					X		
Rabdosiella calycina (Benth.) Codd (KB & MJB 8168)		X					
Rotheca hirsuta s.l. (Hochst.) R.Fern. (SDW & AS 752)	X					X	X

Taxon	CZ	GV	KK	MM	MC	RT	SM
Rotheca louwalbertsii (P.P.J.Herman) P.P.J.Herman & Retief (KB, MJB & CK 8125)		X			X	X	
Rotheca myricoides (Hochst.) Steane & Mabb.	X						
Stachys natalensis Hochst. var. *natalensis* (KB, MJB & CK 8132; KB, SDW & SS 10064 & 9839; SDW 359 & 833)	X	X	X		X	X	X
Stachys nigricans Benth. (KB, MJB & CK 8135; KB, SDW & SS 10058 & 9917)		X				X	
Syncolostemon modestus (Codd) D.F.Otieno (KB, MJB & CK 8124)		X					
Syncolostemon persimilis (N.E.Br.) D.F.Otieno (SDW 775)					X		X
Syncolostemon punctatus (Codd) D.F.Otieno (KB & MJC 2617; KB, MJB & SDW 6684a)							X
Syncolostemon sp.			X			X	
Syncolostemon sp. 2			X				
Syncolostemon transvaalensis (Schltr.) D.F.Otieno (KB 12019; KB, SDW & SS 9929; MS 3006; SDW 728)	X	X			X		X
Thorncroftia sp.					X		
Tinnea galpinii Briq. (KB, MJB & CK 8153; SDW 416; 590 & 550)		X	X				
Vitex harveyana H.Pearson (KB, MJB & SDW 6691a)							X
Vitex obovata E.Mey. subsp. *wilmsii* (Guerke) C.L.Bredenkamp & D.J.Botha (SDW & TH 893)					X		
Vitex obovata E.Mey. subsp. *obovata* (MS 3004; SDW 846 & 502)	X				X		

Taxon	CZ	GV	KK	MM	MC	RT	SM
LINACEAE							
Linum thunbergii Eckl. & Zeyh. (SDW 713)			X			X	X
LOBELIACEAE							
Cyphia elata Harv.			X				
Cyphia stenophylla E.Wimm. (SDW 693)							X
Monopsis decipiens (Sond.) Thulin (KB, SDW & SS 9907)		X					
LOPHIOCARPACEAE							
Corbichonia decumbens (Forssk.) Exell (SDW 801)				X	X		
LORANTHACEAE							
Agelanthus natalitius (Meisn.) Polh. & Weins subsp. *zeyheri* (Harv.) Polh. & Weins (KB, SDW & SS 10074; SDW 852)		X			X		
Erianthemum dregei (Eckl. & Zeyh.) Tiegh. (SDW 424 & 429)	X						
LYTHRACEAE							
Nesaea schinzii Koehne (KB, MJB & SDW 6671a; SDW 576)		X					X
MALPHIGIACEAE							
Sphedamnocarpus pruriens (A.Juss) Szylszyl. subsp. p*ruriens*	X			X	X		

Taxon	CZ	GV	KK	MM	MC	RT	SM
MALVACEAE							
Abutilon sonneratianum (Cav.) Sweet (KB, MJB & CK 8166)		X					
Corchorus asplenifolius Burch.	X	X			X		
Corchorus confusus Wild (KB, SDW & SS 9685; SDW 814)	X			X	X		X
Corchorus sp.				X		X	
Dombeya rotundifolia (Hochst.) Planch. var. *rotundifolia*	X	X		X	X	X	X
Grewia bicolor Juss. var. *bicolor* (KB, SDW & SS 9798)				X			
Grewia monticola Sond.				X			
Grewia sp.	X						
Hermannia antonii I.Verd. (KB & MJC 2612)			X				X
Hermannia boraginiflora Hook. (SDW 677 & 663)				X	X	X	
Hermannia depressa N.E.Br. (KB, SDW & SS 10090; SDW & AS 747)		X	X				
Hermannia glanduligera K.Schum.				X			
Hermannia sp.							X
Hermannia sp. near *H. montana* N.E.Br.	X					X	
Hibiscus aethiopicus L. var. *ovatus* Harv. (KB, SDW & SS 9908; SDW 506)	X	X	X			X	
Hibiscus barbosae Exell					X		
Hibiscus engleri K.Schum.				X			
Hibiscus pusillus Thunb. (SDW 769)				X	X		
Hibiscus trionum L. (SDW 660)						X	
Melhania prostata DC.				X			
Pavonia sp.	X						
Sida acuta Burm.f. subsp. *acuta*					X		

Taxon	CZ	GV	KK	MM	MC	RT	SM
Sida chrysantha Ulbr. (KB, SDW & SS 9806; SDW 555)			X	X			
Sida dregei Burtt Davy				X			
Triumfetta pilosa Roth var. *tomentosa* Szyszyl.ex Sprague & Hutch.							X
Triumfetta welwitschii Mast. var. *welwitschii* (KB, SDW & SS 10138; 9866 & 9691)	X	X	X				X
Waltheria indica L. (KB, SDW & SS 9710 & 9794)	X		X	X			
MOLLUGINACEAE							
Limeum sp.							
Psammotropha myriantha Sond. (KB & MJC 2610; KB, MJB & SDW 6689a)							X
Psammotropha sp.			X				
MORACEAE							
Ficus abutilifolia (Miq.) Miq.				X			
MYRICACEAE							
Morella serrata (Lam.) Killick (KB & MJB 8187)		X					
MYRSINACEAE							
Myrsine africana L. (KB, SDW & SS 10047)		X					
OLACACEAE							
Olax dissitiflora Oliv.				X			
Ximenia caffra Sond. var. *caffra* (SDW 850)				X	X		

Taxon	CZ	GV	KK	MM	MC	RT	SM
Ximenia caffra Sond. var. *natalensis* Sond. (MS 2406)						X	
OLEACEAE							
Menodora africana Hook. (KB, SDW & SS 9868)		X					
Schrebera alata (Hochst.) Welw. (KB, MJB & SDW 6692a)							X
ONAGRACEAE							
*_Oenothera rosea_ L'Hér. ex Aiton (MS 3036)						X	
OROBANCHACEAE							
Alectra sp.			X				
Buchnera dura Benth. (SDW 627)		X					
Graderia scabra (L.f.) Benth. (SDW 824)	X	X			X		
Graderia sp. nov. (SDW 761)						X	
Graderia subintegra Mast. (KB, MJB & CK 8116)		X					
Striga asiatica (L.) Kuntze (SDW 856 & 586)		X			X		X
Striga bilabiata (Thunb.) Kuntze subsp. *bilabiata* (SDW 675; 825; 562 & 568)		X	X		X		X
Striga elegans Benth. (SDW 418 & 567)		X	X				
OXALIDACEAE							
Oxalis obliquifolia Steud. ex A.Rich		X	X	X		X	
Oxalis smithiana Eckl. & Zeyh. (KB, SDW & SS 10158 & 10063)	X	X	X		X		

Taxon	CZ	GV	KK	MM	MC	RT	SM
PASSIFLORACEAE							
Adenia digitata (Harv.) Engl.				X			
Basananthe triloba (Bolus) W.J.de Wilde				X			
Basananthe sandersonii (Harv.) W.J.de Wilde (KB, MJB & CK 8157; KB, SDW & SS 9840; SDW 603; KB 10151)		X	X				X
PHYLLANTHACEAE							
Phyllanthus burchellii (E.Mey. ex Drège) Radcl.-Sm (SDW 571)		X					
Phyllanthus glaucophyllus Sond. (KB, SDW & SS 9817)				X			
Phyllanthus maderaspatensis L. (SDW 806; 813 & 572; KB, SDW & SS 9722 & SDW 499)	X	X			X	X	
Phyllanthus parvulus Sond.var *garipensis* (E.Mey. ex Drége) Radcl.-Sm				X			
Phyllanthus sp 1	X		X	X		X	X
Phyllanthus sp 2			X		X		
PITTOSPORACEAE							
Pittosporum viridiflorum Sims (KB, SDW & SS 10044)		X					
POLYGALACEAE							
Polygala albida Schinz subsp. *albida*				X			
Polygala gracilenta Burtt Davy (KB, SDW & SS 9905)		X					
Polygala hottentotta Presl (KB, MJB, DG, AP & SDW 10872; KB, SDW & SS 10081; SDW 415; 574; 643, 810 & 680b)		X	X	X	X	X	X

Taxon	CZ	GV	KK	MM	MC	RT	SM
Polygala leendertziae Burtt Davy (KB 9044; KB, SDW & SS 9922)		X					
Polygala ohlendorfiana Eckl. & Zeyh. (KB, SDW & SS 9920)		X					
Polygala producta N.E.Br. (KB, SDW & SS 9791)				X			
Polygala serpentaria Eckl. & Zeyh.						X	
Polygala sp.	X						
Polygala sphenoptera Fresen. var. *sphenoptera* (SDW 802)	X				X		
Polygala wilmsii Chod. (SDW 489 & 501)						X	
POLYGONACEAE							
Oxygonum dregeanum Meisn. subsp. *swazicum* (Burtt Davy) Germish (KB, SDW & SS 10184)			X				
Rumex sagittatus Thunb. (KB 1465)							X
PROTEACEAE							
Faurea rochetiana (A.Rich.) Chiov. ex Pic.Serm. (SDW 427; KB 1464)	X	X	X			X	X
Faurea saligna Harv. (SDW 845)	X				X		
Protea caffra Meisn. subsp. *caffra* (KB, SDW & SS 10054)		X				X	
Protea curvata N.E.Br.					X		
Protea gaguedi Gmel. (KB 11839)		X					X
RANUNCULACEAE							
Clematis brachiata Thunb.	X						
Ranunculus multifidus Forssk. (KB, SDW & SS 9918)		X					

Taxon	CZ	GV	KK	MM	MC	RT	SM
RHAMNACEAE							
Berchemia zeyheri (Sond.) Grubov	X			X	X		
Helinus sp.						X	
Rhamnus prinoides L'Hér. (KB 9058)		X					
Ziziphus mucronata Willd. subsp. *mucronata* (SDW 778)	X			X	X		
ROSACEAE							
Cliffortia linearifolia Eckl. & Zeyh. (KB, MJB & CK 8177)		X					
RUBIACEAE							
Anthospermum herbaceum L.f. (KB, SDW & SS 9909)		X				X	X
Anthospermum rigidum Eckl. & Zeyh. subsp. *pumilum* (Sond.) Puff (SDW 410; 575 & 648)	X	X	X			X	
Cephalanthus natalensis Oliv. (KB, SDW & SS 10156 & 10049)		X	X			X	
Coddia rudis (E.Mey. ex Harv.) Verdc. (KB, SDW & SS 9677)	X			X			
Fadogia homblei De Wild.			X				
Galium thunbergianum Eckl. & Zeyh.	X						
Kohautia amatymbica Eckl. & Zeyh. (KB, MJB & CK 8133; KB, SDW & SS 9895; SDW & AS 755; SDW 822)	X	X	X		X	X	X
Oldenlandia herbacea (L.) Roxb. var *herbacea*			X				
Oldenlandia sp.						X	
Pachystigma latifolium Sond. (KB 9083)						X	
Pavetta catophylla K.Schum	X						
Pavetta cooperi Harv. & Sond. (KB, SDW & SS 10042)		X					

Taxon	CZ	GV	KK	MM	MC	RT	SM
Pavetta edentula Sond. (SDW 518 & 857a)	X				X	X	
Pavetta schumanniana F.Hoffm. ex K.Schum.	X						
Pavetta sp.			X	X			
Pentanisia angustifolia (Hochst.) Hochst. (KB, MJB & CK 8126; KB, SDW & SS 9827; KB 10147; SDW & AS 750; SDW 811)	X	X	X		X	X	X
Pentanisia prunelloides (Klotzsch ex Eckl. & Zeyh.) Walp. subsp. *prunelloides* (KB, SDW & SS 9675 & 10067)	X	X	X			X	
Pygmaeothamnus chamaedendrum (Kuntze) Robyns var. *chamaedendrum*			X				
Tricalysia lanceolata (Sond.) Burtt Davy (KB 9050)		X					
Vangueria infausta Burch. subsp. *infausta*	X				X		
RUTACEAE							
Zanthoxylum capense (Thunb.) Harv.				X			
SALICACEAE							
Salix mucronata Thunb. subsp. *mucronata* (KB, MJB & CK 8173)		X					
SANTALACEAE							
Osyridicarpos schimperianus (Hochst. ex A.Rich) A.DC. (KB 1458)							X
Thesium deceptum N.E.Br. (KB, SDW & SS 9899)		X					
Thesium gracilarioides A.W.Hill (SDW 667)					X		
Thesium gracile A.W.Hill (SDW 796 & 812)					X		

Taxon	CZ	GV	KK	MM	MC	RT	SM
Thesium multiramulosum Pilger (KB, SDW & SS 9705)	X						
Thesium sp. 1			X			X	
Thesium sp. 2				X			X
Thesium sp. near *T. gypsophiloides* A.W.Hill (KB, MJB, DG, AP & SDW 10873; SDW 619 & 513)	X				X		
SAPINDACEAE							
Allophylus chaunostachys Gilg	X						
Pappea capensis Eckl. & Zeyh.				X			
SAPOTACEAE							
Mimusops zeyheri Sond.				X			
SCROPHULARIACEAE							
Chaenostoma floribundum Benth. (KB, MJB & CK 8165)		X					
Jamesbrittenia accrescens (Hiern) Hilliard (KB, SDW & SS 9708 & SDW 349)	X						
Jamesbrittenia aurantiaca (Burch.) Hilliard (KB, SDW & SS 9699a)	X						
Jamesbrittenia burkeana (Benth.) Hilliard (KB 9051; SDW 414 & 650)		X				X	
Jamesbrittenia grandiflora (Galpin) Hilliard (KB, SDW & SS 10073; SDW 421; 551 & 862)		X	X		X	X	X
Jamesbrittenia huillana (Diels) Hilliard (SDW 620)					X		
Manulea parviflora Benth. var. *parviflora* (KB 9084)						X	
Nemesia albiflora N.E.Br. (KB, MJB & SDW 6685a)							X

Taxon	CZ	GV	KK	MM	MC	RT	SM
Nemesia fruticans (Thunb.) Benth. (KB, SDW & SS 9900)		X					
Selago villosa Rolfe (SDW 628)		X					
Zaluzianskya elongata Hilliard & B.L.Burtt (SDW 607)		X					
SOLANACEAE							
Solanum supinum Dunal var. *leucophaeum* (Dunal) Bitter				X			
THYMELAEACEAE							
Gnidia caffra (Meisn.) Gilg (KB, MJB & CK 8143; KB, SDW & SS 9836 & SS 9916; SDW 618 & 817)	X	X			X		X
Gnidia gymnostachya (C.A.Mey.) Gilg (KB, SDW & SS 9914)		X					
Gnidia kraussiana Meisn. var. *kraussiana* (KB 10128)			X				
Gnidia sp.			X				
Lasiosiphon capitatus (L.f.) Burtt Davy (SDW 356)	X				X		
TURNERACAEAE							
Tricliceras laceratum (Oberm.) Oberm.				X			
VERBENACEAE							
Chascanum hederaceum (Sond.) Moldenke var. *natalense* (H.Pearson) Moldenke (SDW 816)					X		
Lantana rugosa Thunb. (SDW 820)			X	X	X	X	
Lippia sp.	X					X	
Lippia wilmsii H.Pearson		X	X				

