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A standard method for predicting conservation status from herbarium specimens in the context of a new phylogeny and taxonomy of Loudetia

Kamundi, Dickson Athanasius

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CHAPTER 5

A new method for predicting risk using herbarium data

5.0 Abstract

Species exhibiting widespread spatial distributions and / or many different locations and habitat types are considered to have a greater chance of surviving localized and habitat-specific environmental disturbances and therefore a lower risk of extinction than restricted ones. In addition, the number of herbarium specimens is assumed to indicate relative abundance and the presence of recent collecting records may indicate persistence of a species in the wild. Therefore gleaning information on the number of collecting locations, relative abundance and habitat types from herbarium specimen labels has enabled the risk assessments of especially well-collected species to be undertaken in the absence of field-based data. However, there are disparities between herbarium records and patterns of species distributions, because current changes in the number of locations, range sizes and abundance levels are very rarely captured in herbarium specimens. Rare species may be well-represented and vice versa, leading to inaccurate assessments. There was therefore a need to determine if herbarium records could be useful only in predicting which species require detailed conservation status assessments – thus restricting the often expensive and time consuming field surveys to species that have already been prioritized. A method for predicting which species might be threatened was therefore developed using 2308 herbarium specimens of *Loudetia.* Locality information obtained from specimen labels was plotted on maps to provide indications of the number of locations and habitat fragmentations. The number of habitat types, also obtained from collector notes, supplemented this information. Species not collected over a period of 3 decades formed a benchmark for the apprehension that they might have gone locally extinct or their population or range sizes have diminished to the extent that collectors may easily overlook them. Dates of specimen collections, the number of locations, habitat fragmentation and habitat types were then analysed to identify classes of risk indicators. Four groups were identified and assigned conservation priorities, first, second, third and Low Priorities. Species falling within the High and Intermediate Priority categories require detailed risk assessments, whereas those within the Low Priority category do not. These risk indicators were used to predict the conservation status of species of *Syncolostemon* (including *Hemizygia*). Comparison showed that species assigned first to third priorities using the risk predicting method were also considered to be endangered or vulnerable by the IUCN SSC (2001) method based on field data. On the other hand, the IUCN SSC (2001) method showed that some species assigned to the Low Priority category are threatened. Therefore the risk indicators were adjusted to accommodate as many endangered species as possible in the priority list. This implies that the proposed exclusion of non-priority species from detailed assessments can be undertaken while capturing threatened species. In this way, funding can be more effectively employed by dedicating time and money for detailed conservation status assessments to species that have been prioritized using herbarium records.

5.1 Introduction

5.1.1 *Knowledge about the conservation status of species*

Loss, reduction in size, fragmentation and modification of habitats have been linked to an increase in the extinction risk of species, placing an urgent need for increased protection for endangered or rare species (Burkey, 1989; Turner *et al.,* 1989; Pimm *et al.,* 1995; MacDougall *et al.,* 1998; Mazia *et al.,* 2001; Achard *et al.,* 2002; Sachs, 2002; Tickell, 2002; Willis *et al.,* 2003; Victor & Keith, 2004). The development of effective conservation strategies almost always requires knowledge about plant species that have become threatened due to prevailing environmental stresses. The increased rate of extinction therefore imposes pressing demands for the conservation status assessments of species (MacDougall *et al.,* 1998; Willis *et al.,* 2003). However, only about 2.3% of the estimated 300,000 plant species had been assessed by the year 2000 (Willis *et al.,* 2003). The conservation status of species can be determined through detailed field surveys to obtain quantitative data about the ecological aspects and changes in the range of species due to habitat destruction, which may cause population reductions. Field surveys require much time and consequently the progress in conservation status assessments has been slow, raising concern for conservation managers.

5.1.2 *Status quo*

Herbarium records have been recommended and used to assess species for the conservation status and rarity (Prance, 1984; MacDougall *et al.,* 1998; Prance *et al.,* 2000; Golding, 2000; Schatz, 2002; Willis *et al.,* 2003; Victor & Keith, 2004). By using herbarium records, MacDougall *et al.* (1998) compiled a list of rare and threatened species based on which recommendations were made for increased protection under the Federal Law of the United States of America. More recently, Golding (2002) provided the estimation of the risk of extinction for the southern Africa grass species and Willis *et al.* (2003) assessed the well-collected species of *Plectranthus* based largely on herbarium records. The apparent success of risk and rarity assessments coupled with the ready availability of voucher specimens encouraged workers to recommend, demonstrate and sometimes even employ herbarium records as a quick and cheap alternative source of information for conservation status assessments especially when these are the best available data (Prance, 1984; MacDougall *et al.,* 1998; Prance *et al.,* 2000; Schatz, 2002; Willis *et al.,* 2003). Despite this promising use, shortfalls have been noted, the major one being the disproportional representation of abundances and spatial distributions in the wild by herbarium collections, particularly in Africa, Asia and South America where collections are still not comprehensive for most species (White, 1983; Prance, 1984; Rhoads & Thompson, 1992; MacDougall *et al.,* 1998; Prance *et al.*, 2000; Purvis *et al.,* 2000; Donoghue & Alverson, 2000; Schatz, 2002; Willis *et al.,* 2003). For example, *Hirtella tocantina* Ducke (Chrysobalanaceae) was represented by 3 specimens in 1972, but it turned out to be the most common species on a 3 ha survey plot, albeit its restriction to a small area in the Xingu and Tocantins river basins of Pará, Brazil (Prance, 1984). *Anthodiscus amazonica* Ducke (Cariocaraceae) was previously only known from the Guianas, western Amazonia and the Chocó in Colombia, but it was later documented in the forests of Bahia (Prance *et al.,* 2000). The large population of *Loudetia camerunensis* encountered during the recent field survey at Lajuma Plateau, Soutpansberg, South Africa has apparently not been represented in herbaria before. These 3 examples serve to

indicate that specimens housed in herbaria may be a poor indicator of abundance estimates and the number of localities because many areas and / or species are under-collected. Therefore, because of the disparity between collecting records and abundance and / or the dispersion of species, results of risk assessments based on herbarium data are often likely to be misrepresentations of the situation in the field (Rhoads & Thompson, 1992).

The widely used method for assessing the conservation status of species employs the reduction in population and range sizes over the past 10 years or 3 generations, severe habitat fragmentation, estimated population size and a quantified probability of extinction (Appendix 5.1). This method requires knowledge about the biology of species and quantitative data, including area of occupancy, extent of occurrence, number of mature individuals and reduction in range and population sizes over time (White, 1983; IUCN SSC, 2001; Willis *et al.,* 2003). Such data are not only scanty in literature but also not available in herbarium data. Due to the scarcity of field-based species-specific data, information is gleaned from herbarium specimen labels. Commonly used herbarium data include range sizes, number of locations, range fragmentations, age distribution of collecting efforts, and habitat affinities (Prance, 1984; Rhoads & Thompson, 1992; MacDougall *et al.,* 1998; Purvis *et al.,* 2000; Willis *et al.,* 2003).

5.1.3 *Range properties and habitat fragmentations*

Plotting point distribution maps to depict collecting localities based on herbarium labels reveals patterns of past and present geographical distributions of species (Rhoads & Thompson, 1992; Stern & Eriksson, 1996; Willis *et al.,* 2003). On fine scale maps, including quarter degree square plots, clusters of contiguous points may indicate the existence of a single interbreeding population of a species whereas a disjunction of points would be interpretable as independent populations (Schatz, 2002). Therefore the number of populations of species may be inferred by using herbarium records (Willis *et al.* 2003). A systematic method of estimating the area of occupancy (AOO) and extent of occurrence (EOO) from herbarium records using geographical information systems (GIS) has been suggested. Using at least 3 non-linear points obtained from locality records, a polygon is generated on a map and measured for the AOO or EOO (Willis *et al.,* 2003). An alternative method involves counting contiguous occupied cells. If collecting localities plotted on a map cover few contiguous cells, a species exhibit restricted spatial distribution whereas if points are spread over several grids; a species is widely dispersed (Schatz, 2002). The pattern of distribution may therefore allow the estimation of the size of locations. Species restricted to a single habitat type may be at greater risk of extinction than those which are adapted to more than one habitat type (Figure 5.1), particularly if the habitat is small, modified and has been or is being converted to inhospitable forms of land use (MacDougall *et al.,* 1998). The risk of local extinction due to the demographic stochasticity, natural catastrophe and / or habitat destruction is believed to be higher in spatially restricted than in extensive populations (Figure 5.1) because a widely dispersed species has a chance of surviving elsewhere in the event of a habitat-specific disturbance (Pimm *et al.,* 1988; Purvis *et al.*, 2000). Small populations may also be prone to inbreeding depression and the edge effect, which are perceived to drive species to extinction (Burkey, 1989; Pimm *et al.,* 1995). More importantly, information on suitable

habitat types may indicate ecological restrictions of species to particular environments (Stern & Eriksson, 1996; MacDougall *et al.,* 1998). The devastation of restricted habitats may lead to the lack or reduction of subsequent collections (Prance, 1984).

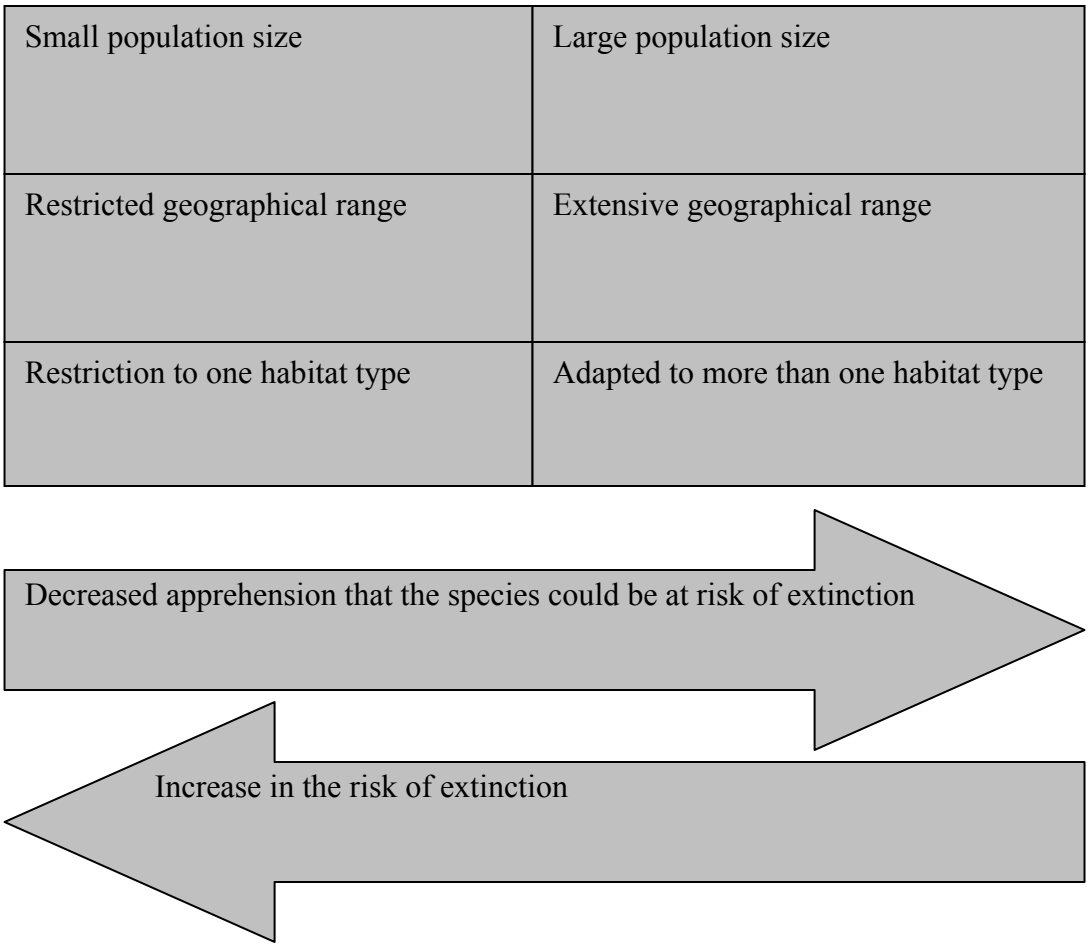


Figure 5.1. General considerations about the risk of species extinction based on population and range sizes and the number of habitat types.

5.1.4 *Temporal distributions*

The age of herbarium records may provide patterns of past and present geographical distributions of species (Stern & Eriksson, 1996). Recently collected specimens indicate the perpetual existence of species in particular localities (MacDougall *et al.,* 1998; Willis *et al.,* 2003). Collection dates have therefore been used to make inferences about the persistence of species in given localities (MacDougall *et al.,* 1998). For example, species not recorded for the past 35 years when collection frequencies were high at Brunswick in Canada have been perceived to have become locally extirpated (MacDougall *et al.,* 1998). A similar conclusion has been made for other species, including in the genus *Orbea* Haw. (Bruyns, 2002). A reduction in the number of recent specimen collections may not only signal a decline in the population size, but also shrinkage in range size.

Therefore an assumption that small and decreasing numbers of specimens may signal the probability that species have become or are becoming threatened has been made (Figure 5.2).

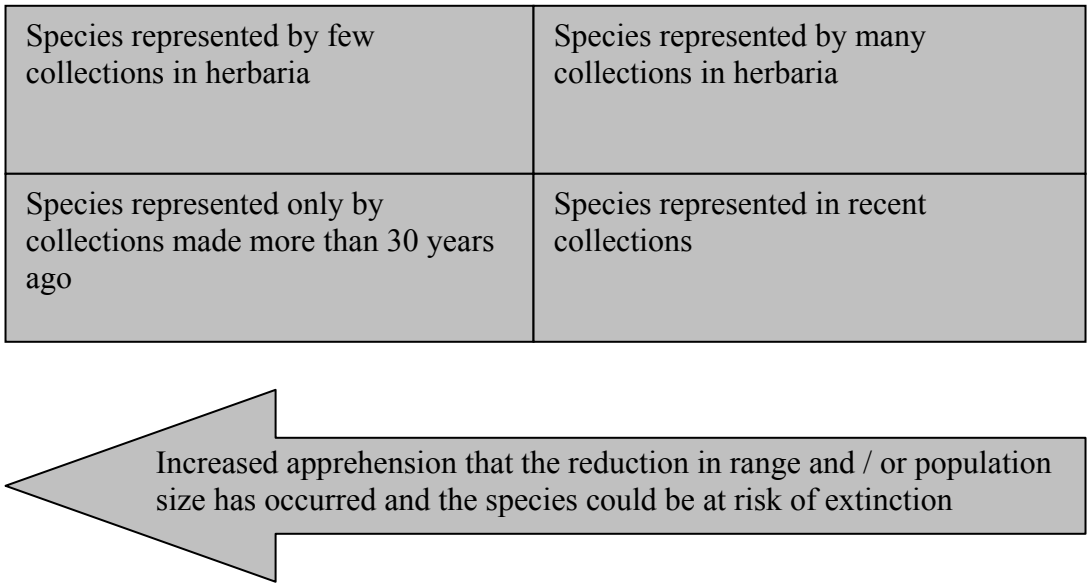


Figure 5.2. Assumption of risk status based on the number and age of herbarium collections.

5.1.5 *Indicators of conservation status*

Species represented by <5, 5-10 and ≥11 herbarium specimens have been classified as very rare, rare and uncommon, respectively (MacDougall *et al.*, 1998). However, MacDougall *et al.* (1998) did not specify the boundary between uncommon and common as the category "≥11 herbarium specimens" could include species that are common. A combination of AOO, EOO, and number of habitats and extent of fragmentation has been used to classify rare species. For example, Critically Rare and Rare species have been defined as having AOO <10 km² or EOO <100 km² and known from only one location or severely fragmented and AOO <500 km² or EOO <5000 km² and no more than 5 locations and is severely fragmented, respectively (Victor & Keith, 2004). The IUCN SSC (2001) method requires quantitative data (Appendix 5.1) that are not available in the largely qualitative herbarium records; hence this method cannot be used with certainty for processing herbarium records. Thus, various classification schemes have been developed to depict different levels of threat or rarity based on herbarium or field data.