Taxon	CZ	GV	KK	MM	MC	RT	SM
VIOLACEAE							
* *Hybanthus enneaspermus* (L.) F.Muell. (SDW 828)					X		
VITACEAE							
Cissus cornifolia (Baker) Planch.		X		X			
Cissus fragilis E.Mey. ex Kunth				X			
Cissus quadrangularis L. var. *quadrangularis*				X			
Cyphostemma anatomicum (C.A.Sm) Wild & R.B.Drumm	X						
Cyphostemma humile (N.E.Br.) Desc. ex Wild & R.B.Drumm subsp. *dolichopus* (C.A.Sm.) Wild & R.B.Drumm	X						
Cyphostemma lanigerum (Harv.) Desc. ex Wild & R.B.Drumm. (SDW 832)					X	X	
Cyphostemma paucidentatum (Klatt) Desc. ex Wild and R.B.Drumm				X			
Cyphostemma puberulum (C.A. Sm.) Wild & R.B. Drumm. (SDW 668)					X		
Cyphostemma schlechteri (Gilg & Brandt) Desc. ex Wild & R.B. Drumm. (KB, MJB, DG, AP & SDW 10874)					X		
Cyphostemma sp. nov. (KB 12021)		X					
Cyphostemma sp. near.*C. heterotrichum* (Gilg. & R.E.Fr.) Desc. ex Wild & R.B.Drumm.							X
Cyphostemma woodii (Gilg & M.Brandt) Desc.					X		
Rhoicissus rhomboidea (E.Mey. ex Harv.) Planch.				X			
Rhoicissus tridentata (L.f.) Wild & R.B.Drumm subsp. *cuneifolia* (Eckl. & Zeyh.) Urton	X						

Taxon	CZ	GV	KK	MM	MC	RT	SM
Monocotyledons:							
AGAPANTHACEAE							
Agapanthus sp.		X					
ALLIACEAE							
Tulbaghia sp.	X						
Tulbaghia transvaalensis Vosa (KB, SDW & SS 10070)		X					
AMARYLLIDACEAE							
Haemanthus montanus Baker (KB, SDW & SS 9931)		X					
ANTHERICACEAE							
Anthericum sp. 1			X	X		X	
Anthericum sp. 2				X		X	
Chlorophytum angulicaule (Baker) Kativu (SDW 413 & 611)		X			X		
Chlorophytum cooperi (Baker) (KB, SDW & SS 10026 & 9923)		X					
Chlorophytum fasciculatum (Baker) Kativu			X				
Chlorophytum galpinii (Baker) Kativu var. *galpinii* (KB, SDW & SS 10137 & 9684; SDW 823; SDW & TH 889)	X		X		X		
ARACEAE							
Stylochaeton natalensis Schott	X			X	X	X	X
ASPARAGACEAE							
Asparagus cooperi Baker					X		

Taxon	CZ	GV	KK	MM	MC	RT	SM
Asparagus laricinus Burch. (KB, SDW & SS 10076)		X		X			
Asparagus setaceus (Kunth) Jessop				X			
Asparagus sp. 1	1					X	
Asparagus sp. 2					X		X
Asparagus suaveolens Burch.				X			
Asparagus virgatus Baker	X						X
ASPHODELACEAE							
Aloe kniphofioides Baker				X			
Aloe marlothii A.Berger subsp. *marlothii* (KB & SDW 10628)		X	X	X	X	X	
Aloe sp. 1	X					X	
Aloe sp. 2			X				X
Aloe thorncroftii Pole Evans (KB, SDW & SS 10075)		X					
Aloe zebrina Baker (KB, MJB & CK 8185)		X					
Trachyandra reflexipilosa (Kuntze) Oberm. (KB, MJB & CK 8147; SDW 595)		X					
Trachyandra saltii (Baker) Oberm. var. *saltii* (SDW 624)					X		
COLCHICACEAE							
Colchicum burkei (Baker) J.C.Manning & Vinn. (KB, SDW & SS 10079 & 9924)		X					
COMMELINACEAE							
Commelina africana L. var. *africana*				X			
Commelina africana L. var. *krebsiana* (Kunth) C.B.Clarke		X				X	

Taxon	CZ	GV	KK	MM	MC	RT	SM
Commelina diffusa Burm.f. subsp. *scandens* (C.B.Clarke) Oberm. (SDW 357)	X						
Commelina eckloniana Kunth (SDW 765)					X	X	
Commelina modesta Oberm. (SDW 764)					X		
Commelina sp.			X				
Cyanotis sp.			X				
Cyanotis speciosa (L.f.) Hassk.				X			
CYPERACEAE							
Abildgaardia ovata (Burm.f.) Kral (KB, SDW & SS 10088 & 9845; SDW 782 & 520)	X	X	X	X	X	X	
Bulbostylis contexta (Nees) Bodard (SDW 780)					X		
Bulbostylis oritrephes (Redley) C.B.Clarke (SDW 591)		X					
Bulbostylus boeckeleriana (Schweinf.) Beetle						X	
Cyperus glaucophyllus Boeckeler		X					
Cyperus obtusiflorus Vahl var. *flavissimus* (Schrad.) Boeck. (KB, SDW & SS 10129 & 10083; SDW 790)		X	X		X		
Cyperus obtusiflorus Vahl var. *obtusiflorus* (KB, SDW & SS 10132, 10084 & 10082)		X	X				
Cyperus sexangularis Nees (SDW 843)					X		
Cyperus sp. 1			X				
Cyperus sp. 2				X			
Cyperus uitenhagensis (Steud.) C.Archer & Goetgh. (KB, SDW & SS 10133)			X				
Fuirena coerulescens Steud. (SDW 842)					X		

Taxon	CZ	GV	KK	MM	MC	RT	SM
Kyllinga alba Nees (KB, SDW & SS 10126)			X				
Schoenoxiphium sparteum (Wahlenb.) C.B.Clarke (SDW & TH 890)					X		
Scirpus sp.		X					
Scleria bulbifera Hochst. ex A.Rich. (KB, SDW & SS 9844)		X				X	
Scleria sp. 1					X		X
Scleria sp. 2							X
DRACAENACEAE							
Sanseviera hyacynthoides (L.) Druce				X			
ERIOSPERMACEAE							
Eriospermum flagelliforme (Baker) J.C.Manning (KB, MJB & SDW 6678a; KB, SDW & SS 10183 & 10145; SDW 757)			X			X	X
HYACINTHACEAE							
Albuca abyssinica Jacq (KB, MJB & CK 8150)		X					
Dipcadi gracillimum Baker (KB, SDW & SS 10087)		X					
Dipcadi marlothii Medik (SDW 554)			X				
Dipcadi viride (L.) Moench (KB, SDW & SS 10085)		X					
Ledebourea sp. 1			X	X		X	
Ledebourea sp. 2			X	X		X	
Ledebourea sp. 3			X				
Ledebouria burkei (Baker) J.C.Manning & Goldblatt (KB, SDW & SS 10091)		X					

Taxon	CZ	GV	KK	MM	MC	RT	SM
Ledebouria leptophylla (Baker) S.Venter (KB, SDW & SS 10055)		X					
Ledebouria revoluta (L.f.) Jessop (KB, SDW & SS 9925)		X					
Merwilla plumbea (Lindl.) Speta (KB, MJB & CK 8136)		X			X		X
Schizocarphus nervosa (Burch.) Van der Merwe			X				
HYPOXIDACEAE							
Hypoxis filiformis Baker (KB, SDW & SS 10161)			X				
Hypoxis hemerocallidea Fisch. & C.A.Meyer (KB, SDW & SS 10143)			X				
Hypoxis multiceps Buchinger ex Baker (KB, SDW & SS 10116 & 9902)		X	X				
Hypoxis rigidula Baker var. *pilosissima* Baker (KB, SDW & SS 9901; SDW 853)		X			X		
Hypoxis rigidula Baker var. *rigidula* (KB, SDW & SS 10119)			X				
Hypoxis sp. near *H. acuminata* Baker & *H. rigidula* Baker (KB, MJB & CK 8131)		X					
Hypoxis sp. 1	X					X	X
Hypoxis sp. 2				X		X	X
IRIDACEAE							
Aristea torulosa Klatt. (SDW 657)						X	
Gladiolus elliotii Baker (SDW 412)		X					
Gladiolus serpenticola Goldblatt & Manning (KB, MJB, DG, AP & SDW 10865)					X		
Gladiolus sp. 1	X					X	X

Taxon	CZ	GV	KK	MM	MC	RT	SM
Gladiolus sp. 2							X
Gladiolus vinosomaculatus Kies (KB 9056)		X					
Gladiolus woodii Baker (KB, SDW & SS 10134)			X				
Watsonia densiflora Baker (SDW 633)		X					
ORCHIDACEAE							
Eulophia speciosa (R.Br. ex Lindl.) Bolus		X		X			
POACEAE							
**Cymbopogon pospishilii* (K.Schum.) C.E.Hubb.						X	
Agrostis lachnantha Nees var. *lachnantha* (MS 2901)						X	
Alloteropsis semialata (R.Br.) Hitchc. subsp. *eckloniana* (Nees) Gibbs-Russ. (KB, SDW & SS 9958; SDW 787)		X			X		
Andropogon fastigiatus Sw. (MS 2897)						X	
Andropogon shirensis Hochst. ex A.Rich.		X					
Aristida bipartita (Nees) Trin. & Rupr.		X					
Aristida congesta Roem. & Schult. subsp. *barbicollis* (Trin. & Rupr.) De Winter (MS 3026)				X	X	X	
Aristida congesta Roem. & Schult. subsp. *congesta*							X
Aristida sp.				X			X
Aristida transvaalensis Henrard		X					
Bewsia biflora (Hack.) Gooss. (SDW 584)		X	X		X	X	X
Bothriochloa insculpta (A.Rich.) A.Camus (SDW 509; MS 3028)	X				X	X	
Brachiaria brizantha (A.Rich.) Stapf (SDW 517 & 784)	X				X		X

Taxon	CZ	GV	KK	MM	MC	RT	SM
Brachiaria serrata (Thunb.) Stapf (KB, MJB & CK 8160; KB, SDW & SS 10095)		X			X		X
Cymbopogon caesius (Hook. & Arn.) Stapf (MS 3025)		X		X	X		X
Cymbopogon sp.	X		X				X
Digitaria diagonalis (Nees) Stapf. var. *diagonalis*		X				X	X
Digitaria eriantha Steud.				X	X		
Digitaria sp. 1			X	X			X
Digitaria sp. 2			X				
Diheteropogon amplectens (Nees) Clayton (MS 3027; SDW 626)		X		X	X	X	X
Diheteropogon sp.							X
Elionurus muticus (Spreng.) Kunth (KB, SDW & SS 10086 & 9957; SDW 622 & 783)				X	X		
Enneapogon cenchroides (Roem. & Schult.) C.E.Hubb. (MS 3029)					X		
Enneapogon scoparius Stapf (MS 2898)				X		X	
Eragrostis barbinodis Hack.(MS 3035)						X	
Eragrostis capensis (Thunb.) Trin. (KB, SDW & SS 9934a)		X	X				
Eragrostis chloromelas Steud. (MS 3024)					X		
Eragrostis cilianensis (All.) F.T.Hubb. (MS 2933)						X	
Eragrostis curvula (Schrad.) Nees							X
Eragrostis heteromera Stapf (KB, SDW & SS 9940)		X					
Eragrostis patentipilosa Hack. (KB, SDW & SS 9938)		X					
Eragrostis racemosa (Thunb.) Steud. (SDW 781)		X	X		X	X	

Taxon	**CZ**	**GV**	**KK**	**MM**	**MC**	**RT**	**SM**
Eragrostis rigidior Pilg.				X			
Eragrostis sp. 1	X			X	X		X
Eragrostis sp. 2	X		X				
Eragrostis superba Peyr. (SDW 788)				X	X		
Eragrostis tenuifolia (A.Rich.) Steud.			X			X	
Eulalia villosa (Thunb.) Nees (SDW 610)		X				X	
Eustachys paspaloides (Vahl) Lanza & Mattei (SDW 840)					X		
Fingerhutia africana Lehm.				X			
Heteropogon contortus (L.) Roem. & Schult. (SDW 785)	X	X	X	X	X	X	X
Hyparrhenia hirta (L.) Stapf (MS 3021)		X			X		
Hyperthelia dissoluta (Nees ex Steud.) Clayton (SDW 515)	X					X	
Imperata cylindrica (L.) Reausch. (KB, SDW & SS 10089)		X					
Koeleria capensis (Steud.) Nees (KB, MJB & CK 8164)		X					
Loudetia simplex (Nees) C.E. Hubb (KB 9047; KB, MJB & CK 8163; MS 3022; SDW 583; 617 & 519)	X	X	X		X		X
Melinis nerviglumis (Franch.) Zizka (KB, SDW & SS 10153 & 9937; SDW 779)		X	X	X	X		
Melinis repens (Willd.) Zizka subsp. *repens* (KB, SDW & SS 10093; SDW 792)	X	X	X	X	X	X	X
Microchloa caffra Nees		X	X				
Monocymbium ceresiformi (Nees) Stapf							X
Panicum coloratum L. var. *coloratum*				X			
Panicum maximum Jacq. (MS 3023; SDW 671)	X			X	X		
Panicum natalense Hochst. (SDW 582)		X	X		X		
Panicum sp.				X			

Taxon	CZ	GV	KK	MM	MC	RT	SM
Paspalum scrobiculatum L.	X		X				
Sartidia dewinteri J. Munday & L. Fish. (KB 9043; SDW 579 & 640)		X				X	X
Schizachyrium sanguineum (Retz.) Alston (SDW 580b)		X					X
Setaria incrassata (Hochst.) Hack. (MS 2967)						X	
Setaria nigrirostris (Nees) T.Durand & Schinz (KB, SDW & SS 9934)		X					
Setaria sp.			X				
Setaria sphacelata (Schumach.) Moss var. *sphacelata* (KB, SDW & SS 9935; SDW 581)	X	X			X	X	X
Sporobolus africanus (Poir.) Robyns & Tournay			X				
Sporobolus pectinatus Hack. (KB, MJB & CK 8162)		X					
Sporobolus sanguineus Rendle (SDW 789)					X		
Themeda triandra Forssk. (KB, SDW & SS 9933; SDW 514 & 786)	X	X	X	X	X	X	X
Trachypogon spicatus (L.f.) Kuntze (SDW 580a)		X		X	X	X	X
Tragus berteronianus Schult.				X			
Tricholaena monachme (Trin.) Stapf & C.E.Hubb.					X		X
Tristachya leucothrix Nees (KB, SDW & SS 10092 & 10152; SDW 791)		X	X	X	X	X	X
SMILACACEAE							
Smilax anceps Willd.	X						
VELLOZIACEAE)							

Taxon	CZ	GV	KK	MM	MC	RT	SM
Xerophyta retinervis Baker (KB, MJB & CK 8127; KB, SDW & SS 10078)		X			X		
Xerophyta villosa (Baker) Smith & Ayensu (KB, MJB & SDW 6683a							X

Appendix B

Plant taxa recorded from adjacent non-serpentine areas but not found on serpentine outcrops. # indicates a family and ^ indicates a genus not represented in the serpentine flora.

Family	Species	
Dicotyledons		
Acanthaceae	*Barleria elegans* S.Moore ex C.B.Clarke	Perennial herb
Amaranthaceae	^*Pupalia lappacea* (L.) A.Juss. var *lappacea*	Annual herb
Apiaceae	*Heteromorpha stenophylla* Welw. ex *Schinz var. transvaalensis* (Schltr. & H.Wolff) P.J.D.Winter	Suffrutex
Apocynaceae	^*Acokanthera oppositifolia* (Lam.) Codd	Shrub/tree
	Ceropegia carnosa E.Mey.	Herbaceous climber
	^*Riocreuxia polyantha* Schltr.	Herbaceous climber
	^*Sarcostemma viminale* (L.) R.Br.	Woody climber
Asteraceae	*Berkheya densifolia* Bohnen ex Roessler	Perennial herb
	^*Chrysocoma ciliata* (L)	Shrub
	^*Psiadia punctulata* (DC.) Vatke	Shrub
	Senecio inornatus DC.	Perennial herb
	Senecio polyanthemoides Sch.Bip.	Annual herb
	Vernonia centaureoides Klatt	Annual herb
Boraginaceae	^*Ehretia rigida* (Thunb.) Druce	Shrub/tree
Burseraceae	*Commiphora mollis* (Oliv.) Engl.	Tree
Capparaceae	*Maerua rosmarinoides* (Sond.) Gilg & Gilg-Ben.	Shrub/tree
Celastraceae	*Maytenus acuminata* (L.f.) Loes.	Shrub/tree
Chrysobalanaceae	*Parinari curatellifolia* Planch. Ex Benth.	Tree
Convolvulaceae	^*Cuscuta cassytoides* Engelm.	Annual herb
	^*Hewittia malabarica* (L.) Suresh	Herbaceous climber
Fabaceae	^*Abrus laevigatus* E.Mey.	Woody climber
	Acacia tortilis (Forssk.) Hayne	Shrub/tree
	Eriosema lucipetum C.H.Stirt.	Perennial herb

Family	Species	
	^*Mundulea sericea* (Willd.) A.Chev.	Shrub/tree
	^*Pterocarpus angolensis* DC.	Tree
	^*Pterocarpus rotundifolius* (Sond.) Druce	Shrub/tree
#Kirkiaceae	*Kirkia wilmsii* Engl.	Tree
Lamiaceae	*Leucas glabrata* (Vahl) Sm. var glabrata	Perennial herb
	Leucas sexdentata Skan	Annual herb
Lythraceae	^*Galpinia transvaalica* N.E.Br.	Shrub/tree
Malvaceae	*Grewia flavescens* Juss.	Shrub
	Grewia hexamita Burret	Shrub/tree
	Melhania didyma Eckl. & Zeyh.	Dwarf shrub
	Melhania integra I.Verd.	Dwarf shrub
	Sida pseudocordifolia Hochr.	Perennial herb
Menispermaceae	*Cissampelos mucronata* A.Rich.	Woody climber
Molluginaceae	*Psammotropha mucronata* (Thunb.) Druce	Perennial herb
Moraceae	*Ficus ingens* (Miq.) Miq.	Tree
Phyllanthaceae	^*Antidesma venosum* E.Mey. ex Tul.	Shrub/tree
Rubiaceae	*Fadogia tetraquetra* K.Krause var *tetraquetra*	Perennial herb
	^*Psydrax locuples* (K.Schum.) Bridson	Shrub/tree
Rutaceae	^*Vepris reflexa* I.Verd.	Shrub/tree
Sapindaceae	*Allophyllus decipiens* (Sond.) Radlk.	Shrub/tree
Sapotaceae	*Englerophytum natalense* (Sond.) T.D.Penn.	Shrub/tree
Scrophulariaceae	*Zaluzianskya distans* Hiern	Perennial herb
#Strychnaceae	^*Strychnos decussata* (Pappe) Gilg	Shrub/tree
	^*Strychnos madagascariensis* Poir	Shrub/tree
Monocotyldons		
#Amaryllidaceae	^*Boophone disticha* (L.f.) Herb.	Geophyte
Poaceae	*Andropogon chinensis* (Nees) Merr.	Perennial
	Aristida adscensionis (L.)	Annual

Family	Species	
	Sporobolus nitens Stent	Perennial
	^*Trichoneura grandiglumis* (Nees) Ekman	Perennial
	^*Urelytrum agropyroides* (Hack.) Hack.	Perennial

Appendix C

List of voucher specimens collected for each taxon endemic to the serpentine outcrops of the Barberton Greenstone Belt, excluding outcrops in Swaziland. Taxa are listed in alphabetical order.

Aloe thorncroftii Pole Evans

- 2530DD South Africa, Mpumalanga, Carolina District, Stolzburg Syncline, on the farm Groenvaly 701JT. On serpentine. 12/10/1997 (K. Balkwill, S. Williamson & S. Smith 10075)
- 2531CC South Africa, Mpumalanga, Barberton District, Barberton. ??/07/1917 (F.A. Rogers 20250)

Asystasia subbiflora C.B. Clarke

- 2530DD South Africa, Mpumalanga, Barberton District, Queens River Valley, on road from Barberton to Nelshoogte Forestry Reserve. 21.3 km from Barberton. 11/01/1985 (K. Balkwill & M-J. Cadman 2615)
- 2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. On SERPENTINE, on light coloured ridge. 09/01/1985 (K. Balkwill & M-J. Cadman 2590)
- 2530DD South Africa, Mpumalanga, Barberton District, about 10 km westof Barberton, in the valley above Agnes mine. Property of Twello Forestry. ON SERPENTINE. 20/03/1991 (S.D. Williamson, K. Balkwill and M-J. Balkwill 54)
- 2530DD South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, on the farm Goudgenoeg 758 JT, on serpentine. 10/12/1992 (K. Balkwill 7763)
- 2530DD South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, on the farm Goudgenoeg 758 JT, on serpentine. At the end of the track. 05/12/1991 (K. Balkwill, M-J. Balkwill & S.D. Williamson 6680)
- 2530DD South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, on the farm Goudgenoeg 758 JT, on serpentine. 05/12/1991 (M. Stalmans 2532)

2530DD South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, on the farm Goudgenoeg 758 JT, on serpentine. Central part, saddle on watershed Komati-Msauli. 16/04/1992 (M. Stalmans 2760)

2530DD South Africa, Mpumalanga, Carolina District, Stolzburg Syncline, on the farm Groenvaly 701JT. On serpentine. 10/10/1997 (K. Balkwill, S. Williamson & S. Smith 9873)

Athrixia sp. nov.

2530DD South Africa, Mpumalanga, Carolina District, Stolzburg Syncline, on the farm Groenvaly 701JT. On serpentine. 30/03/1994 (K. Balkwill 9052)

2530DD South Africa, Mpumalanga, Carolina District, Stolzburg Syncline, on the farm Groenvaly 701 JT. On serpentine. 20/01/1998 (S.D.Williamson 587)

Berkheya coddii Roessler

2530DC South Africa, Mpumalanga, Carolina District, Stolzburg Syncline, on the farm Groenvalei. ON SERPENTINE. 04/03/1997 (S.D. Williamson 406)

2530DD South Africa, Mpumalanga, Barberton District, 22 km from Barberton Queens River Valley. In enclosure with sawmill. 13/10/1997 (K. Balkwill, S. Williamson & S. Smith 10219)

2530DD South Africa, Mpumalanga, Barberton District, Queens River Valley, 20.5 km from Barberton on road to Nelshoogte Forestry Reserve. ON SERPENTINE. 26/04/1984 (K. Balkwill 1460)

2530DD South Africa, Mpumalanga, Barberton District, Queens River Valley, on road from Barberton to Nelshoogte Forestry Reserve. 21.3 km from Barberton. Near the border of the forest reserve. 06/12/1991 (K. Balkwill, M-J. Balkwill & S.D. Williamson 6680a)

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. 23/01/1992 (K. Balkwill 6884)

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. On slope below road facing ridge. ON SERPENTINE. 09/01/1985 (K. Balkwill & M-J. Cadman 2566)

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. ON SERPENTINE. 20/03/1991 (S.D. Williamson, K. Balkwill and M-J. Balkwill 52)

2530DD South Africa, Mpumalanga, Carolina District, about 25 km east of Badplaas, on the Stolzburg Syncline. ON SERPENTINE. 03/05/1987 (K. Balkwill, M-J. Cadman & D.R. Morrey 3750)

2530DD South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, on the farm Goudgenoeg 758 JT, on serpentine. At the end of the track. 05/12/1991 (K. Balkwill, M-J. Balkwill & S.D. Williamson 6693)

2530DD South Africa, Mpumalanga, Barberton District, Nelshoogte Forestry Reserve, on the farm Hilversum 696 JT. On SERPENTINE. Queens River Valley, on the farm Nelshoogte. 10/06/1996 (S.D. Williamson & C. Payet 278)

2530DD South Africa, Mpumalanga, Carolina District, Stolzburg Syncline, on the farm Groenvaly 701JT. On serpentine. Road running from boundary serpentine back to Moses' house. 27/03/1993 (K. Balkwill & M-J. Balkwill 8188)

2530DD South Africa, Mpumalanga, Carolina District, Stolzburg Syncline, on the farm Groenvaly 701JT. On serpentine. Near south western border, along forest track. On turn off-38 km on R38 road. 12/06/1996 (S.D. Williamson & C. Payet 290)

2531CC South Africa, Mpumalanga, Barberton District, Barberton. 24 km west of town. 08/12/1953 (L.E.W. Codd 8148)

2531CC South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve, Dunbar, Msauli Valley, southern part of Msauli Serpentine. 12/04/1991 (M. Stalmans 2203)

2531CC South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve, Diepgezet, on serpentine. About 2 km north of the gate north of Msauli Mine. 05/12/1991 (K. Balkwill, M-J. Balkwill & S.D. Williamson 6646)

2531CC South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve, on the farm Dunbar 383 JU. On Serpentine. 08/12/1992 (G.V. Cron 126);

2531CC South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve, on the farm Dunbar 383 JU. On Serpentine. Near head of Dunbar Valley. 23/03/1995 (K. Balkwill 9207)

2531CC South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve, on the farm Dunbar 383 JU. On Serpentine. 08/12/1992 (K. Balkwill et al. 7710)

2531DC South Africa, Mpumalanga, Kamhlushwa District, Kangwane-Nkomazi west region. Along Swaziland boundary, near the eastern boundary of Jeppes Rust, 2km NW of Nsulaze. 04/05/1992 (M. Stalmans 2774)

2531DC South Africa, Mpumalanga, Kamhlushwa District, Swaziland border, Nzulase Koppie. On Serpentine. 19/11/1993 (K. Balkwill & M-J. Balkwill 8607)

2630BB South Africa, Mpumalanga, Eerstehoek District, Songimvelo Nature Reserve, on the farm Rozentuin 159 IT. On serpentine. Valley on Rozentuin-Goudgenoeg boundary. 09/10/1991 (M. Stalmans 2359)

2631AA South Africa, Mpumalanga, Eerstehoek Dist., Songimvelo Game Reserve, About 30 km S, of Barberton, on the farm Diepgezet, near Msauli Mine. Lower reaches on east facing side of valley but north facing slope of small spur. 06/03/1993 (K. Balkwill & C. Kidger 7946)

Berkheya nivea N.E. Br.

2530DB South Africa, Mpumalanga, Nelspruit District, Barberton Game Reserve, on the farm Hillside 459 JT. ON SERPENTINE. 11/10/1997 (K. Balkwill, S. Williamson & S. Smith 10016)

2531CA South Africa, Mpumalanga, Barberton District. Cerro de Pasco Mine. ??/11/1891 (E.E. Galpin 1356)

2531CA South Africa, Mpumalanga, Barberton District, Barberton phase 2, Mundt's , Concession. On SERPENTINE. 31/01/1999 (K. Balkwill, M-J. Balkwill, D. Goyder, A. Paton & S.D. Williamson 10869)

2531CA South Africa, Mpumalanga, Barberton District, Barberton phase 2, Mundt's , Concession. On SERPENTINE. Hill 737, east of Opsaal.18/11/1998 (M. Stalmans 3005)

2531CA South Africa, Mpumalanga, Barberton District, Barberton phase 2, Mundt's , Concession. On SERPENTINE. 28/11/1998 (S.D. Williamson 815)

2632AA Swaziland, North west of Mbabane, Silotfwane Hills, Malolotja Game, Reserve. Around a saddle at parking spot at Silotfwane Viewpoint. 24/06/1995 (K. Balkwill & M-J. Balkwill 9328)

Berkheya sp. aff. *B. seminivea* Harv. & Sond.

2530DD South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, on the farm Goudgenoeg 758 JT, on serpentine. At the end of the track. 05/12/1991 (K. Balkwill, M-J. Balkwill & S.D. Williamson 6694)

2530DD South Afria. Mpumalanga. Eerstehoek District. Songimvelo Game Reserve, on the farm Geluk 732 JT, south west corner of farm. 24/02/1995 (K. Balkwill 9194)

Berkheya zeyheri (Sond. & Harv.) Oliv. & Hiern ssp. *rehmannii* (Thell.)Roessl. var. *rogersiana* (T-.) R-.

2530DB South Africa, Mpumalanga, Nelspruit District, 17 km from Nelspruit on Barberton Road. ON SERPENTINE. 28/04/1996 (S.D. Williamson & J. Williamson 256)

2530DB South Africa, Mpumalanga, Nelspruit District, Berlin State Forest, about 6 km east of Kaapsehoop Ridge opposite Kaapsehoop Asbestos mine. On SERPENTINE. 30/10/1986 (K. Balkwill, M-J.Cadman 3579)

2530DD South Africa, Mpumalanga, Barberton District, Nelshoogte Forestry Reserve, on the farm Hilversum 696 JT. On SERPENTINE. Queens River Valley, on the farm Nelshoogte. 10/06/1996 (S.D. Williamson & C. Payet 279a)

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. 20/03/1991 (K. Balkwill, M-J. Balkwill & S. Wiliamson 6262)

2530 DD South Africa, Mpumalanga, Carolina District, Stolzburg Syncline, on the farm , Groenvaly 701JT. On serpentine, on the boundary with Doyershoek 10/10/1997 (K. Balkwill, S. Williamson & S. Smith 9874)

2530DD South Africa, Mpumalanga, Carolina District, on the farm Groenvaly 701 JT. On serpentine, on firebreak adjacent to Morgenzon. Not Stolzburg Syncline. 10/10/1997 (K. Balkwill, S. Williamson & S. Smith 9944)

2530DD South Africa, Mpumalanga, Carolina District, Stolzburg Syncline, on the farm Groenvaly 701 JT. 26/03/1993 (K. Balkwill, M-J. Balkwill & C. Kidger 8149)

Brachystelma dyeri K. & M-J. Balkwill

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. ON SERPENTINE. 29/02/1987 (K. Balkwill & M-J. Cadman 3393)

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton ridge overlooking Agnes mine. On slope nearest the road. 26/01/1992 (S.D. Williamson & J. Williamson 114)

Brachystelma sp. nov. aff *B longifolium* (Schltr.) N.E. Br.