5.1.6 *Shortfalls of herbarium data*

Herbarium specimens therefore provide ready data that can be processed to provide estimations of the number of locations, population sizes, habitat fragmentations, EOO, AOO, species-habitat affinities and rarity ratings, each independently or synergistically having a bearing on species extinction risks. However, data gleaned from herbarium specimen labels exhibit limitations. The accumulation of specimens is dependent on the

availability and interest of botanists in the area, accessibility of collecting localities, the nature of species, specimen storage policies and political factors, making herbarium records unsystematic (Prance, 1984; MacDougall *et al.,* 1998; Donoghue & Alverson, 2000; Prance *et al.,* 2000). For example, common species may be better represented than rare ones or vice versa and collections tend to be biased towards settlements and readily accessible locations (Prance, 1984; MacDougall *et al.,* 1998; Willis *et al.,* 2003). While widespread and common species may be well represented in herbaria, collectors may repeatedly ignore them in favour of rare and restricted ones (Prance, 1984; Stern & Eriksson, 1996). Botanists may concentrate on hotspots and flagship species, which may be well collected (Prance, 1984; Willis *et al.,* 2003). Thus, species rare in herbaria may not necessarily be rare in the wild (Stern & Eriksson, 1996). Similarly, a high rate of accumulation of herbarium specimens may indicate that plant collectors have visited the area often, giving pseudo-abundance levels of populations. This implies that some populations may not have been documented in localities where they occur and / or during subsequent visits to the same site while others might be well- or over-represented in herbaria, making it difficult to interpret the absence and abundance of records in apparently suitable habitats or over time. Table 5.1 gives some of the species that have been poorly known due to collecting bias. The lack of relationships between specimen accrual and changes in population and range sizes therefore makes it impossible to devise direct mathematical relationships between quantities and rates of accumulation of herbarium records on one hand and degrees of risks on the other. In addition, accurate species distribution maps involving specimens collected prior to 1960 may be impossible because locality information has often been geo-referenced to a country, province or nearest town, lake, river or mountain range, which may be fuzzy or several kilometres away. Furthermore, some collecting localities cannot be traced using gazetteers or maps, thus causing a failure in assigning latitude and longitude references. Plotting point distribution maps using locality information obtained from herbarium specimen labels therefore does not give the correct picture of the number and size of localities and habitat fragmentations.

A large sample of herbarium specimens may be required for discerning informative patterns of species distributions and inferences about the number, size and fragmentation of locations. However, large samples of herbarium records cannot provide a remedy for collections which are inherently non-representative of species distributions. Obtaining specimens on loan to cover the entire range of species distributions can be difficult, but this problem can be circumvented by enhancing access to electronic data. The mandatory deposition of geo-referenced primary (locality) data in electronic form as a requirement for publishing revisions may help build the locality database and enhance access to information, but a lot still needs to be done (Schatz, 2002). Due to space limitations, most herbaria are already forced to limit the storage of additional specimens collected from the same area (Prance, 1984). Restricting the storage of additional specimens from the same locality may directly reduce the number of specimens and distort age distribution patterns, particularly for recent collections.

Table 5.1. Examples of common and / or abundant species that have been poorly known in herbaria due to collecting bias.

Species	Reason for the poor collection	Reference
Quaqua multiflora	Succulents are difficult to press	Bruyns (1999)
Laccosperma acutiflorum	Leaves are large and thorny	Prance *et al.* (2000)
Loudetia filifolia	Access to mountainous areas	This study
Erisma japura	Apparently overlooked by collectors	Donoghue & Alverson (2000)
Mucuna irritans	The handling of pods that cause itching is difficult	Personal experience
Mollugo namaquensis	Plants are small (2-4 mm high), short-lived and sparse	Adamson (1958)

Knowledge of range expansion for most species, including *Orbea* (Apocyaceae), a bacterium *Prochlorococcus* now known to be one of the major phytoplanktons in tropical and subtropical seas and a shrub *Neviusia cliftonii* (Rosaceae) locally abundant in Shaska, northern California, is increasing (Prance, 1984; Cracraft, 2000; Prance *et al.*, 2000; Donoghue & Alverson, 2000). This apparent range expansion is reminiscent of gaps in knowledge of the number and size of localities at a particular time. Because of the limitations mentioned above, the need for caution in the interpretation of data gleaned from herbarium specimen labels for conservation status and ecological pattern assessments has therefore been expressed (Rhoads & Thompson, 1992; Stern & Eriksson, 1996; MacDougall *et al.*, 1998; Willis *et al.*, 2003). However, the data may be useful in setting priorities for species that may require detailed conservation status assessments and identifying those which do not. To do this, a method has to be developed.

5.1.7 *New proposal*

Ages of herbarium collections and numbers of herbarium holdings per species, localities and habitat types for species of *Loudetia* have been analysed to yield a new method for predicting risks of species. The method was applied to predict risks for species of *Syncolostemon,* tested by using field data of species of *Loudetia* and *Syncolostemon* occurring in South Africa and Swaziland and confirmed by comparing results of risk assessments based on herbarium records with the IUCN SSC (2001) method based on field data for the same species (Figure 5.3).

5.2 Aim and objectives

The aim of this study is to offer a new method for predicting the risk of extinction based on herbarium records of species of *Loudetia.* This aim has been achieved through the following objectives: (1) determining useful parameters for predicting the risk of extinction using herbarium specimens of *Loudetia*, (2) predicting the conservation status of selected species of *Syncolostemon* using the method so developed (3) testing the method so developed by gathering and analysing field-based data using the IUCN SSC (2001) method.

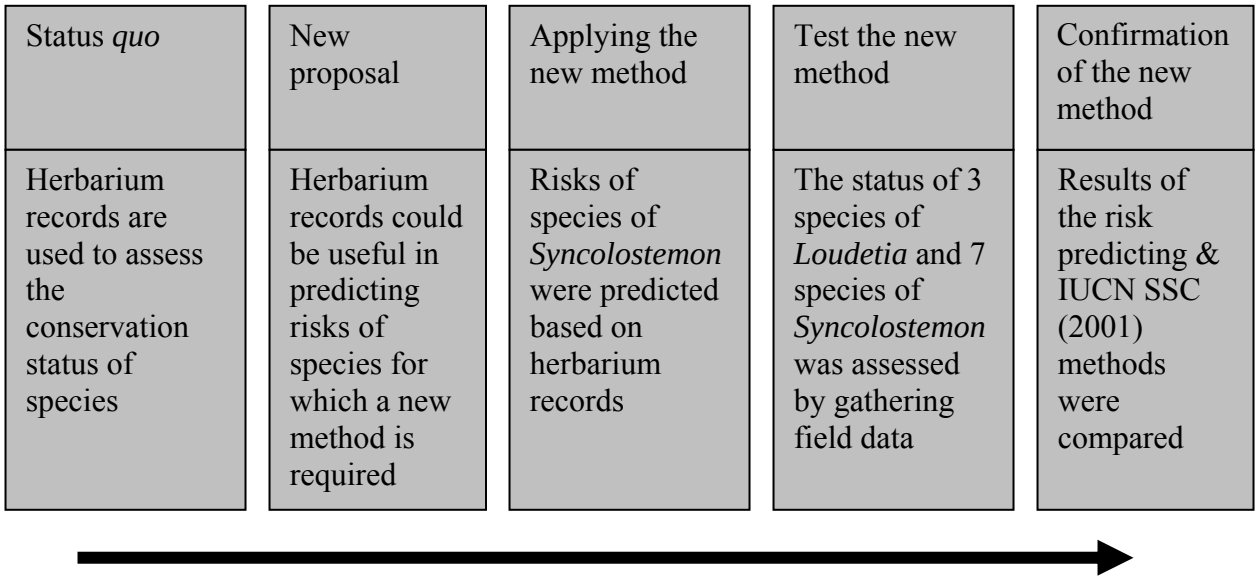


Figure 5.3. Formulation of the proposed method for assessing risk of species.

5.3 Motivation for the risk assessment method

Methods that recommend the use of herbarium records have provided guidelines for the Red Listing or rarity classifications for use in designing conservation strategies (Prance, 1984; MacDougall *et al.,* 1998; Willis *et al.,* 2003; Victor & Keith, 2004). While the assessment of the conservation status of endemic and well-collected groups is possible, the degree of uncertainty inherent in the available data for many species would have compelled one to consider herbarium records useful primarily in setting priorities for detailed conservation status assessments. The detailed field-based assessments to follow could dampen shortfalls arising from intermittent accumulations of specimens. By restricting conservation status assessment studies to species already prioritized in preliminary analyses based on herbarium records, funding can be more effectively deployed.

5.4 *Materials and methods*

5.4.1 *Materials*

The taxonomy and nomenclature of *Loudetia* (this study) and *Syncolostemon* (Otieno, in preparation) have been revised with specimens studied still available. Data collected in the field and herbaria during the course of these systematic studies provided an opportunity for choosing these genera to develop the risk predicting method. Materials were 2308 herbarium specimens belonging to 18 species of *Loudetia* (average 128 per species) collected over the past 17 decades throughout Africa (latitudes: 14ºN and 20ºS) and Madagascar. Of these, 726 specimens (representing 31.5%; average 40 per species) were collected during the past 3 decades. These specimens were studied at J, MAL, PRE, SDNH and SRGH. In addition, K and B provided electronic images, ETH locality data and BR, EA, ETH, K, PAT, PRE, SRGH, UPS and UWO loans of herbarium specimens

(abbreviations for herbaria follow Holmgren *et al.* (1991) and Smith & Willis (1999)). The recently revised *Syncolostemon* and *Hemizygia* provided 773 specimens which were studied for locality information and numbers of specimens per species.

5.4.2 *Data collection*

Habitat types, dates of specimen collection and geo-references were obtained from herbarium specimen labels when available. Coordinates for specimens collected from Namibia, South Africa and Swaziland were available in or converted to ¼-degree squares using Leistener & Morris (1976), while the rest were in degrees, minutes and seconds. Both ¼-degree squares and the coordinates were used to plot distributions of species on maps. Maps and gazetteers were used to trace geo-references when localities are not accompanied by grid references (Willis et al., 2003). Gazetteers used include http://gnswww.nga.mil/geonames/GNS/index.jsp, Polhill (1970), Skead (1973), Leistner & Morris (1976), Pope (1998) and The Times Atlas of the World, 5th ed. Grid references and estimations of population and habitat sizes for *Loudetia camerunensis, L. filifolia, L. simplex, Syncolostemon albiflorum, S. argenteus, S. concinnus, S. parviflora, S. pretoriae, S. rehmannii, S. subvelutinus, S. thorncroftii* and *S. transvaalensis* were also obtained in the field in Swaziland and South Africa using GPS. The field data were used to assess the risk based on the IUCN SSC (2001) method, results of which were compared with those obtained using the risk predicting method based on herbarium records for the same species.

5.4.3 *Data analyses and inferences*

Criteria A to C (Table 5.3) were developed using herbarium records as follows:

5.4.4 *Criterion A: Accumulation of specimens over time*

Ages of herbarium records collected during the past 17 decades were plotted on a graph from which periods of high and low collecting intensities were captured following MacDougall *et al.* (1998). Relative frequencies of specimen accumulations during the period of high collecting intensity (Criterion A1) and over the past 3 decades (Criterion A2) and rates of specimen accumulations during the recent 3 decades were classified into 4 groups, each assumed to represent ranges defining a risk indicator. These groups were classified as 1st, 2nd, 3rd and Low Priorities. Each class of values was standardised by dividing by the highest value.

5.4.5 *Criterion B: Spatial distributions*

Collecting localities were plotted on grid maps using MAPPIT (1995) to reveal the patterns of spatial distributions of species. The ¼-degree square grids were selected under the assumption that genetic exchange through pollen or seed dispersal between plants lying more than 20 km apart may be ineffective. The number of locations of populations, apparent habitat fragmentation and indications of range sizes were inferred from these maps. Clusters of points on maps constituted independent populations separated by unfilled grids (Criterion B1). The number of empty grids between clusters of points was then counted and regarded as indicating habitat fragmentations or naturally isolated patches and the degree of population isolation (Criterion B2). Filled grids were counted to provide an estimation of apparent range sizes (Criterion B3). The data were classified

to correspond with risk indicators as in Criterion A. The results were standardized by dividing by the highest value.

5.4.6 *Criterion C: Habitat types*
The number of habitat types was used to provide indications of the risk of population extirpation due to habitat affinities. The habitat-types data set was classified into 4 groups corresponding to those in Criteria A & B and standardized by dividing by the highest value.

5.4.7 *Assigning priority ranks to species*
The range of values in each species was compared with the range of the scale for each risk indicator and a matching code was recorded. Codes representing risk indicators assigned to each species were compared using correlation coefficients to determine whether ranks correspond to each other within a particular species. A student's *t-test* was calculated to determine if there were differences between risk indicators.

5.4.8 *Determination of priorities for conservation status assessments*
All codes assigned to a particular species were multiplied by each other to obtain the product value(p). An inverse of p $(1/p)$ was calculated to obtain an overall risk-predicting index (RPI) ranging between 0 and 1. Principal coordinates analysis (PCOORDA) was performed using NTSYS-pc (Rohlf, 1998) on codes assigned to species to help identify groups. PCOORDA was performed by standardizing the raw data matrix, calculating the distance matrix, double centering the distance matrix, calculating eigen values and projecting these into 2- and 3-dimension plots. These groups were then used to determine the range of RPI for each category.

5.4.9 *Application of the new method by risk assessment of* Syncolostemon *using herbarium records*
A total of 773 herbarium specimens belonging to 32 species of *Syncolostemon* were examined. Data collected from herbarium specimens included the number of specimens collected over a period of 5 decades, number of specimens gathered during the past 3 decades, collecting localities and habitat types. Collecting localities were converted into degrees and minutes or degrees, minutes and seconds and ¼-degree squares using the gazetteers. These coordinates were used to plot maps using ¼-degree square grids. Inferences about the number of locations, habitat fragmentation and range size were drawn from these maps. Extinction risks for each species of *Syncolostemon* were predicted using the risk predicting method developed with species of *Loudetia*.

5.4.10 *Testing of the method*
Population sizes, number and size of locations for *Loudetia camerunensis, L. filifolia* and *L. simplex* and selected species of *Syncolostemon* were estimated in the field in Swaziland and Mpumalanga & Limpopo provinces of South Africa. Latitudes and longitudes of locations were recorded on site in degrees, minutes and seconds using a GPS. The data were used to assess the conservation status based on the IUCN SSC (2001) method (Appendix 5.1). The results of the risk predicting and IUCN SSC (2001)

methods were coded and compared using correlation coefficients. A student's *t-test* was calculated to determine within-method and between-method variation.

5.4.11 *Modifying the method*

Comparison with the IUCN SSC (2001) method revealed that differences exist between the risk method and the conservation assessment method. The risk method was then modified to accommodate as many threatened plants as possible within the 1st, 2nd and 3rd priority ranks for assessment. The number of specimens collected over a period of 3 decades was thus reclassified by expanding the range of each class of values.

5.5 Results

5.5.1 *Missing and imprecise information*

The distribution of missing information in the 2308 herbarium specimen labels for species of *Loudetia* examined is as follows: 2.2% without the year of collection, 1.6% no country of origin and 3.1% omit the locality. In addition, the collecting localities in 11.9% of the specimen labels were vague and 13.3% were not traceable using gazetteers and maps for converting into latitudes and longitudes (Figure 5.4). Thus, the localities for 30.5% of the specimens of species of *Loudetia* could not be traced.

5.5.2 *Accumulation of herbarium specimens*

Collecting intensities exhibit variations with time, being low between 1831 and 1900 for all species of *Loudetia* (Figure 5.5), but increasing fast from 1901, 1911 and 1921 and slowly after 1951 and 1980 (Figure 5.5a to 5.5f). *Loudetia camerunensis* and *L. simplex* have increased fast from 1971 to 1980 (Figure 5.5c), reflecting a recent surge in specimen collecting activities in southern Africa or difficulties in accessing specimens north of the southern Africa region. All species are represented in collecting records gathered during high intensities, but some are missing in low activities (1831 to1900 and 1971 to 2000, respectively). Species that are missing in herbarium collections gathered between 1971 and 2000 include *L. angolensis* and *L. coarctata* (Figure 5.5a & 5.5f), *L. phragmitoides* (Figure 5.5e), *L. kagerensis, L. tisserantii* and *L. togoensis* (Figure 5.5a, 5.5b & 5.5d), *L. densispica* and *L. pennata* (Figure 5.5c, 5.5b & 5.5e) and *L. hordeiformis* (Figure 5.5f). The general pattern shows that collecting activities have been unsystematic. Unsystematic collecting activities are revealed by the isolated collections in *L. annua, L. arundinacea, L. togoensis* and *L. densispica* between 1831 and 1920 (Figures 5.5a to 5.5c & 5.5e to 5.5f, respectively). In addition, *L. demeusei* is not represented in records collected between 1981 and 1990, but it has been documented in the preceding and subsequent decades (Figure 5.5a) and high collecting intensities in *L. tisserantii* and *L. densispica* each follow a decade in which records are missing (Figure 5.5b & 5.5c).

5.5.3 *Risk predictors*

Three groups of variables have been identified using the data set of the period of high collecting intensity. Each of these groups was considered to represent a risk predictor. Thus 4 risk predictors: 1st, 2nd, 3rd and Low Risk (LR) were identified. While detailed conservation status assessments are urgently required for species falling under the 1st to 3rd categories, they may not be necessary for the LR category.