2531CA South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve, on the farm Heemstede 378 JU. On Serpentine. 19/11/1993 (K. Balkwill & M-J. Balkwill 8559)

Cyphia bolusii E. Phillips

2530DB South Africa, Mpumalanga, Nelspruit District, Berlin State Forest, about 6 km east of Kaapsehoop Ridge opposite Kaapsehoop Asbestos mine. On SERPENTINE. 30/12/1987 (K. Balkwill & M-J. Balkwill 3982)

2530DB South Africa, Mpumalanga, Nelspruit District, Berlin State Forest, about 6 km east of Kaapsehoop Ridge opposite Kaapsehoop Asbestos mine. On lower slopes, near asbestos claims. ON SERPENTINE ridge. 30/10/1986 (K. Balkwill & M-J. Cadman 3580)

2530DD South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, on the farm Uitval 376 JT. Boundary with Granville Grove. 09/12/1992 (W.N. Ellery & K. Ellery 92/120)

Dicoma swazilandica S. Ortiz, Rodr. Oubina & Pulgar

2530DD South Africa, Mpumalanga, Carolina District, Stolzburg Syncline, on the farm Groenvaly 701 JT. 27/03/1993 (K. Balkwill & M-J. Balkwill 8178)

2531 CC South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve on the farm Dunbar 383 JU. On Serpentine. 01/04/1996 (A. Lee 107)

2630BB South Africa, Mpumalanga, Eerstehoek District, Songimvelo Nature Reserve, on the farm Onverwacht 733 JT. On serpentine. In from gate with day visitors centre. 19/03/1992 (K. Balkwill & E.R. Robinson 6831)

Gladiolus serpenticola Goldblatt & Manning

2531CA South Africa, Mpumalanga, Barberton District, Barberton phase 2, Mundt's Concession. On SERPENTINE. 22/07/2000 (S.D. Williamson 869)

Gymnoporium sp. nov.

2530DB South Africa, Mpumalanga, Nelspruit District, 17 km from Nelspruit on Barberton Road. On the farm Hillside 459 JT. ON SERPENTINE. 07/10/2001 (S.D. Williamson 945)

2531CA South Africa, Mpumalanga, Barberton District, Barberton phase 2, Mundt's Concession. On SERPENTINE. 31/01/1999 (K. Balkwill, M-J. Balkwill, D. Goyder, A. Paton & S.D. Williamson 10865)

2531CA South Africa, Mpumalanga, Barberton District, Barberton phase 2, Mundt's Concession. On SERPENTINE. 1/10/2000 (S.D. Williamson & T. Herron 887)

Helichrysum sp. nov. c.f. *H. albo-brunneum* S.Moore

2531CC South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve on the farm Dunbar 383 JU. On Serpentine. 01/04/1996 (A. Lee s.n.)

2531CC South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve, on the farm Dunbar 383 JU. On Serpentine. 25 58' 78.9" S; 31 04' 24.2" E. 16/03/1995 (F. Hologne 551)

Indigofera crebra N.E. Br.

2530DD South Africa, Mpumalanga, Barberton District, Queens River Valley, 14.5 km from Barberton on road to Nelshoogte Forestry Reserve. 04/12/1991 (K. Balkwill, M-J. Balkwill & S.D. Williamson 6617a)

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. 01/11/1986 (K. Balkwill & M-J. Cadman 3620)

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. 04/12/1991 (K. Balkwill, M-J. Balkwill & S.D. Williamson 6603)

2530DD South Africa, Mpumalanga, Barberton District, east of Barberton, ridge overlooking Agnes mine between Thorncroft and Cythna Letty. Near Agnes mine. 31/12/1987 (K. Balkwill & M-J. Balkwill 3999)

2530DD South Africa, Mpumalanga, Carolina District, on the farm Groenvaly 701 JT. On serpentine. 10/10/1997 (K. Balkwill, S. Williamson & S. Smith 9894)

2531CC South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve, on the farm Loenen 381 JU. Mlembe Mountain. 07/12/1992 (G. Germishuizen 5677)

2632AA Swaziland, North west of Mbabane, Malolotja Game Reserve. On SERPENTINE. Luhhumanani School. 11/10/1997 (D.A. McCallum & M-J. Balkwill 396)

2631AC Swaziland, Mbabane District, about 5 km south of Oshoek, Mbabane road, Motjane Serpentine Band. Atlas Engineering site. 10/11/1997 (D.A. McCallum 381)

2631AC Swaziland, Mbabane District, about 5 km south of Oshoek, Mbabane road, Motjane Serpentine Band. Near school. 12/11/1997 (D.A. McCallum 453)

Macledium zeyheri subsp. *thyrsifolium* (Klatt)

2531CB South Africa, Mpumalanga, Malelane District, in conservancy on the farm Strathmore 214 JU, 9/10/1997 (K. Balkwill, S. Williamson & S. Smith 9819)

Ocimum sp. nov. 1

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. On SERPENTINE., On light coloured ridge 20/03/1991 (K. Balkwill, M-J. Balkwill & S. Williamson 6273)

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. On SERPENTINE, On light coloured ridge 04/12/1991 (K. Balkwill, M-J. Balkwill & S. Williamson 6613)

2530DD South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, on the farm Goudgenoeg 758 JT, on serpentine. 10/12/1992 (K. Balkwill 7765)

2530DD South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, on the farm Goudgenoeg 758 JT, on serpentine. 10/12/1992 (S.D. Williamson 211)

2531CC South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve, Dunbar Msauli Valley, southern part of Msauli Serpentine. 08/12/1992 (I.H. Hartley 1291)

2531CC South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve, Diepgezet, on serpentine. Top part. 25/03/1993 (K. Balkwill 8250a)

2531CC South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve, on the farm Dunbar 383 JU. On Serpentine. 08/12/1992 (K. Balkwill et al. 7706)

2531CC South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve, on the farm Dunbar 383 JU. On Serpentine. 07/12/1992 (S.D. Williamson 160)

2531CA South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve, on the farm Heemstede 378 JU. On Serpentine. 19/11/1993 (K. Balkwill & M-J. Balkwill 8564)

2630BB South Africa, Mpumalanga, Eerstehoek District, Songimvelo Nature Reserve, on the farm Doornhoek South West. Ultramafic soils. Ridge above saddle. 02/02/1999 (K. Balkwill, M-J. Balkwill, M. Stalmans, D. Goyder & A. Paton 10901)

2630BB South Africa, Mpumalanga, Eerstehoek District, Songimvelo Nature Reserve, Hooggenoeg 160 IT. On serpentine. Above Komati River. 11/12/1992 (K. Balkwill 7814)

2631AC Swaziland, Mbabane District, about 5 km south of Oshoek Mbabane road, Motjane Serpentine Band. East of Makhwana Primary School. 10/10/1997 (D.A. McCallum & M-J. Balkwill 352)

2630BB South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, on the farm Rosentuin 159 IT, On Serpentine. 12/08/1998 (S.D. Williamson 751)

2531CA South Africa, Mpumalanga, Barberton District, Barberton Reserve Phase 1 on the farm Hillside 459 JT. 16/01/2001 (S.D. Williamson 899)

Ocimum sp. nov. 2

2530DD South Africa, Mpumalanga, Barberton District, 22 km from Barberton Queens River Valley. In enclosure with sawmill. 13/10/1997 (K. Balkwill, S. Williamson & S. Smith 10208)

Ozoroa barbertonensis Retief

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. On the whitish green band of serpentine. 31/12/1987 (K. Balkwill & M-J. Balkwill 4001)

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. On the ridge facing the road. ON SERPENTINE. 01/11/1986 (K. Balkwill & M-J. Cadman 3604)

2530 DD South Africa, Mpumalanga, Barberton District, about 22 km W of Barberton on the farm Hilversum 696 JT. 07/08/1998 (S. Williamson 741)

Ozoroa sp. nov.

2530DD South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, on the farm Doornhoek. North of Komati River on border with Geluk. 08/04/1993 (M. Stalmans 2856)

2531CC South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve, on the farm Dunbar 383 JU. On Serpentine. Hill south of game scout hut. 04/04/1996 (A. Lee 166;

2630BB South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, On the farm Doornhoek. On serpentine. Northern part. 24/04/1993 (K. Balkwill et al. 8270;

2630BB South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, On the farm Doornhoek. On serpentine. South west. On ridge above saddle. Near western border of reserve. 24/04/1993 (K. Balkwill et al. 8948)

2630BB South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, On the farm Doornhoek. On serpentine. South west. Near base of hill. Near western border of reserve. 24/04/1993 (K. Balkwill et al, 8968;

2630BB South Africa, Mpumalanga, Eerstehoek District, Songimvelo Nature Reserve, on the farm Doornhoek South West. On saddle between hills 1014 & 1008. 29/09/2000 (K. Balkwill 12014)

2630BB South Africa, Mpumalanga, Eerstehoek District, Songimvelo Nature Reserve, on the farm Onverwacht 733 JT. 10/12/1992 (E. Masilo E 90)

2631AA South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, on the farm Kromdraai. Kromdraai Hill. 26/03/1993 (K. Balkwill, M-J. Balkwill & C. Kidger 8096)

2630BB South Africa, Mpumalanga, Eerstehoek District, Songimvelo Nature Reserve, Hooggenoeg 160 IT. On serpentine. Above Komati River. 11/12/1992 (K. Balkwill 7840)

Protea curvata N.E.Br.

2531CA South Africa, Mpumalanga, Barberton District, Barberton phase 2, Mundt's Concession. On SERPENTINE. 22/07/2000 (S.D. Williamson 864)

Salpinctium hirsutum T.J. Edwards

2530DB South Africa, Mpumalanga, Nelspruit District, Berlin State Forest, about 7.5 km E of Kaapsehoop, same side of road as Kaapeshoop Asbestos Mine. Lower section of serpentine. 07/12/1991 (K. Balkwill, M-J. Balkwill & S.D. Williamson 6704)

2530DB South Africa, Mpumalanga, Nelspruit District, on the road to Kaapsehoop. Ca. 22.5 km W of Nelspruit. 02/03/1999 (T.F. Daniel, K. Balkwill & M. Butterwick 9390)

2531CC South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve, Diepgezet, on serpentine. About 2 km north of the gate north of Msauli Mine. At the point where track crosses river. 05/12/1991 (K. Balkwill, M-J. Balkwill & S.D. Williamson 6659)

2631AA South Africa, Mpumalanga, Eerstehoek Dist., Songimvelo Game Reserve, about 30 km south of Barberton, on the farm Diepgezet 388 JU. Just below the picket. 05/12/1991 (K. Balkwill, M-J. Balkwill & S.D. Williamson 6666)

2631AC Swaziland, Mbabane District, about 5 km south of Oshoek, Mbabane road, Motjane Serpentine Band. Near school. 12/11/1997 (D.A. McCallum 445)

Sartidia dewinteri J.Munday & L.Fish

2530DB South Africa, Mpumalanga, Nelspruit District, Berlin State Forest, about 6 km east of Kaapsehoop Ridge opposite Kaapsehoop Asbestos mine. Near top of ridge. 08/01/1993 (K. Balkwill 7879)

2530DB South Africa, Mpumalanga, Nelspruit District, Berlin State Forest, about 6 km east of Kaapsehoop Ridge opposite Kaapsehoop Asbestos mine. On SERPENTINE. 30/12/1987 (K. Balkwill & M-J. Balkwill 3983)

2530DB South Africa, Mpumalanga, Nelspruit District, Berlin State Forest, about 6 km east of Kaapsehoop Ridge opposite Kaapsehoop Asbestos mine. ON SERPENTINE. 27/04/1991 (K. Balkwill, M-J. Balkwill & S.D. Williamson 6434)

2530DB South Africa, Mpumalanga, Nelspruit District, Berlin State Forest, about 6 km east of Kaapsehoop, ridge opposite Kaapsehoop Asbestos mine. 23/01/1992 (K. Balkwill 6876)

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. Between Cythna Letty and Thorncroft Nature Reserves. On light coloured band facing the road. 31/12/1987 (K. Balkwill & M-J. Balkwill 4016)

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. On SERPENTINE. 09/01/1985 (K. Balkwill & M-J. Cadman 2584)

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. ON SERPENTINE, 20/03/1991, (S.D. Williamson, K. Balkwill and M-J. Balkwill 53)

2530DD South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, on the farm Goudgenoeg 758 JT, on serpentine. Manana's Kraal. 10/12/1992 (M. Stalmans 2829)

2530DD South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, on the farm Geluk 732 JT. South west corner of farm, but south east corner of portion within Songimvelo. 24/02/1995 (K. Balkwill 9195)

2530DD South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, on the farm Geluk 732 JT. Western boundary fence. 20/02/1992 (M. Stalmans 2679)

2530DD South Africa, Mpumalanga, Barberton District, east of Barberton, ridge overlooking Agnes mine, between Thorncroft and Cythna Letty. Near Agnes mine. 31/12/1987 (K. Balkwill & M-J. Balkwill 3996)

2530DD South Africa, Mpumalanga, Carolina District, Stolzburg Syncline, on the farm Groenvaly 701 JT. On serpentine. 30/03/1994 (K. Balkwill 9043)

2531CC South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve, on the farm Dunbar 383 JU. On Serpentine. North-west of the game scout camp. 06/04/1996 (A. Lee 126)

2531CC South Africa, Mpumalanga, Barberton District, Songimvelo Game Reserve, on the farm Dunbar 383 JU. On Serpentine. Near head of floor of valley. 23/02/1995 (K. Balkwill 9167)

2630BB South Africa, Mpumalanga, Eerstehoek District, Songimvelo Nature Reserve, on the farm Rosentuin 159 IT. On serpentine. 4/2/1998 (S.D.Williamson 640)

2630BB South Africa, Mpumalanga, Eerstehoek District, Songimvelo Nature Reserve, on the farm Kortbegrip 168 IT. On serpentine. 23/02/1995 (K. Balkwill 9148)

2631AA South Africa, Mpumalanga, Eerstehoek District, Songimvelo Game Reserve, on the farm Kromdraai. Kromdraai Hill. 26/03/1993 (K. Balkwill, M-J. Balkwill & C. Kidger 8097)

Sclerochiton triacanthus A. Meeuse

2531CA South Africa, Mpumalanga, Barberton District, 8 km north of Barberton, Caledonian. ??/11/1970 L.E. Davidson s.n.)

2531CA South Africa, Mpumalanga, Barberton District, north of Barberton, about 13 km along, M40 to Noordkop. Track towards north, to hills with serp. 04/03/2000 (L.A. McDade & K. Balkwill 1255)

Searsia pygmaea (Moffett) Moffett

2530DD South Africa, Mpumalanga, Barberton District, Queens River Valley, on road from Barberton to Nelshoogte Forestry Reserve. 09/01/1985 (K. Balkwill & M-J. Cadman 2611)

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. 23/01/1992 (K. Balkwill 6883)

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. Foot of facing slope. 04/12/1991 (K. Balkwill, M-J. Balkwill & S.D. Williamson 6596)

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, in the valley above Agnes mine. Property of Twello Forestry. On ridge facing the road.ON SERPENTINE. 02/05/1987 (K. Balkwill, M-J. Cadman & D.R. Morrey 3728)

2530DD South Africa, Mpumalanga, Barberton District, about 10 km west of Barberton, ridge overlooking Agnes mine. West of Agnes Mine. Between mine and Twello Plantation. 13/04/1995 (R.O. Moffett 4904)

2531CA South Africa, Mpumalanga, Barberton District, Barberton phase 2, Mundt's Concession. On SERPENTINE. North of Noordkaap Road, at the junction of the road to Consort Mine. 11/02/1998 (S.D. Williamson 623)

Senecio sp. aff. *S. anomalochrous* Hilliard

2530DB South Africa, Mpumalanga, Nelspruit District, Berlin State Forest, about 6 km east of Kaapsehoop, on same ridge as Kaapsehoop Asbestos mine. ON SERPENTINE. 27/04/1991 (K. Balkwill, M-J. Balkwill & S.D. Williamson 6441)

2530DB South Africa, Mpumalanga, Nelspruit District, Berlin State Forest, about 6 km east of Kaapsehoop, ridge opposite Kaapsehoop Asbestos mine. 23/01/1992 (K. Balkwill 6869)

Senecio sp. aff. *S. coronatus* (Thunb.) Harv.

2530DB South Africa, Mpumalanga, Nelspruit District, Berlin State Forest, about 6 km east of Kaapsehoop Ridge opposite Kaapsehoop Asbestos mine. 21/03/1991 (K. Balkwill, M-J. Balkwill & S.D. Williamson 6288)

2530DB South Africa, Mpumalanga, Nelspruit District, Berlin State Forest, about 6 km east of Kaapsehoop Ridge opposite Kaapsehoop Asbestos mine. At eastern end of ridge. 03/12/1991 (K. Balkwill, M-J. Balkwill & S.D. Williamson 6566)

2530DD South Africa, Mpumalanga, Carolina District, on the farm Groenvaly 701 JT. On serpentine. On firebreak adjacent to Morgenzon. NOT STOLZBURG SYNCLINE. 10/10/1997 (K. Balkwill, S. Williamson & S. Smith 9950)

Senagalia lotterii N.Hahn

2531CA South Africa, Mpumalanga, Barberton District, Barberton phase 2, Mundt's Concession. On SERPENTINE. 08/03/2003 (S.D. Williamson 954)

Appendix D – Levels of threat and recommended IUCN Red Data categories for endemic species

This appendix has been inserted as the Journal article:
Williamson, S.D. and Balkwill, K. (2006) Factors determining levels of threat to serpentine endemics. South Afican Journal of Botany 72: 619–626.

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Factors determining levels of threat to serpentine endemics

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Abstract

Twenty-nine species endemic to outcrops of serpentine soils of the Barberton Greenstone Belt have been identified. Of these only 11 were previously Red Data Listed, six as Insufficiently Known (K) or Data Deficient (DD). The populations of these endemics are extremely fragmented and many are threatened by afforestation. IUCN criteria were used to re-assess their conservation status. One taxon has been assessed as Critically Endangered, eight as Endangered, 12 as Vulnerable, three as Lower Risk and four as Data Deficient. Additional factors, crucial for setting conservation priorities, are identified.

Keywords: Serpentine; Endemics; Conservation status; IUCN categories

1. Introduction

Over the past 15 years extensive studies have been conducted on the serpentine vegetation in Mpumalanga, South Africa (Morrey et al., 1989, 1992; Williamson, 1994; Hologne, 1994; Balkwill et al., 1997; Williamson et al., 1997). The term 'serpentine' is often applied to ultramafic rocks in general, but in this study it is used in its strict sense and applied to rocks derived from serpentinite. There are about 80 serpentine outcrops in south-eastern Mpumalanga (Fig. 1). These form part of the Barberton Greenstone Belt, which is a triangular geological intrusion extending from Malelane in the east, to Badplaas and Barberton in the south and ending just west of Nelspruit. The outcrops vary in size from 0.1 km^2 to ca. 19 km^2 with a mean size of 2.6 km^2. Some are separated from other outcrops by up to 20 km (Balkwill et al., 1997). The outcrops occur in mountainous areas and are heterogeneous in altitude, slope, soil depth etc. The serpentine vegetation falls within the Mixed Lowveld Bushveld, Sour Lowveld Bushveld and North-eastern Mountain Grassland vegetation types (Low and Rebelo, 1996).

The vegetation of serpentine outcrops is often distinct from the surrounding vegetation, as a result of the presence of high concentrations of heavy metals such as nickel and chromium and high magnesium to calcium ratios (Roberts and Proctor, 1992). To date, 29 species endemic to serpentine soils of the Barberton Greenstone Belt have been identified. Van Wyk and Smith (2001) described the Barberton Centre of Plant Endemism and suggested that the serpentine vegetation contributes significantly to the total endemism and the total number of species of this region. Only nine of the serpentine endemic species were previously listed in the Red Data Lists (Hilton-Taylor, 1996; Victor, 2002), six of these as Insufficiently Known (K) or Data Deficient (DD).

Five endemic species are restricted to single sites and are therefore rare. Serpentine patches are effectively islands surrounded by soils derived from granite, shale and slate, in which serpentine endemics do not grow. Therefore the populations of these endemics are extremely fragmented. Fragmentation has been increased by extensive afforestation of the area. The Barberton Centre of Plant Endemism is the most transformed centre in Mpumalanga at 31% with 22% of its surface area under plantations (Lötter et al., 2002). Only about 6% of its serpentine sites are conserved (Balkwill and Balkwill, 1999) and many remaining sites are threatened by further afforestation and mining. Serpentine ecosystems worldwide are threatened or are being destroyed and therefore a “Resolution” highlighting their unique biology, was endorsed by delegates (including South African representatives) of the First International Conference on Serpentine Ecology. These delegates

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Abbreviations: IUCN, World conservation Union.

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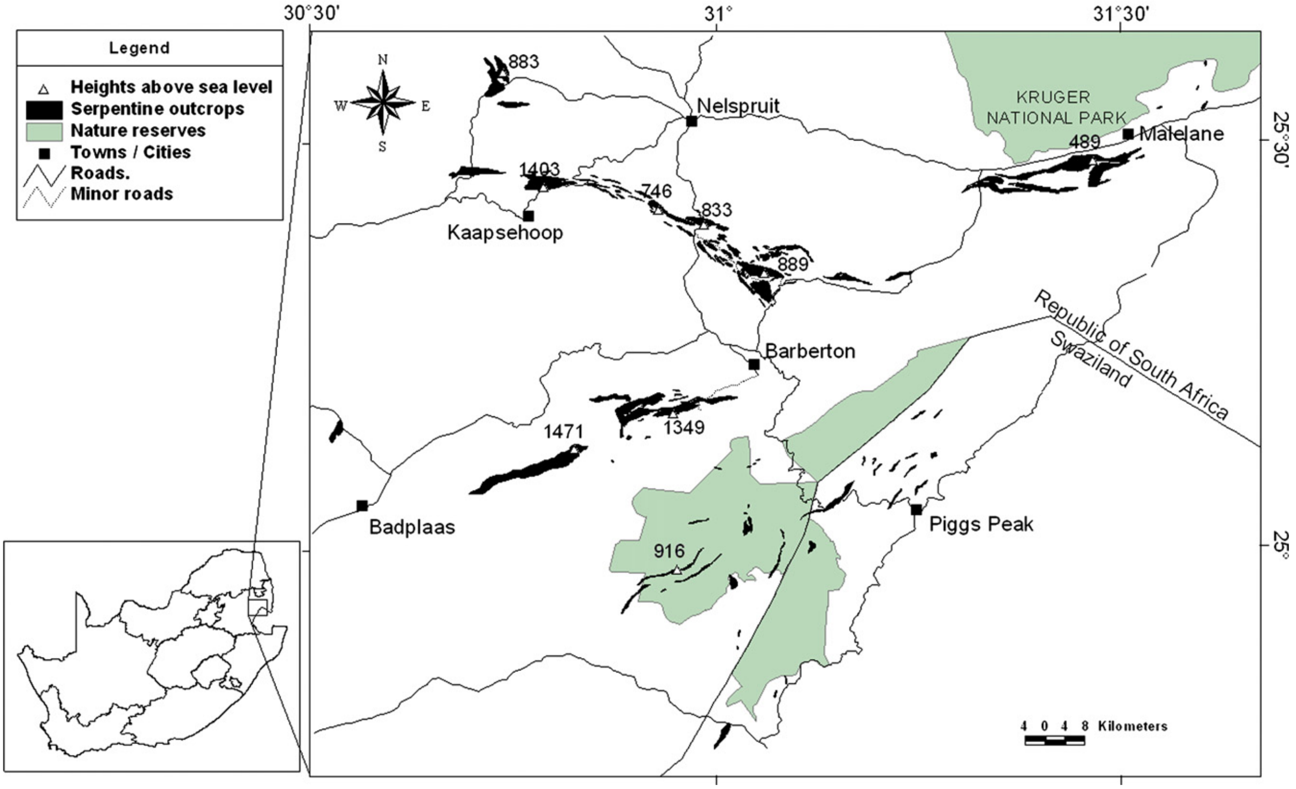

Fig. 1. Map indicating serpentine outcrops of the Barberton Greenstone Belt in Mpumalanga, South Africa.

resolved to support the conservation of the serpentine floras worldwide (Kruckeberg, 1992).

This paper is part of a more extensive study to assess the conservation status of the vegetation of serpentine outcrops in Mpumalanga. The assessment of extinction risk is pivotal in setting conservation priorities for threatened species, being one of many factors that contribute to the setting of priorities (Gärdenfors et al., 2002). The extinction risk of each serpentine endemic is presented and the contributory factors are highlighted.

The current IUCN (2001) criteria that are appropriate for the assessment of these endemics are outlined. Red Data categories are essential as they are used by conservation authorities to set clearly defined priorities for conservation of some areas (Lötter, pers. comm.[1]). However, Red Data Lists do not take into account whether or not species are utilized or whether they are high profile species (Golding, 2001).

Five endemic species have been found to hyperaccumulate nickel and may have uses in the biorecovery of nickel. Hyperaccumulating taxa (especially *Berkheya coddii* Roessl. and *Senecio* sp. aff. *S. coronatus*) are propagated worldwide to determine the mechanisms and evolution of nickel hyperaccumulation (Mesjasz-Przybylowicz et al., 2001a,b) and for phytoremediation and phytomining (Brooks et al., 2001; Angle et al., 2001). Should hyperaccumulation be considered when determining the conservation priorities of serpentine endemics? Does potential use give conservation authorities an additional responsibility to conserve particular species? In addition, taxa such as *B. coddii* and *Berkheya nivea* N.E.Br possess many of the features that characterise most invasive plants (Bromilow, 2001). Away from natural pathogens and competitors these plants have the potential of becoming exotic invaders of serpentine soils in other countries, thus, we question whether there is an additional responsibility to conserve this plant in its natural habitat as a potential source of biological control agents. Currently these factors are not taken into account when determining the conservation status of species using IUCN criteria but could have important implications for the conservation and management of serpentine areas.

2. Materials and methods

2.1. Data collection

Selected outcrops were visited regularly (from 1997 to 2001) to compile detailed checklists of the vegetation and to collect the data needed to assess the conservation status of each endemic. Information was also obtained from publications that report additional localities of some of the endemic species (Dyer, 1983; Balkwill and Balkwill, 1988; Edwards and Getliffe Norris, 1989; Phillips, 1917; Vollesen, 1991; Moffett, 1999). The type of data collected was determined by the definitions of population, subpopulation, extent of occurrence and area of occupancy, outlined in the IUCN documents (1994, 2001). For each

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[1] Mervyn Lötter—Mpumalanga Parks Board, Private Bag X11338, Nelspruit, 1200, South Africa.

Table 1
Current and recommended conservation status (according to new IUCN categories and criteria; IUCN, 2001z) of serpentine endemics

Recommended status taxon	Estimated population size	Estimated future decline (%)	Threats and potential threats	Extent of occurrence (EOO) (km^2)	Area of occupancy (AOO) (km^2)	Number of subpopulations	Recommended status	Current status in RDL
Critically endangered								
Brachystelma dyeri	50–100	?	Over collection and afforestation	14.5	3–4	1	CR C2a(ii)	Vu D2[b]
Endangered								
Athrixia sp. nov.	200–250	20–30	Afforestation	20–25	5–7	2–3	EN B1ab(iii,v)+2ab(iii,v);C1+2a(i)	Not listed
Brachystelma longifolium	100–150	10–15	Afforestation and mining	5–10	1–3	3–4	EN B1ab(v)+2ab(v),C1+2a(i); D	LR-lc[b]
Cyphia bolusii	100–150	3–7	Afforestation	15–20	3–5	1(2)	EN D[d]	DD[b]
Helichrysum sp. nov.	100–150	0	None known	2–5	1–2	1	EN D	Not listed
Inezia speciosa	200–250	10–20	Afforestation	15–20	2–4	2	EN B1ab(v)+2ab(v),C1+2a(i)[d]	VuD2[a]
Rhus pygmaea	100–500	5–10	Afforestation	38–40	4–6	5–6	EN D1+2	K[a]
Salpinctium hirsutum	200–250	4–5	Afforestation	15–20	2–3	2	EN C1[d]	K[a]
Senecio sp. aff. *S. anomalochrous*	150–250	5–10	Afforestation	14	50–80 m^2	1–2	EN D	Not listed
Vulnerable								
Aloe thorncroftii	1500–3000	10–20	Afforestation	30–35	5–7	4–7	VU B1ab(v)+2ab(v)D2	Vu D2[b]
Asystasia subbiflora	500–1000	5–10	Mining and afforestation	38–40	5–6	5–8	VU D1+2	Not listed
Berkheya nivea	2000–3000	0	Mining	17–20	4–5	4–5	VU D2	Not listed
Berkheya rehmannii var. *rogersiana*	800–1000	5–10	Afforestation	32–35	8–10	10–14	VU D1+2	Not listed
Gladiolus serpenticola	500–1000	5–8	Minor threat of afforestation	26–30	4–5	4–5	VU D1+2	Not listed
Gymnosporia sp. nov.	500–800	5–8	Mining claims	16–20	5–6	3–4	VU D1+2	Not listed
Indigofera crebra	700–1500	10–20	Afforestation	30–40	15–20	6–7	VU D1+2	Not listed
Ozoroa barbertonensis	500–1000	5–10	Afforestation	29–35	4–6	4–5	VU D1+2	DD[c]
Ozoroa sp. nov.	500–1000	5	Afforestation	20–25	3–4	3–4	VU D1	Not listed
Protea curvata	500–1000	5	Sasol pipeline and collection of flowers	16–20	4–5	4–5	VU D1+2	VU (D2)
Sclerochiton triacanthus	250–300	0–5	Mining claims and apparent failure to reproduce sexually	16–20	1–2	2–4	VU D1+2	DD[c]
Senecio sp. aff. *S. coronatus*	500–1000	5	Afforestation	26–30	3–5	4–5	VU D1+2	Not listed
Lower risk taxa								
Berkheya coddii	30000–50000	10–15	Afforestation and over–collection for research and biorecovery	44–45	20–25	10–15	LR-lc	DD[c]
Ocimum sp. nov. (Barberton species)	1000–2000	0	Minor threat of afforestation	27–30	10–15	9–10	LR-nt	Not listed
Sartidia sp. nov.	10000–20000	5–10	Afforestation	30–35	10–15	10–14	LR-lc	Threatened[c]

(continued on next page)

Table 1 (*continued*)

Recommended status taxon	Estimated population size	Estimated future decline (%)	Threats and potential threats	Extent of occurrence (EOO) (km^2)	Area of occupancy (AOO) (km^2)	Number of subpopulations	Recommended status	Current status in RDL
Data deficient taxa								
Berkheya sp. nov. aff. *B. seminivea*	?	?	?	?	?	?	DD	Not listed
Cheilanthes sp. nov.	?	?	?	?	?	?	DD	Not listed
Macledium zeyheri subsp. *thyrsiflorum*	?	?	?	?	?	?	DD	Not listed
Dicoma swazilandica	?	?	?	?	?	?	DD	Not listed

##Regional assessment (according to guidelines in Gärdenfors et al., 2002).

[a] (Hilton-Taylor, 1996).

[b] (Victor, 2002).

[c] (SABONET, 2003).