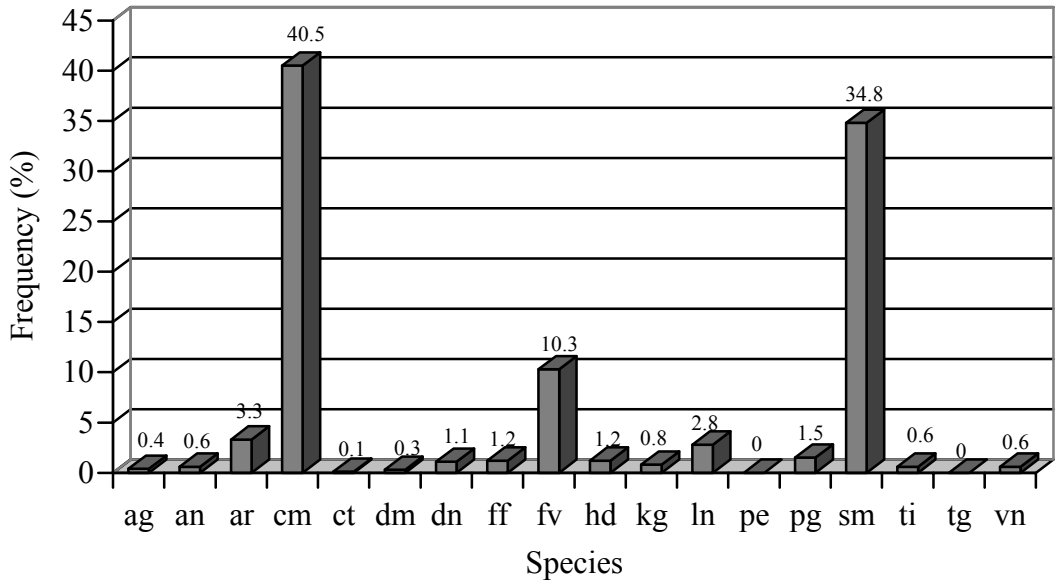


Figure 5.4. Frequency of missing and imprecise information from specimen labels for collections of species of *Loudetia* dating from 1831 to 2000. Vague collecting locality refers to imprecise reference to collecting locality such as the name of an extensive geographical feature (Soutpansberg, Lake Kivu, Zambezi River, Luangwa Valley, Abercorn – Mpulungu road) or country (Benin), region (Transvaal) and city (Pretoria). Abbreviations used: ag = *Loudetia angolensis,* an = *L. annua,* ar = *L. arundinacea,* cm = *L. camerunensis,* ct = *L. coarctata,* dm = *L. demeusei,* dn = *L. densispica,* ff = *L. filifolia,* fv = *L. flavida,* hd = *L. hordeiformis,* ln = *L. lanata,* kg = *L. kagerensis,* pe = *L. pennata,* pg = *L. phragmitoides,* sm = *L. simplex,* ti = *L. Tisserantii,* tg = *L. togoensis* and vn = *L. vanderystii*.

5.5.4 *Rates of specimen accumulations*

Over three decades, rates of specimen accumulation have decreased in recent years (Figure 5.6). However, there are increases between 1981 and 1990 in *L. arundinacea, L. densispica, L. lanata, L. tisserantii* and *L. vanderystii* and almost constant rates between 1971 to 1980 and 1991 to 2000 in *L. demeusei.* About 81% of specimens which have been accumulated during the period of high collecting intensities belong to *L. camerunensis* (35.9%), *L. simplex* (28.8%), *L. flavida* (11.8 %) and *L. arundinacea* (4.3%; Figure 5.7a). This trend is maintained for specimens accumulated over the past 3 decades of which about 89% are distributed among *L. camerunensis* (40.5%), *L. simplex* (34.8%), *L. flavida* (10.3 %) and *L. arundinacea* (3.3%), the rest of the species being represented by less than 3% each (Figure 5.7b). Distributions of collecting records examined show a concentration of points for *L. camerunensis* and *L. simplex* in Malawi, South Africa, Zambia and Zimbabwe, but relatively sparse elsewhere (see Chapter 3).

Species also fall into 4 classes using the number of specimens collected over a period of 3 decades, but members have lower values than during the period of high collecting intensities. Ranges of accumulated specimens were initially classified as: no specimen

during the past 3 decades for 1st priority, at least 1-12 (standardized to 0.05-0.95) for 2nd priority, 13-20 for the 3rd priority and ≥21 (≥1.00) for LR (Criterion A2; Table 5.3; Appendices 5.2 & 5.3). These classes were later modified to 0-5, 6-19, 20-29 and ≥30 for the LR category to accommodate as many threatened species as possible within the 1st to 3rd risk categories (Table 5.11).

5.5.5 *Distribution patterns of species*

Data sets of number of locations, number of empty grids and number of filled grids were each classified into 4s corresponding to 1st to 4th categories and standardized (Table 5.3).

5.5.6 *Habitat types*

Species of *Loudetia* predominantly grow in open environments, mainly in grasslands and open patches in woodlands (Table 5.2). They are associated with a variety of habitats, including rock crevices in *L. filifolia,* seepage areas (*L. phragmitoides*), sandy soils (*L. lanata*), shallow rocky soils (*L. simplex*) and drainage-poor areas (*L. camerunensis* and *L. coarctata.* Only *L. densispica* and *L. togoensis* have been documented in disturbed environments. Seven species are restricted to one habitat type, 7 others have been recorded in 2 to 3 and 4 species in 4 to 7 habitat types each. Four groups can be identified with respect to habitat types: classified as species occurring in 1, 2, 3-4 & ≥5 habitat types corresponding to 1st, 2nd, 3rd and LR categories, respectively (Table 5.3).

5.5.7 *Predicting priorities for conservation status assessments*

Predicted risks of species of *Loudetia* are presented in Table 5.4. Species with a risk predicting index (RPI) <0.1000 (*L. arundinacea*, *L. camerunensis, L. simplex* and *L. vanderystii*) are clearly separated from those with RPI >0.1000 (Figure 5.9). Species with the same RPI value (*L. camerunensis L. simplex*) are plotted on the same spot, with 5 clusters identified, two of which define one risk category (Figure 5.9).

5.5.8 *Comparison of parameters*

The number of specimens collected during the period of high collecting intensity (parameter A1) is moderately associated with specimen accrual over three decades (A2) with moderate correlation coefficient and no significant difference (correlation coefficient (corr.) = 0.88, p = <0.001; Table 5.5). Both A1 and A2 are weakly associated with the predicted risk (PR) from which they are significantly different (corr. = 0.40 & p < 0.001 & 0.015, respectively). The number of locations (B1) is not associated with parameters A1-A2, B1 & B3, but weakly associated with C (Table 5.4). There are significant differences between B1 and all other parameters, including the PR, but with the exception of B2. All parameters are individually significantly different to PR.