[d] National assessment (according to guidelines in Gärdenfors, 2001).

endemic considered the quantitative data collected included; size of subpopulations on each site, estimated number of mature individuals in the entire population and how many sites are inhabited by the endemic. In addition, note was taken of past events that could have affected these populations and any possible future threats in terms of mining, forestry, farming etc. The data used to assess each taxon are summarized in Table 1.

2.2. Taxon assessment

Each endemic species was assessed with the aid of RAMAS Red list version 2.0, which is recommended for assessors evaluating species for the IUCN Red List Program (Akçakaya and Ferson, 2001). RAMAS allows the user to specify a precautionary attitude or a more evidentiary attitude to risk. This software implements the IUCN threatened species criteria (IUCN, 2001) using all the data collected for each species and its habitat after allowing for explicit incorporation of uncertainties in the input data. Depending on these uncertainties, the resulting classification is often a single IUCN category, accompanied by a range of plausible categories.

3. Results and discussion

3.1. Evaluation of extinction risk (voucher specimens are listed in Appendix A)

Brachystelma dyeri K. and M. Balkwill occurs at only one locality and appears to be restricted to the amphibolite band that occurs in this area. The population is estimated to number less than 50 mature individuals. This taxon has been listed in the Red Data Book (Hilton-Taylor, 1996) as insufficiently known (K). However, this research has revealed that it should be listed as Critically Endangered (CR C2a (ii)), on the basis of its small population size and due to the threats of further afforestation in the area and of collectors of rare plant species.

Athrixia sp. nov. (Balkwill 9052) only occurs at two localities, one of which is extensively planted to pine. *Inezia speciosa* Brusse is considered a local endemic to serpentine as it has also been recorded from the Iron Crown area near Haenertsburg in Limpopo Province. In Mpumalanga this taxon is only known to occur at the same localities as *Athrixia* sp. nov. We estimate that afforestation caused at least a 20–30% decline in these populations. The populations of these taxa consist of less than 250 mature individuals and it is predicted that the presence of pine plantations will result in further decline in the size of the populations due to degradation of the habitat and shading. For these reasons it is recommended that *Athrixia* sp. nov. is listed as globally Endangered and *I. speciosa* as regionally Endangered (Table 1).

Brachystelma longifolium (Schltr.) N.E.Br. and *Rhus pygmaea* Moffett occur at three, possibly four localities, but the total area of occupancy of each taxon is less than 1 km^2 and there are only 50–100 mature individuals in the entire population of each species. One sub-population of *B. longifolium* is in an area planted to pine and another is very near a large mine. The largest sub-population of *R. pygmaea* occurs in a forestry area near a sawmill where there is also informal human habitation. *B. longifolium* is listed as Rare (Hilton-Taylor, 1996) using the old categories. Based on the new criteria and new information, it is recommended that *B. longifolium* and *R. pygmaea* be placed on the Red Data list as Endangered (Table 1).

Helichrysum sp. nov. and *Senecio* sp. aff. *S. anomalochrous* (Balkwill 6869) were assessed as Endangered (D) based on the small number of mature individuals (<150 and <250 respectively) that occur at only 1 location in each case. No threats have been recorded for *Helichrysum* sp. nov., but the population of *S.* sp. aff. *S. anomalochrous* is in an area undergoing extensive afforestation and is severely threatened by extensive planting of alien species of *Pinus* and *Eucalyptus*.

Aloe thorncroftii Pole Evans was listed as Vulnerable (Hilton-Taylor, 1996) and recently this taxon was reassessed as Least Concern because of the large numbers of mature individuals in each subpopulation (Smith et al., 2000). However, the area of occupancy of this species is about 5 km^2 at only four or five localities and a catastrophic event at any one of these localities could result in a significant decline of the population. In addition, these populations occur in areas of extensive afforestation and an increase of planting and environmental degradation associated with sylviculture threatens them. This threat includes altered burning regimes and a massive increase in weeds associated with the disturbance. It is recommended that this threat be considered in the assessment and therefore it is listed as Vulnerable (B1ab(v)+2ab(v) D2). Continued monitoring by Mpumalanga Parks Board (the statutory authority responsible for nature conservation in the province) will determine if there is any decline or increase in the size of the population, requiring a reassessment.

The individual areas of occupancy for the entire populations of *Asystasia subbiflora* C.B.Cl., *Gladiolus serpenticola* Goldblatt, *Gymnosporia* sp. nov. (Williamson 945), *Indigofera crebra* N. E.Br., *Ozoroa barbertonensis* Retief, *Ozoroa* sp. nov. (Balkwill 7840) and *Sclerochiton triacanthus* Meeuse are less than 2 km^2. Each taxon is known from two to five localities and their populations are estimated to consist of 500–1000 mature individuals. *S. triacanthus* has only been recorded at two localities and the largest population could be threatened by existing mining claims. Despite numerous attempts to collect flowering or fruiting material of this population, it has never been observed to be reproducing sexually. At present the populations of *S. triacanthus* are stable and this taxon has been assessed as Vulnerable (D1 + 2), but this could be reclassified as Endangered if further populations are not found and mining operations begin.

Most of the populations of *G. serpenticola* and *Gymnosporia* sp. nov. and a subpopulation of *A. subbiflora* and *O. barbertonensis* occur on land to be incorporated into the Barberton Nature Reserve. Many of the subpopulations of *A. subbiflora*, *I. crebra*, *O. barbertonensis* and *Ozoroa* sp. nov. could be threatened in the future by the forestry industry. Due to the small areas occupied by these taxa and the low numbers of mature individuals in these populations they have been assessed as Vulnerable (Table 1).

Berkheya rehmannii Thell. var. *rogersiana* Thell. and *Senecio* sp. aff. *S. coronatus* (Balkwill, Balkwill and Williamson 6566) were assessed as Vu D1 + 2 and *B. nivea* as Vu D2 based on small numbers of mature individuals (≤ 1000) and/or small area of occupancy (<20 km^2). The smallest subpopulation of *B. nivea* is in the Barberton Nature Reserve whereas the largest occurs on State land with existing mining claims, which will be incorporated into this reserve in the future (Stalmans, pers. comm.[2]). Many populations of *B. rehmannii* Thell. var. *rogersiana* Thell. and *Senecio* sp. aff. *S. coronatus* occur in areas undergoing extensive afforestation and are severely threatened.

Protea curvata N.E.Br was previously classified as Vulnerable (D2) (Hilton-Taylor, 1996). This classification suggests that there is little or no threat to this species. One of the largest populations occurs on State land, which is under the management of Mpumalanga Parks Board and will be incorporated into the Barberton Reserve in the future. The other populations are on private land, but development in these areas is unlikely due to their low agricultural and afforestation potential. Therefore the recommendation is that the classification of Vulnerable (D2) remains.

The extent of occurrence (EOO) of *B. coddii*, a hyperaccumulator of nickel, is less than 100 km^2 (Table 1) and thus applying a precautionary attitude would suggest this taxon be listed as threatened. However, category B cannot be applied further as there are more than 10 subpopulations and *B. coddii* can also not be categorized as threatened under criterion D due to the large number of individuals in each subpopulation (Table 1). In addition, at least half of the sites where *B. coddii* is found occur in conservation areas. Therefore the status of this taxon is Least Concern (LC). However, this status could change to threatened as some of the populations occur in areas undergoing afforestation.

Ocimum sp. nov. and *Sartidia* sp. nov. occur at more than 8 localities and at each of these sites large numbers have been found and many of these subpopulations occur within conservation areas such as the Songimvelo Game Reserve. It is recommended that these taxa are listed as Least Concern, however, their status could be changed to threatened if afforestation expands in areas outside the reserve boundaries.

Cyphia bolusii Phill. and *Salpinctium hirsutum* T.J.Edwards appear to be local endemics, as in Swaziland they occur off serpentine, but in Mpumalanga they have only been found on serpentine outcrops. *C. bolusii* only occurs at one locality in Mpumalanga and at this serpentine outcrop there has been some afforestation and a major road has been built through it. Both these events caused a decline in the numbers of this taxon, although the exact number cannot be determined. It is thought to only occur at one locality in Swaziland. This cannot be confirmed at present and thus it has been decided to assess this taxon on a national basis using the guidelines outlined by Gärdenfors (2001). The national status was thus determined to be Endangered (Table 1). *S. hirsutum* occurs at a minimum of two sites in Mpumalanga, with less than 250 mature individuals. Swaziland sub-populations could not be assessed and it was therefore also classified as nationally Endangered (Table 1).

An evaluation of *Berkheya* sp. nov. aff. *B. seminivea*, *Macledium zeyheri* (Sond.) S.Ortiz subsp. *thyrsiflorum* (Klatt) N.C. Netnou, *Dicoma swazilandica* S.Ortiz, Rodr. Oubiña and Pulgar and *Cheilanthes* sp. nov. is not possible at present due to insufficient data. Each taxon occurs in less than five localities, which would place each in a threatened category. Due to the lack of information these taxa should be listed as Data Deficient until they can be reassessed.

It is difficult to apply the IUCN criteria to serpentine endemics as in most cases the sub-populations are restricted to few outcrops, which results in extreme fragmentation. More than half of the serpentine endemics assessed here have less than five subpopulations and the rest have less than 16 subpopulations. In many instances subpopulations have large numbers of individuals (*B. coddii*, *B. nivea* and *Sartidia* sp. nov.) (Table 1), but areas of occupancy are very small. It is then difficult to make a decision based on these criteria. Due to this inconsistency when the data were entered into RAMAS, the outcome would be a list of plausible categories but a specific status with criteria could not be achieved at the recommended Burden of Proof (50%). Applying the threat data was also difficult, as these populations have not been studied long enough to determine by what percentage the populations have declined and the percentage future decline was thus predicted.

Categories A and E (describing the rate at which the populations are declining and probability of extinction) were not used in any of the assessments as no quantitative data were available for the taxa considered here. The distribution size and the

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[2] Mark Stalmans—International Conservation Services, P.O. Box 19139, Nelspruit, 1200.

numbers of mature individuals of each taxon, together with any threats recorded were used to calculate the extent of occurrence and/or the area of occupancy and to estimate any decline in distribution or population size. These data were used to apply Criteria B and/or C. Criterion D (describing very small or restricted populations) was the most appropriate criterion in most assessments, where there was little or no evidence of threats to the taxa.

In summary, seven taxa, which were insufficiently known (K) (Hilton-Taylor, 1996), have been reassessed and placed in appropriate categories (Table 1). Eighteen serpentine endemics have been assessed for the first time and 11 of these have been categorised as threatened (Table 1). Most of these are assessed as Vulnerable (37%) or Endangered (29%). Only one taxon is Critically Endangered, representing 3.7% of the taxa.

Subpopulations of serpentine endemics are severely fragmented and occur at few localities. Therefore they are vulnerable to single events, which could destroy large proportions of the population. These endemics also occur in areas that are largely planted to pine or they are threatened by mining operations. It is recommended that these taxa be placed on the Red Data List in the categories discussed so that conservation authorities and private land owners can be made aware of the value of serpentine outcrops and implement appropriate management actions.

Further assessment is needed to determine the levels of threat of the taxa that have been classified as Data Deficient. *M. zeyheri* subsp. *thyrsiflorum* is recorded from only one locality with a relatively small population and should be placed in a threatened category once data are available.

Additional factors not considered by the IUCN criteria, such as the extensive use of these plants for research and bioremediation and the potential of some serpentine endemics to become invasive weeds, need to be considered when conservation priorities and management plans for serpentine areas are compiled.

Acknowledgments

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Appendix A

Voucher specimens (precise localities have not been included to protect the endangered taxa from unscrupulous collectors of rare plants.)

Aloe thorncroftii Pole Evans

Carolina District, Stolzburg Syncline (2530DD), *Balkwill*, *Williamson and Smith 10075* (J); Barberton District, Barberton (2531CC), *Rogers 20250* (J)

Asystasia subbiflora C.B.Cl.

Barberton District (2530DD), *Balkwill and Cadman* 2590 (J); Barberton District, Queens River Valley (2530DD), *Balkwill and Cadman 2615* (J); Eerstehoek District, Songimvelo Game Reserve (2530DD), *Balkwill 7763* (J); Carolina District, Stolzburg Syncline (2530DD), *Balkwill*, *Williamson and Smith 9873* (J)

Athrixia sp. nov.

Carolina District, Stolzburg Syncline (2530DD), *Balkwill 9052* (J)

Berkheya coddii Roessl.

Carolina District, Stolzburg Syncline (2530DC), *Williamson 406* (J); Barberton District (2530DD), *Balkwill 6884* (J); Barberton District, Queens River Valley (2530DD), *Balkwill*, *Williamson and Smith 10219* (J); Eerstehoek District, Songimvelo Game Reserve (2530DD), *Balkwill*, *Balkwill and Williamson 6693* (J); Barberton District, Songimvelo Game Reserve (2531CC), *Stalmans 2203* (J); Barberton District, Songimvelo Game Reserve (2531CC), *Balkwill*, *Balkwill and Williamson 6646* (J); Kamhlushwa District, Kangwane–Nkomazi west region (2531DC), *Stalmans 2774* (J); Eerstehoek District, Songimvelo Nature Reserve (2630BB), *Stalmans 2359* (J)

Berkheya nivea N.E.Br.

Nelspruit District (2530DB), *Balkwill*, *Williamson and Smith 10016* (J); Barberton District (2531CA), *Stalmans 3005* (J); Swaziland, North West of Mbabane, Malolotja Game Reserve (2631AA), *Balkwill and Balkwill 9328* (J)

Berkheya rehmannii Thell. var. *rogersiana* Thell.

Nelspruit District, Berlin State Forest (2530DB), *Balkwill*, *Williamson and Smith 9786* (J); Barberton District (2530DD), *Williamson and Payet 279a* (J); Swaziland, North West of Mbabane, Malolotja Game Reserve (2631AA), *McCallum and Balkwill 210* (J)

Brachystelma dyeri K. and M. Balkwill

Barberton District (2530DD), *Williamson and Williamson 114* (J)

Brachystelma longifolium (Schltr.) N.E.Br.

Barberton District, Songimvelo Game Reserve (2531CC), *Ellery and Ellery 92/55* (J); Barberton District, Songimvelo Game Reserve (2531CC), *Balkwill and Balkwill 8559* (J); Eerstehoek District, Songimvelo Nature Reserve (2630BB), *Hartley 1469* (J)

Cyphia bolusii Phill.

Nelspruit District (2530DB), *Balkwill and Balkwill 3982* (J); Eerstehoek District, Songimvelo Game Reserve (2530DD), *Ellery and Ellery 92/120* (J)

Dicoma swazilandica S.Ortiz, Rodr. Oubiña and Pulgar

Eerstehoek District, Songimvelo Nature Reserve (2630BB), *Balkwill and Robinson 6831* (J)

Macledium zeyheri (Sond.) S.Ortiz subsp. *thyrsiflorum* (Klatt) N.C.Netnou

Barberton District, west of Malelane (2531CB) *Williamson and Williamson 263* (J)

Gladiolus serpenticola Goldblatt and Manning

Barberton District (2531CA), *Balkwill, Balkwill, Goyder, Paton and Williamson 10865* (J)

Indigofera crebra N.E.Br.

Barberton District, Queens River Valley (2530DD), *Balkwill, Balkwill and Williamson 6617a* (J); Carolina District (2530DD), *Balkwill, Williamson and Smith 9894* (J); Barberton District, Songimvelo Game Reserve (2531CC), *Germishuizen 5677* (J); Swaziland, North west of Mbabane, Malolotja Game Reserve (2632AA), *McCallum and Balkwill 396* (J); Swaziland, Mbabane District, (2631AC), *McCallum 381* (J)

Ocimum sp. near *O. obovatum* E. Mey. ex Benth.

Barberton District (2530DD), *Balkwill, Williamson and Smith 10208* (J); Eerstehoek District, Songimvelo Game Reserve (2530DD), *Balkwill 7765* (J); Barberton District, Songimvelo Game Reserve (2531CC), *Hartley 1291* (J); Barberton District, Songimvelo Game Reserve (2531CA), *Balkwill and Balkwill 8564* (J); Eerstehoek District, Songimvelo Nature Reserve (2630BB), *Balkwill, Balkwill, Stalmans, Goyder and Paton 10901* (J)

Ozoroa barbertonensis Retief

Barberton District (2530DD), *Balkwill and Balkwill 4001* (J)

Ozoroa sp. nov.

Eerstehoek District, Songimvelo Game Reserve (2530DD), *Stalmans 2856* (J); Barberton District, Songimvelo Game Reserve (2531CC), *Lee 166* (J); Eerstehoek District, Songimvelo Nature Reserve (2630BB), *Masilo E90* (J); Eerstehoek District, Songimvelo Game Reserve (2631AA), *Balkwill, Balkwill and Kidger 8096* (J)

Rhus pygmaea Moffett

Barberton District (2530DD), *Balkwill, Balkwill and Williamson 6596* (J); Barberton District (2531CA), *Williamson 623* (J)

Salpinctium hirsutum Edwards

Nelspruit District, Berlin State Forest (2530DB), *Balkwill, Balkwill and Williamson 6704* (J); Barberton District, Songimvelo Game Reserve (2531CC), *Balkwill, Balkwill and Williamson 6659* (J); Swaziland, Mbabane District (2631AC), *McCallum 445* (J)

Sartidia sp. nov.

Nelspruit District, Berlin State Forest (2530DB), *Balkwill, Balkwill and Williamson 6434* (J); Barberton District (2530DD), *Williamson, Balkwill and Balkwill 53* (J); Eerstehoek District, Songimvelo Game Reserve (2530DD), *Stalmans 2829* (J), Carolina District, Stolzburg Syncline (2530DD), *Balkwill 9043* (J); Barberton District, Songimvelo Game Reserve (2531CC), *Lee 126* (J); Eerstehoek District, Songimvelo Nature Reserve (2630BB), *Balkwill 9148* (J); Eerstehoek District, Songimvelo Game Reserve (2631AA), *Balkwill, Balkwill and Kidger 8097* (J)

Sclerochiton triacanthus Meeuse

Barberton District (2531CC), *Davidson s.n.* (J)

Senecio sp. aff. *S. anomalochrous* Hilliard

Nelspruit District, Berlin State Forest (2530DB), *Balkwill, Balkwill and Williamson 6441* (J)

Senecio sp. aff. *S. coronatus* Hilliard

Nelspruit District, Berlin State Forest (2530DB), *Balkwill, Balkwill and Williamson 6566* (J); Carolina District (2530DD), *Balkwill, Williamson and Smith 9950* (J)

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Appendix E

Matrices of species turnover determined for each pair of 1000 m^2 plots placed at each serpentine site and adjacent non-serpentine site, using Whittaker's β_W, Wilson and Shmida's β_T, Sorensen's C_S and Jaccard's C_J. Plots were placed at the base of hills (B), on south-facing slopes (S), north-facing slopes (N) and at the crest of hills (C).

Rosentuin (RT) serpentine

		C_S			
		B	S	N	C
C_J	B	-	0.372	0.387	0.359
	S	0.229	-	0.351	0.528
	N	0.240	0.213	-	0.352
	C	0.219	0.358	0.214	-

		β_W			
		B	S	N	C
β_T	B	-	0.628	0.613	0.641
	S	0.814	-	0.649	0.472
	N	0.806	0.824	-	0.648
	C	0.846	0.736	0.824	-

RT non-serpentine

		C_S			
		B	S	N	C
C_J	B	-	0.221	0.409	0.248
	S	0.124	-	0.467	0.496
	N	0.257	0.304	-	0.573
	C	0.142	0.330	0.402	-

		β_W			
		B	S	N	C
β_T	B	-	0.779	0.592	0.752
	S	0.890	-	0.533	0.504
	N	0.796	0.767	-	0.427
	C	0.876	0.752	0.713	-

Sawmill (SM) serpentine

		C_S			
		B	S	N	C
C_J	B	-	0.339	0.520	0.357
	S	0.203	-	0.595	0.554
	N	0.351	0.423	-	0.539
	C	0.217	0.383	0.368	-

		β_W			
		B	S	N	C
β_T	B	-	0.662	0.480	0.643
	S	0.831	-	0.405	0.446
	N	0.740	0.703	-	0.462
	C	0.821	0.723	0.731	-

SM non-serpentine

		C_S			
		B	S	N	C
C_J	B	-	0.411	0.566	0.354
	S	0.259	-	0.466	0.635
	N	0.394	0.305	-	0.433
	C	0.215	0.465	0.276	-

		β_W			
		B	S	N	C
β_T	B	-	0.589	0.434	0.646
	S	0.794	-	0.533	0.365
	N	0.717	0.766	-	0.567
	C	0.823	0.683	0.784	-

Groenvaly (GV) serpentine

		C_S			
		B	S	N	C
C_J	B	-	0.413	0.510	0.419
	S	0.260	-	0.483	0.465
	N	0.342	0.319	-	0.621
	C	0.419	0.465	0.621	-

		β_W			
		B	S	N	C
β_T	B	-	0.588	0.490	0.581
	S	0.794	-	0.517	0.535
	N	0.745	0.758	-	0.379
	C	0.797	0.709	0.752	-

GV non-serpentine

		C_S			
		B	S	N	C
C_J	B	-	0.511	0.478	0.416
	S	0.343	-	0.582	0.474
	N	0.314	0.410	-	0.483
	C	0.263	0.311	0.319	-

		β_W			
		B	S	N	C
β_T	B	-	0.489	0.522	0.584
	S	0.745	-	0.418	0.526
	N	0.761	0.703	-	0.517
	C	0.792	0.756	0.758	-

Magnesite (MM) serpentine

		C_S			
		B	S	N	C
C_J	B	-	0.453	0.470	0.497
	S	0.293	-	0.375	0.486
	N	0.307	0.231	-	0.403
	C	0.330	0.321	0.252	-

		β_W			
		B	S	N	C
β_T	B	-	0.547	0.530	0.503
	S	0.774	-	0.625	0.514
	N	0.765	0.813	-	0.597
	C	0.752	0.757	0.724	-

MM non-serpentine

		C_S			
		B	S	N	C
C_J	B	-	0.170	0.320	0.291
	S	0.093	-	0.245	0.224
	N	0.190	0.140	-	0.377
	C	0.170	0.126	0.232	-

		β_W			
		B	S	N	C
β_T	B	-	0.830	0.680	0.709
	S	0.915	-	0.755	0.776
	N	0.840	0.877	-	0.623
	C	0.855	0.888	0.811	-

Mundt’s (MC) serpentine

		C_S			
		B	S	N	C
C_J	B	-	0.297	0.248	0.435
	S	0.174	-	0.390	0.455
	N	0.142	0.242	-	0.376
	C	0.278	0.294	0.231	-

		β_W			
		B	S	N	C
β_T	B	-	0.703	0.752	0.565
	S	0.852	-	0.610	0.545
	N	0.876	0.805	-	0.624
	C	0.783	0.773	0.812	-

MC non-serpentine

		C_S			
		B	S	N	C
C_J	B	-	0.283	0.423	0.286
	S	0.165	-	0.368	0.258
	N	0.269	0.225	-	0.338
	C	0.167	0.148	0.203	-

		β_W			
		B	S	N	C
β_T	B	-	0.717	0.577	0.714
	S	0.858	-	0.632	0.742
	N	0.788	0.816	-	0.662
	C	0.857	0.871	0.831	-

Core Zone (CZ) serpentine

		C_S			
		B	S	N	C
C_J	B	-	0.605	0.587	0.611
	S	0.434	-	0.594	0.590
	N	0.416	0.422	-	0.571
	C	0.440	0.418	0.400	-

		β_W			
		B	S	N	C
β_T	B	-	0.395	0.413	0.389
	S	0.697	-	0.406	0.410
	N	0.706	0.703	-	0.429
	C	0.694	0.705	0.714	-

CZ non-serpentine

		C_S			
		B	S	N	C
C_J	B	-	0.488	0.395	0.398
	S	0.323	-	0.374	0.350
	N	0.246	0.230	-	0.486
	C	0.248	0.212	0.321	-

		β_W			
		B	S	N	C
β_T	B	-	0.512	0.605	0.602
	S	0.756	-	0.626	0.650
	N	0.802	0.813	-	0.514
	C	0.801	0.825	0.757	-

Kalkloof (KK) serpentine

		C_S			
		B	S	N	C
C_J	B	-	0.429	0.415	0.484
	S	0.273	-	0.466	0.529
	N	0.262	0.304	-	0.435
	C	0.319	0.359	0.278	0

		β_W			
		B	S	N	C
β_T	B	-	0.571	0.585	0.516
	S	0.785	-	0.534	0.471
	N	0.792	0.767	-	0.565
	C	0.764	0.736	0.782	-

KK non-serpentine

		C_S			
		B	S	N	C
C_J	B	-	0.471	0.474	0.431
	S	0.308	-	0.451	0.447
	N	0.310	0.291	-	0.318
	C	0.275	0.288	0.189	0

		β_W			
		B	S	N	C
β_T	B	-	0.529	0.526	0.569
	S	0.764	-	0.549	0.553
	N	0.770	0.768	-	0.682
	C	0.784	0.777	0.841	-

Appendix F

Keys to species names used in Figure 5.8 graphs and additional graphs for Chapter 5 showing the ranges of various climatic conditions in which serpentine-tolerant taxa are found.

Table F1: Key to taxon numbers for Graph showing the altitudes at which the plant taxa with apparent narrow altitude tolerances have been recorded (Figure 5.8)

Taxon number on graph	Taxon name
1	*Acacia caffra*
2	*Aerva leucura*
3	*Blepharis* sp
4	*Corbichonia decumbens*
5	*Digitaria eriantha*
6	*Elionurus muticus*
7	*Eragrostis superba*
8	*Euphorbia* sp.
9	*Hibiscus pusillus*
10	*Jatropha* sp.
11	*Senna italica* subsp. *arachoides*
12	*Tephrosia multijuga*
13	*Ximenia caffra*. var. *caffra*
14	*Bauhinia galpinii*
15	*Berchemia zeyheri*
16	*Combretum apiculatum* subsp. *apiculatum*
17	*Combretum zeyheri*
18	*Dichrostachys cinerea* subsp. *africana*
19	*Indigofera* sp. 1
20	*Ipomoea obscura* var. *obscura*
21	*Panicum maximum*
22	*Ziziphus mucronata* subsp. *mucronata*
23	*Coddia rudis*.
24	*Convolvulus farinosus*
25	*Sclerocarya birrea* subsp. *caffra*
26	*Searsia leptodictya*
27	*Peltophorum africanum*
28	*Sphedamnocarpus pruriens* subsp. *pruriens*
29	*Lepidagathis scabra*
30	*Xenostegia tridentata* subsp. *angustifolia*
31	*Corchorus confusus*
32	*Acacia davyii*
33	*Elephantorrhiza* sp.
34	*Faurea saligna*
35	*Lasiosiphon capitatus*
36	*Ocimum serratum*
37	*Polygala sphenoptera* Fresen. var. *sphenoptera*
38	*Searsia pentheri*
39	*Thesium* sp. near *T. gypsophiloides*
40	*Annona senegalensis* Pers. subsp. *senegalensis*
41	*Cheilanthes involuta* var. *obscura*
42	*Eragrostis chloromelas*
43	*Sporobolus sanguineus*
44	*Stenostelma corniculatum*
45	*Brachiaria brizantha*
46	*Brachystelma macropetalum*
47	*Clematis brachiata*.
48	*Cyphostemma humile* subsp. *dolichopus*

49 *Searsia pyroides* var. *gracilis*
50 *Rhoicissus tridentata* subsp. *cuneifolia*
51 *Hyperthelia dissoluta* (
52 *Faurea rochetiana*
53 *Hilliardiella aristata*
54 *Bowkeria cymosa*
55 *Ceropegia rendalii*
56 *Chironia purpurascens* ssp. *purpurascens*
57 *Diospyros lycioides* ssp. *nitens*
58 *Eulophia clavicornis*. var. *nutans*
59 *Gladiolus ecklonii*
60 *Hemizygia transvaalensis*
61 Heteromorpha involucrata
62 *Heteropyxis canescens*
63 Hypoxis iridifolia
64 *Kalanchoe paniculata*
65 *Lithospermum officinale*
66 *Plectranthus verticillatus*
67 *Elephantorrhiza elephantina*
68 *Monocymbium ceresiformi*
69 *Pearsonia aristata*
70 *Sisyranthus randii*
71 *Xerophyta villosa*
72 *Hibiscus trionum*
73 *Geigeria burkei* subsp. *burkei* var. *burkei*
74 *Berkheya coddii*
75 *Hypericum aethiopicum*. subsp. *sonderi*
76 *Alepidea setifera*
77 *Argyrolobium robustum*
78 *Berkheya rehmanii* var. *rogersiana*
79 *Digitaria diagonalis*. var. *diagonalis*
80 *Pimpinella transvaalensis*
81 *Cyphia stenophylla*.
82 *Psammotropha myriantha*
83 *Thunbergia galpinii*
84 *Aster bakerianus*
85 *Inezia integrifolia*
86 *Schistostephium heptalobum*
87 *Eriospermum flagelliforme*
88 *Linum thunbergii*
89 *Alepidea peduncularis*
90 *Felicia muricata* subsp. *muricata*
91 *Anthospermum herbaceum*
92 *Berkheya* sp. nov.
93 *Eragrostis tenuifolia*
94 *Eriosema* sp.
95 *Helichrysum kraussii*
96 *Helichrysum* sp 3
97 *Pseudarthria hookeri*
98 *Syncolostemon* sp.
99 *Thesium* sp. 1
100 *Gazania krebsiana* subsp. *serrulata*
101 *Gerbera piloselloides*
102 *Leobordea eriantha*
103 *Argyrolobium wilmsii*
104 *Aspidoglossum araneiferum*
105 *Commelina africana* var. *krebsiana*
106 *Jamesbrittenia burkeana*
107 *Searsia dentata*
108 *Searsia rehmanniana* var. *rehmanniana*
109 *Senecio lydenburgensis*
110 *Stachys nigricans*
111 **Crepis hypochoeridea*
112 *Hermannia depressa*
113 *Hypoxis multiceps*
114 *Launaea rarifolia* var. *rarifolia*
115 *Microchloa caffra*
116 *Ocimum obovatum* subsp. *obovatum* var. *obovatum*

117	*Senecio glanduloso-pilosus*
118	*Striga elegans*
119	*Thunbergia natalensis*
120	*Acalypha* sp. near *A. wilmsii*
121	*Ipomoea ommanneyi*
122	*Euryops laxus*
123	*Lippia wilmsii*
124	*Searsia discolor*
125	*Chaetacanthus* sp. nov.
126	*Crassula vaginata* subsp. *vaginata*
127	*Graderia subintegra*
128	*Helichrysum pallidum*
129	*Senecio scitus*
130	*Tricalysia lanceolata*