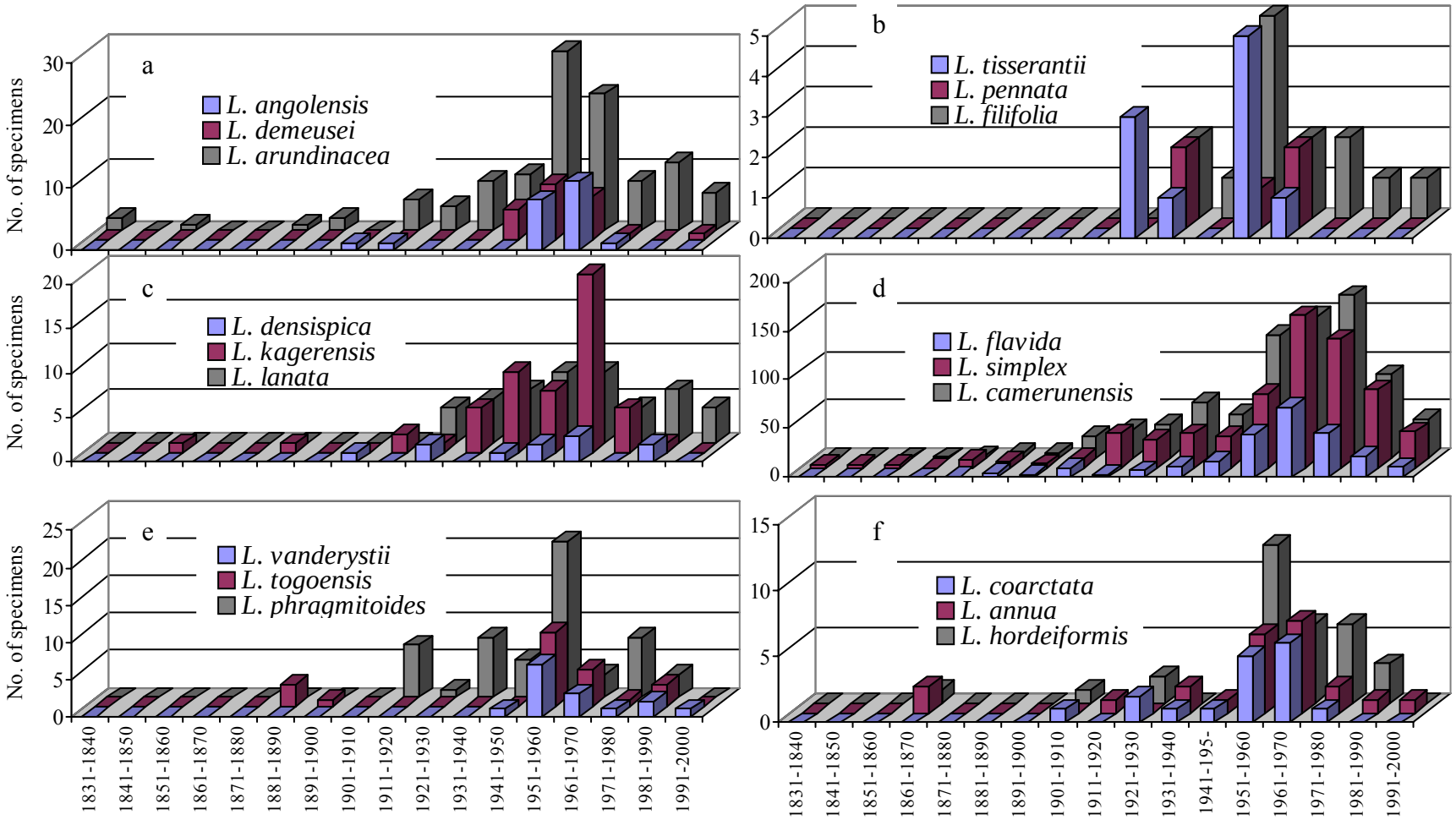


Figure 5.5. Temporal distributions of collecting records of species of *Loudetia* in Africa. Collecting intensities were low between about 1831 and about 1920. There was a steady increase in collecting intensities from about 1921 to about 1970. After about 1970 to present, the rate has steadily decreased. The lag period between collection and accession in herbaria might exert an effect on specimens gathered per decade, but its influence is considered negligible. n = 2308 specimens.

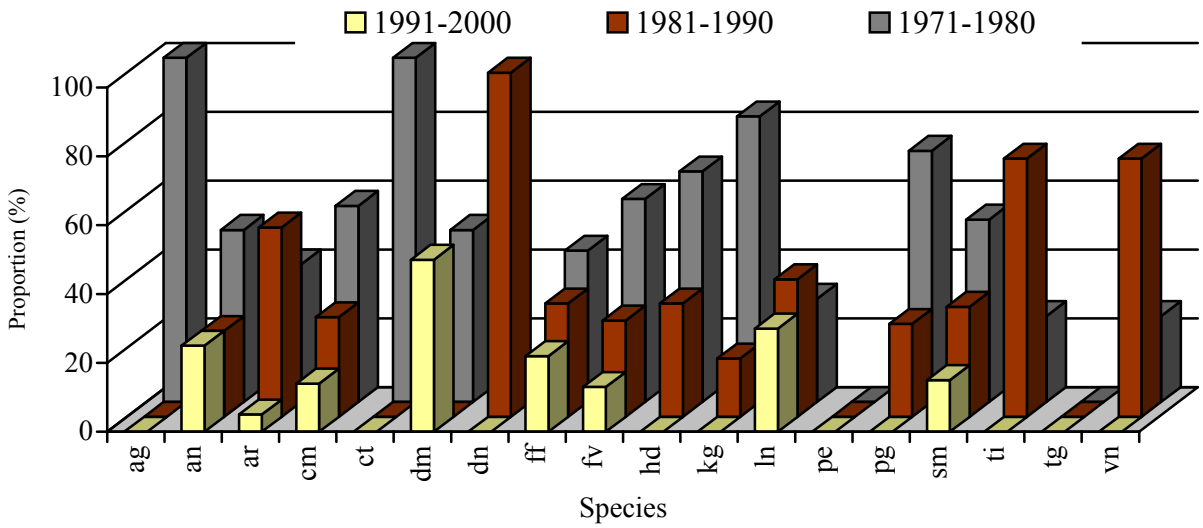


Figure 5.6. Comparison of proportions of specimens collected over a period of 3 decades in species of *Loudetia* with a total of specimens per species representing 100%. n = 726 specimens. Abbreviations used and sample sizes for the period 1991-2000: ag = *Loudetia angolensis* (n = 1), an = *L. annua* (n = 4), ar = *L. arundinacea* (n = 25), cm = *L. camerunensis* (n = 293), ct = *L. coarctata* (n = 1), dm = *L. demeusei* (n = 2), dn = *L. densispica* (n = 8), ff = *L. filifolia* (n = 9), fv = *L. flavida* (n = 75), hd = *L. hordeiformis* (n = 9), ln = *L. lanata* (n = 20), kg = *L. kagerensis* (n = 6), pe = *L. pennata* (n = 0), pg = *L. phragmitoides* (n = 11), sm = *L. simplex* (n = 249), ti = *L. tisserantii* (n = 4), tg = *L. togoensis* (n = 0) and vn = *L. vanderystii* (n = 4).

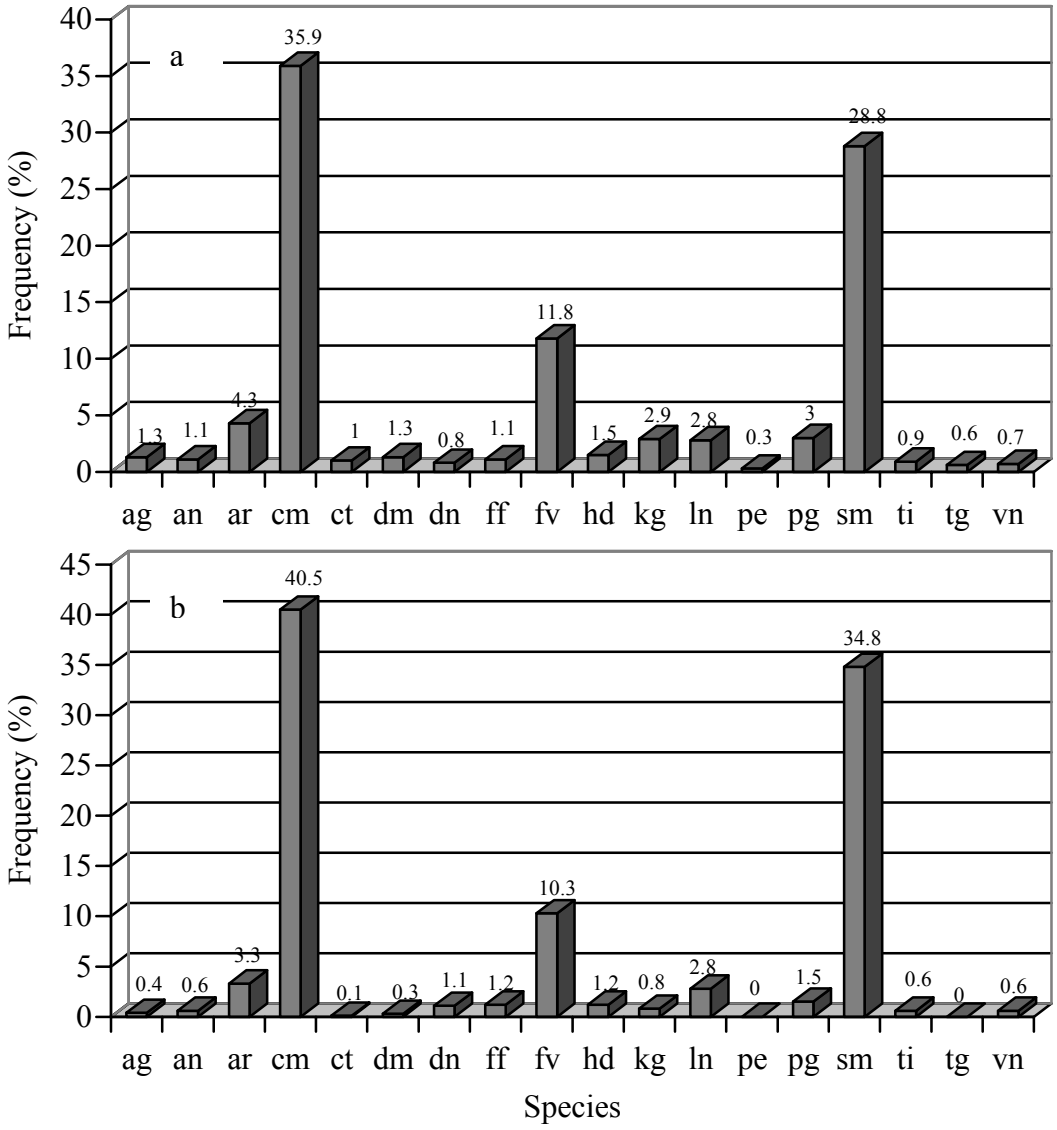


Figure 5.7. Relative frequencies of herbarium specimens of *Loudetia* collected: a = during the period of high collecting intensities from 1951 to 1970 (n = 1756) and b = over a period of 3 decades from 1971 to 2000 (n = 726). Only *L. camerunensis, L. simplex* and *L. flavida* exceed 10%, with the rest below 5% of the total number of specimens accrued. Abbreviations of names used are as in Figure 5.4.