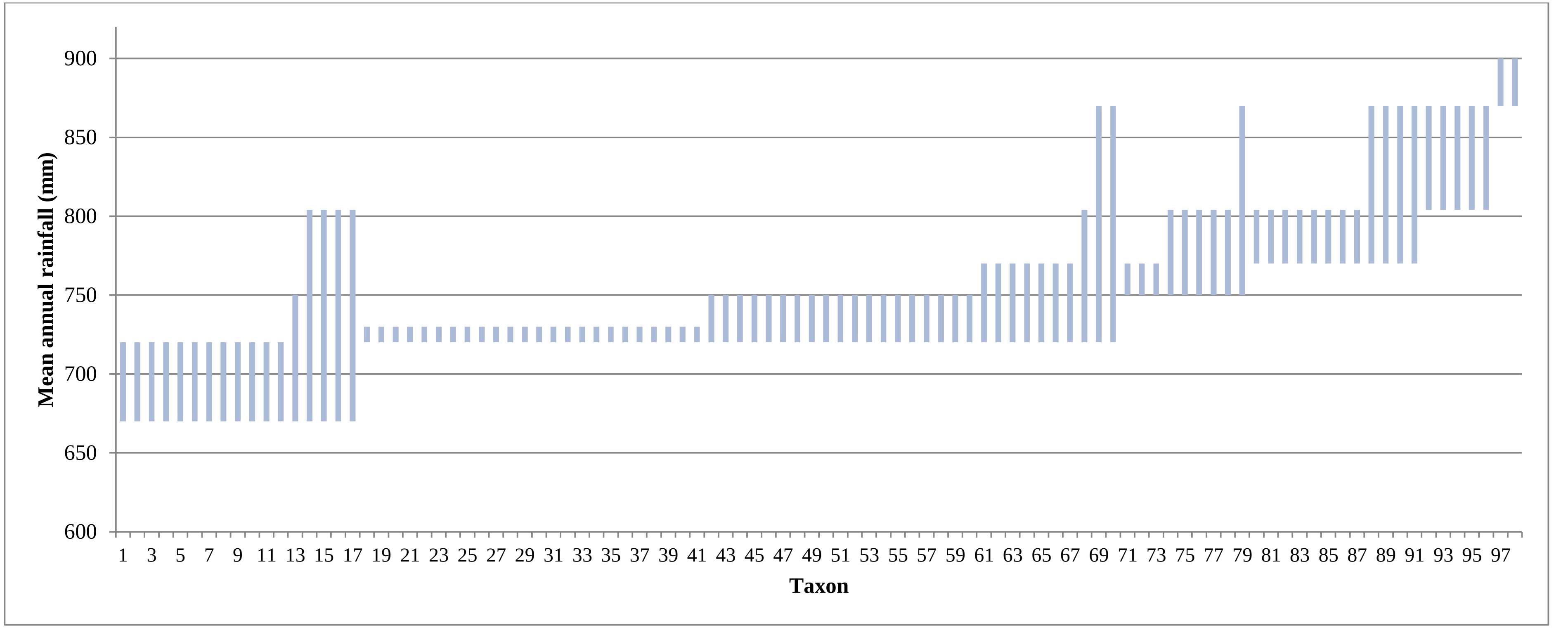

Figure F1: Graph showing the rainfall levels at which the plant taxa with apparent narrow rainfall tolerances have been recorded. The numbered plant taxa are listed in Table F2.

Table F2: Key to taxon numbers for Graph showing the rainfalls at which the plant taxa with apparent narrow rainfall tolerances have been recorded (Figure F1)

Taxon number on graph	Taxon name
1	*Acalypha punctata*. var. *punctata*
2	*Agelanthus natalitius* subsp. *zeyheri*
3	*Asparagus* sp. 2
4	*Chlorophytum angulicaule*
5	*Combretum hereroense* subsp. *hereroense*
6	*Hyparrhenia hirta*
7	*Merwilla plumbea*
8	*Rhynchosia nitens*
9	*Scleria* sp. 1
10	*Searsia pygmaea*
11	*Syncolostemon persimilis*
12	*Tricholaena monachme*
13	*Striga asiatica*
14	*Diheteropogon amplectens*
15	*Rhynchosia totta* var. *totta*
16	*Trachypogon spicatus*
17	*Tristachya leucothrix*
18	*Acalypha glandulifolia*
19	*Argyrolobium rupestre*
20	*Aristida* sp.
21	*Asparagus laricinus*
22	*Aster lydenburgensis*
23	*Asystasia subbiflora*
24	*Carissa bispinosa*
25	*Chlorophytum cooperi*
26	*Cissus cornifolia*
27	*Cyanotis speciosa*
28	*Eriosema ellipticifolium*
29	*Eulophia speciosa*
30	*Euphorbia schinzii*
31	*Helichrysum nudifolium* var. *pilosellum*
32	*Heteromorpha pubescens*
33	*Indigofera crebra*
34	*Lactuca* sp.
35	*Ledebouria revoluta*
36	*Phyllanthus glaucophyllus*
37	*Polygala leendertziae*
38	*Senecio bupleuroides*
39	*Senecio glaberrimus*
40	*Sonchus dregeanus*
41	*Thesium* sp. 2
42	*Acalypha angustata*
43	*Aristida transvaalensis*
44	*Asparagus setaceus*
45	*Bowkeria cymosa*
46	*Ceropegia rendalii*
47	*Chironia purpurascens* ssp. *purpurascens*
48	*Diospyros lycioides* ssp. *nitens*
49	*Eulophia clavicornis* var. *nutans*
50	*Gladiolus ecklonii*
51	*Gnidia gymnostachya*
52	*Hemizygia transvaalensis*
53	*Heteromorpha involucrata*
54	*Heteropyxis canescens*
55	*Hypoxis iridifolia*
56	*Kalanchoe paniculata*
57	*Lithospermum officinale*
58	*Plectranthus verticillatus*
59	*Protea gaguedi*
60	*Trachyandra reflexipilosa*
61	*Basananthe sandersonii*
62	*Berkheya insignis*

63	*Cyperus obtusiflorus* var. *obtusiflorus*
64	*Cyphia elata*
65	*Eragrostis capensis*
66	*Pelargonium luridum*
67	*Ruellia stenophylla*
68	*Hibiscus trionum*
69	*Faurea rochetiana*
70	*Hilliardiella aristata*
71	*Chlorophytum fasciculatum*
72	*Erica drakensbergensis*
73	*Rhynchosia monophylla*
74	*Jatropha latifolia* var. *angustata*
75	*Ocimum* sp. nov.
76	*Pachystigma latifolium*
77	*Polygala serpentaria*.
78	*Senecio latifolius*
79	*Hyperthelia dissoluta*
80	*Berkheya* sp. nov.
81	*Eragrostis tenuifolia*.
82	*Eriosema* sp.
83	*Helichrysum kraussii*
84	*Helichrysum* sp 3
85	*Pseudarthria hookeri*
86	*Syncolostemon* sp.
87	*Thesium* sp. 1
88	*Eragrostis* sp. 2
89	*Euphorbia gueinzii* Boiss. var. *gueinzii*
90	*Lannea edulis* var. edulis
91	*Paspalum scrobiculatum*
92	*Aloe* sp. 1
93	*Asparagus* sp. 1
94	*Hermannia* sp. near *H. montana*
95	*Lippia* sp.
96	*Pearsonia* sp.
97	*Rotheca myricoides*
98	*Vigna vexillata* var. *vexillata*

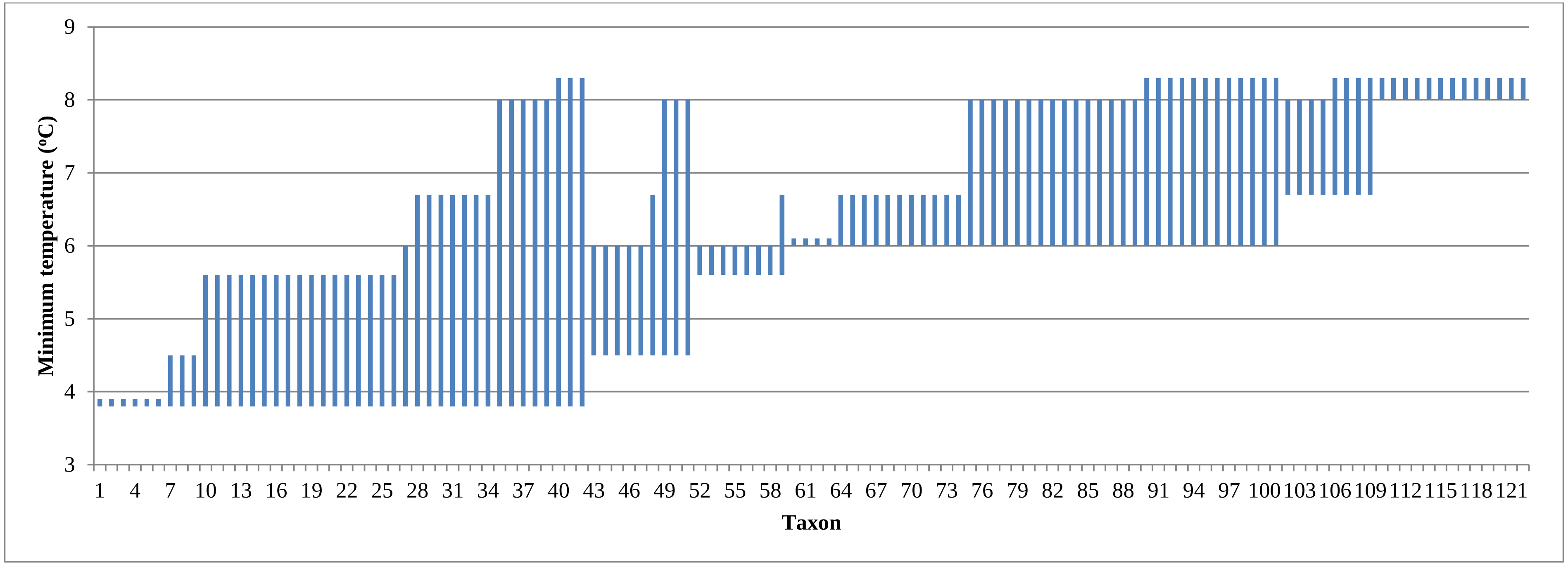

Figure F2: Graph showing the minimum temperatures at which the plant taxa with apparent narrow temperature tolerances have been recorded. The numbered plant taxa are listed in Table F3.

Table F3: Key to taxon numbers for Graph showing the minimum temperature at which the plant taxa with apparent narrow temperature tolerances have been recorded (Figure F2)

Taxon number on graph	Taxon name
1	*Aloe* sp. 2
2	*Aster bakerianus*
3	*Cyphia stenophylla*
4	*Hypoxis rigidula* var. *rigidula*
5	*Psammotropha myriantha* Sond.
6	*Senecio* sp. near *S. latifolius*
7	*Eriospermum flagelliforme*
8	*Linum thunbergii*
9	*Thunbergia galpinii*
10	*Aloe arborescens*
11	*Aristea woodii*
12	*Ocimum* sp. near *O. obovatum*
13	*Berkheya echinacea*
14	*Cheillanthes viridis* var. *glauca*
15	*Diheteropogon filifolium*
16	*Ekebergia pterophylla*
17	*Hermannia antonii*
18	*Leobordea pulchra*
19	*Pachystigma macrocalyx*
20	*Polygala wilmsii*)
21	*Rapanea melanophloeos*
22	*Raphionacme hirsuta*
23	*Scilla natalensis*
24	*Sebaea leiostyla*
25	*Senecio macrocephalus*
26	*Senecio sp. bupleurioides*
27	*Hibiscus trionum*
28	*Acalypha peduncularis*
29	*Argyrolobium robustum*
30	*Digitaria diagonalis* var. *diagonalis*
31	*Dipcadi viride*
32	*Hilliardiella aristata*
33	*Pimpinella transvaalensis*
34	*Scleria bulbifera*
35	*Athrixia phylicoides*
36	*Callilepis laureola*
37	*Cheilanthes hirta* var. *hirta*
38	*Nidorella auriculata*
39	*Phyllanthus maderaspatensis*
40	*Barleria ovata*
41	*Diheteropogon amplectens*
42	*Leobordea carinata*
43	*Jatropha latifolia* var. *angustata*
44	*Ocimum* sp. nov.
45	*Pachystigma latifolium*
46	*Polygala serpentaria*
47	*Senecio latifolius*
48	*Wahlenbergia undulata*
49	*Convolvulus sagittatus*
50	*Euclea crispa* subsp. *crispa*
51	*Zornia capensis* subsp. *capensis*
52	*Cheilanthes involuta* var. *involuta*
53	*Cyperus obtusiflorus* var. *sphaerocephalus*
54	*Dais cotinifolia*
55	*Dicoma zeyheri* ssp. *zeyheri*
56	*Loudetia flavida*
57	*Rotheca myricoides*
58	*Vigna vexillata* var. *vexillata*
59	*Andropogon shirensis*
60	*Brachystelma macropetalum*
61	*Clematis brachiata*
62	*Cyphostemma humile* subsp. *dolichopus*
63	*Searsia pyroides* var. *gracilis*

64	*Acalypha wilmsii*
65	*Asclepias aurea*
66	*Berkheya setifera*
67	*Bulbostylis oritrephes*
68	*Dalbergia armata* E.Mey. (
69	*Dicoma swazilandica* S.Ortiz
70	*Eriosema distinctum*
71	*Hypoxis* sp. near *H. acuminata* & *H. rigidula*
72	*Imperata cylindrica*
73	*Pegolettia lanceolata*
74	*Wahlenbergia virgata*
75	*Acacia davyii*
76	*Annona senegalensis* subsp. *senegalensis*
77	*Cheilanthes involuta* var. *obscura*
78	*Combretum molle*
79	*Corchorus asplenifolius*
80	*Elephantorrhiza* sp.
81	*Faurea saligna*
82	*Geigeria burkei* subsp. *burkei* var. *hirtella*
83	*Graderia scabra*
84	*Hypoxis rigidula* var. *pilosissima*
85	*Lasiosiphon capitatus*
86	*Ocimum serratum*
87	*Polygala sphenoptera* var. *sphenoptera*
88	*Searsia pentheri*
89	*Thesium* sp. near *T. gypsophiloides*
90	*Bauhinia galpinii*
91	*Berchemia zeyheri*
92	*Combretum apiculatum* subsp. *apiculatum*
94	*Combretum zeyheri*
95	*Dichrostachys cinerea* subsp. *africana*
96	*Gerbera jamesonii*
97	*Indigofera* sp. 1
98	*Ipomoea obscura*
99	*Panicum maximum*
100	*Peltophorum africanum* Sond.
101	*Sphedamnocarpus pruriens* subsp. *pruriens*
102	*Ziziphus mucronata* subsp. *mucronata*
103	*Acalypha punctata* var. *punctata*
104	*Agelanthus natalitius* subsp. *zeyheri*
105	*Chlorophytum angulicaule*
106	*Hyparrhenia hirta*
107	*Asparagus laricinus*
108	*Cissus cornifolia*
109	*Eulophia speciosa*
110	*Euphorbia schinzii*
111	*Acacia caffra*
112	*Aerva leucura*
113	*Blepharis* sp
114	*Corbichonia decumbens*
115	*Digitaria eriantha* Steud.
116	*Elionurus muticus*
117	*Eragrostis superba*
118	*Euphorbia* sp.
119	*Hibiscus pusillus*
120	*Jatropha* sp.
121	*Senna italica* subsp. *arachoides*
122	*Tephrosia multijuga*
123	*Ximenia caffra* var. *caffra*