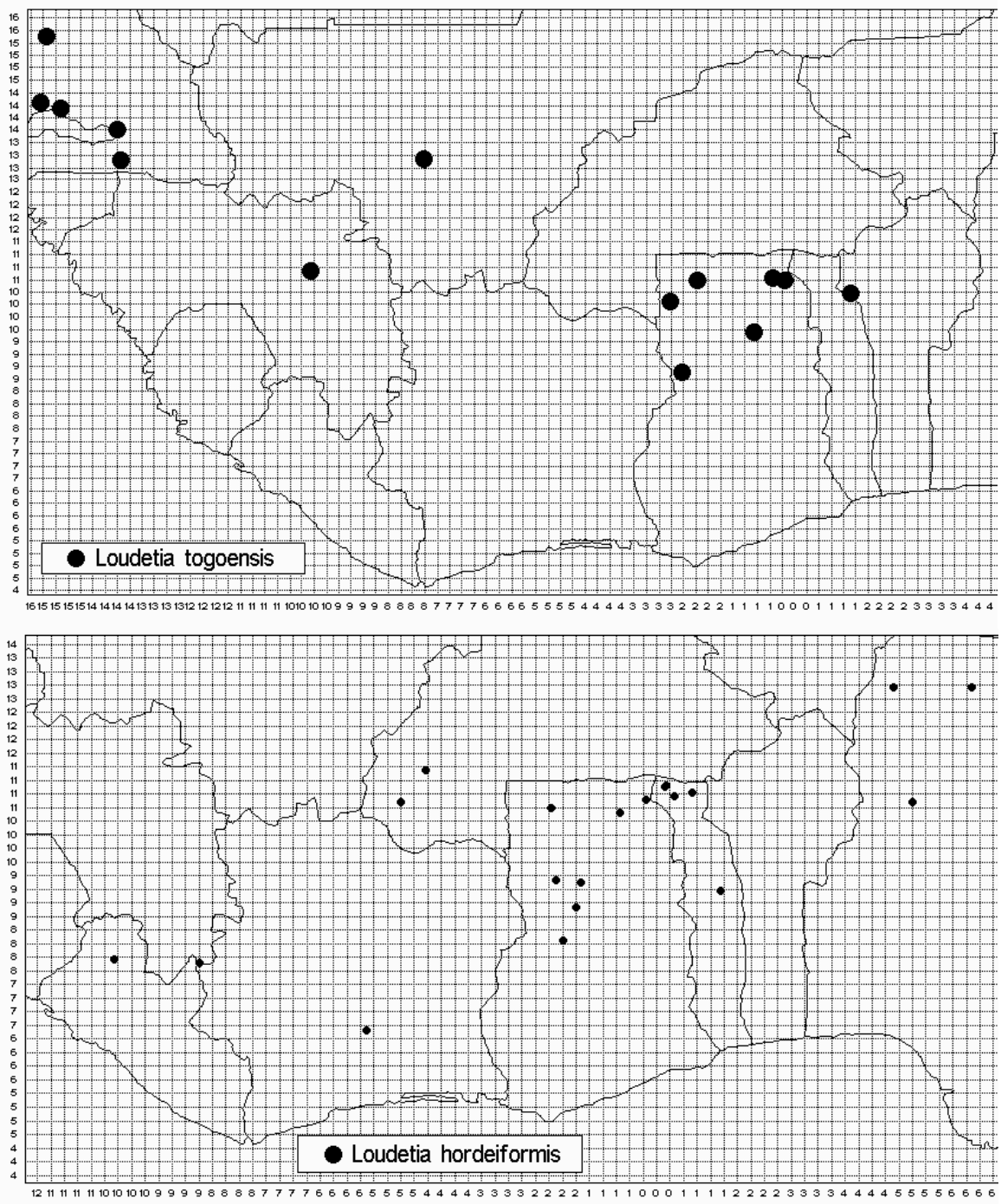


Figure 5.8. The distribution of selected species of *Loudetia,* illustrating the quarter degree square grids which were used to determine the number of locations and habitat fragmentations. Dots in adjacent quarter-degree grids, or separated by only two empty grids are assumed to represent components of a single interbreeding population whereas those separated by three or more quarter degree-squares are assumed to represent independent populations.

Table 5.2. Types of habitat in which species of *Loudetia* have been documented. Symbols used 1 = present and 0 = absent. Abbreviations used follow Figure 5.4.

Habitat type	ag	an	ar	cm	ct	dm	dn	ff	fv	hd	kg	ln	pe	pg	sm	ti	tg	vn
Open *Danielia* Woodland	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
Open Miombo Woodland	1	0	1	1	0	1	0	0	1	0	0	0	0	0	1	0	0	0
Hillside Miombo Woodland	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
Open *Combretum* Woodland	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Open *Acacia* Woodland	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
Open *Burkea* / *Baikea* / *Terminaria* / *Parinari* / *Ochna* Woodland	0	0	0	0	0	0	1	0	0	0	0	1	0	0	0	0	0	0
Open Mixed Woodland	0	1	1	1	0	0	0	0	1	1	1	1	0	0	0	0	0	0
Savanna Woodland	0	1	1	1	0	0	0	0	1	1	0	0	0	0	0	0	1	0
Cultivated / Regenerating Woodland	0	0	0	1	0	0	1	0	0	0	0	0	0	0	0	0	1	0
Savanna Grassland	0	0	0	0	0	1	0	0	0	0	0	0	1	0	0	1	1	1
Upland Grassland	0	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
Riverine Grassland	0	0	0	1	0	0	0	0	0	0	1	0	0	0	0	0	0	0
Swamp	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
Flood plain	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
Serpentine	0	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
Sourveld	0	0	0	0	0	0	1	0	0	0	0	0	0	0	1	0	0	0
Rock crevices	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0
Total	1	2	4	7	1	2	3	1	5	2	2	2	1	1	5	1	3	1

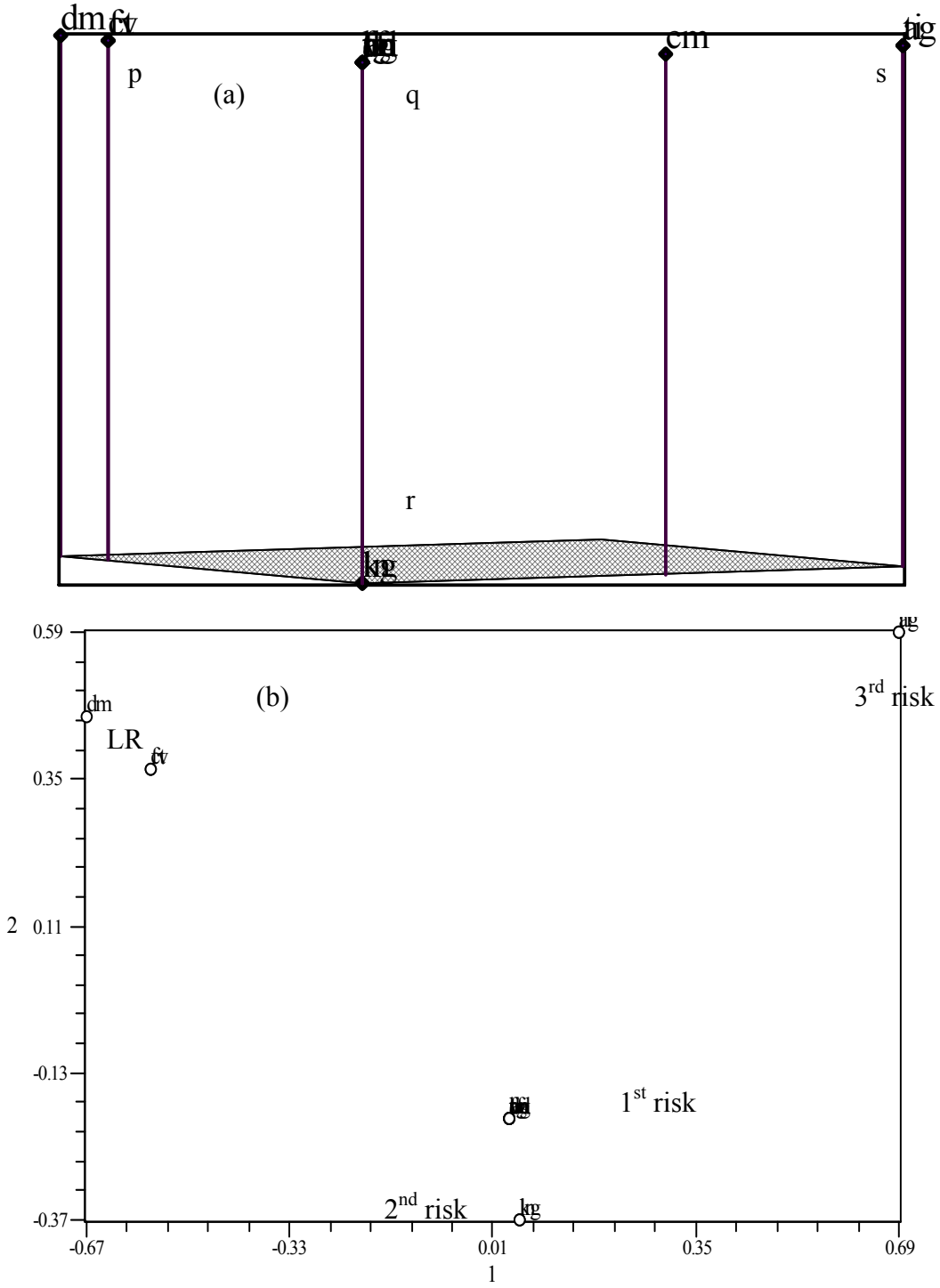


Figure 5.9. PCOORDA of the predicted conservation status data set. Five clusters assignable to the 1st Risk, 2nd risk, 3rd risk and LR can be identified in both 3-dimentional (a) and 2-dimensional (b) plots. p = *L. coarctata, L. demeusei* & *L. flavida,* q = *L. densispica, L. filifolia, L. hordeiformis, L. lanata, L. phragmitoides* & *L. vanderystii,* s = *L. angolensis* & *L. tiserrantii,* r = *L. kagerensis.* Abbreviations remain as in Figure 5.4.

Table 5.3. Risk-predicting parameters for species of *Loudetia* derived using herbarium data. Risk indicator: 1 = 1st Priority with respect to a particular parameter, 2 = 2nd Priority, 3 = 3rd Priority and 4 Lower Priority with respect to a particular parameter. These may be equivalent to Critically Endangered, Endangered, Vulnerable and Least Concern of the IUCN SSC (2001) method, respectively. Conservation status assessment might be required for risk categories 1 to 3 and apparently no need for field-based conservation status assessments for category 4.

Criteria	Considerations	Scale of risk indicator		
		1	2	3
A	Number of specimens collected during: 1. the period of high collecting intensities 2. the past 3 decades	1-15 / decade 0	16-30 / decade 1-12 / decade	31-45 / dec 13 / 24 dec
B	1. Number of locations of population occurrence	1-2	3-5	6-10
	2. Number of empty grids separating occupied ones indicating the apparent extent of habitat fragmentation	≥6 grids	4-5 grids	2-3 grids
	3. Number of filled contiguous grids indicating the area of occupancy	1-10 grids	11-20 grids	21-40
C	1. Number of habitat types	1	2	3-4

Table 5.4. The conservation status of species of *Loudetia* predicted using herbarium data. A, B, C are criteria (Table 5.3), *p* = product of codes = A1 X A2 X B1 X B2 X B3 X C, PR = predicted risk, RPI = Risk Predicting Index defined as $1/p$, I = First Priority for detailed conservation status assessment with RPI of 0.5000 to 1.0000, II = 2nd Priority with RPI ≥0.1000 but <0.5000, III = 3rd Priority with RPI <0.1000 but >0.0800, IV = Low Priority with RPI <0.0800, apparently detailed conservation status assessment are not urgently needed in the fourth category, Concl. = conclusion, LP = Low Priority.

Species	A1	A2	B1	B2	B3	C	*p*	RPI	PR	Concl.
L. pennata	1	1	1	1	1	1	1	1.0000	I(A1,2B1,2,3C)	1st PR
L. coarctata	1	2	2	1	1	1	4	0.2500	I(A1,2B2C)	1st PR
L. lanata	1	2	2	1	1	2	8	0.1250	II(B2,3C)	2nd PR
L. demeusei	1	2	1	3	1	2	12	0.0833	III(A1B1,3C)	3rd PR
L. filifolia	2	2	3	1	1	1	12	0.0833	III(A1B2,3C)	3rd PR
L. tisserantii	1	2	2	3	1	1	12	0.0833	III(A1,2B3C)	3rd PR
L. angolensis	1	2	2	3	1	1	12	0.0833	III(A1,2B3C)	3rd PR
L. annua	1	2	2	2	1	2	16	0.0625	IV(A1B2,3C)	LR
L. arundinacea	2	3	4	3	4	3	864	0.0012	IV(B2)	LP
L. camerunensis	4	4	4	4	4	4	4096	0.0003	IV(B2)	LP
L. densispica	1	2	3	3	1	3	54	0.0185	IV(A1,2B3)	LP
L. flavida	3	3	3	3	4	4	1296	0.0008	IV(B2)	LP
L. hordeiformis	1	2	2	3	1	2	24	0.0417	IV(A1,2B3C)	LP
L. kagerensis	2	2	3	3	3	2	216	0.0046	IV(A1,2B3C)	LP
L. phragmitoides	1	2	4	3	3	1	72	0.0139	IV(A2C)	LP
L. togoensis	1	1	2	4	1	3	24	0.0417	IV(A1,2B3)	LP
L. simplex	4	4	4	4	4	4	4096	0.0002	IV	LP
L. vanderystii	1	2	2	4	1	1	16	0.0625	IV(A1B3C)	LP

Table 5.5. Comparison of risk predicting parameters. Association measures the correlation coefficient between two data sets. All correlation coefficient are positive. Parameters A to C are as in Table 5.3. For the *t-test,* + = significantly different, - = not significantly different.

Student's *t-test* (*P*-values at 95% confidence level)

	A1	A2	B1	B2	B3	C
A1						
A2	<0.001 (+)					
B1	<0.001 (+)	0.027 (+)				
B2	<0.001 (+)	0.029 (+)	0.273 (-)			
B3	0.068 (-)	0.055 (-)	0.001 (+)	0.007 (+)		
C	0.007 (+)	0.289 (-)	0.044 (+)	0.015 (+)	0.180 (-)	
PR	<0.001 (+)	<0.001(+)	0.007 (+)	0.008 (+)	<0.001(+)	<0.001 (+)

Association

	A1	A2	B1	B2	B3	C
A1						
A2	0.88					
B1	0.69	0.72				
B2	0.37	0.41	0.38			
B3	0.82	0.79	0.82	0.43		
C	0.74	0.68	0.52	0.51	0.68	
PR	0.40	0.42	0.50	0.73	0.52	0.42

5.5.9 *Applying the new method by risk prediction of species of* Syncolostemon

The new method was used to predict risks of species of *Syncolostemon* using herbarium specimens. Of the 42 species of *Syncolostemon,* 36 are represented by 1-30 specimens in herbaria (falling within 0.01-0.42; Table 5.3), which could place them within the first and second priorities for detailed conservation status assessment (Criterion A1; Tables 5.3 & 5.6). Thirty-five species have been represented in collections made over three decades, which is an indication of their persistence in the wild (Criterion A2). Thirty-two species have been documented in 1-2 locations (falling within 0.17-0.33 (Table 5.3), but there is no apparent range fragmentation from herbarium data (Criteria B1 & B2, respectively). Thirty-eight species exhibit range restriction to 1-20 filled grids (falling within 0.03-0.57, Table 5.3) and growing in 1-2 (0.2-0.4, Table 5.3) habitat types (Criteria B3 & C, respectively). Due to small numbers of specimens (Criterion A1), locations (Criterion B1), range and habitat type restrictions (Criteria B3 & C, respectively), 4 species of *Syncolostemon* fall within the 1st priority, 9 species in the 2nd priority and 4 species in the 3rd priority, whereas 25 species fall within the low priority for detailed conservation status assessment (Table 5.6).

5.5.10 *Testing the new method by assessing the conservation status using the IUCN SSC (2001) method based on field data*

A large portion of the natural habitats for *Loudetia camerunensis, L. simplex* and species of *Syncolostemon* in Mpumalanga and parts of Swaziland have been modified for plantation forestry, mainly species of *Pinus* for the paper industry. Patches of the

remaining suitable habitats are either conserved or threatened with the expansion of forestry plantations, which is underway in parts of Mpumalanga. Forestry plantations and human settlements in Swaziland have reduced the locations of species of *Loudetia* to isolated and fragmented patches. While species of *Sporobolus, Melinis, Hyparrhenia* and the bracken fern *Pteridium aquilinum* have invaded old pine plantations that have now been clear-cut for at least 2 years, *L. camerunensis* and *L. simplex* were not seen in such environments at Buffelskloof and Long Tom Pass in Lydenburg during the field survey (February to March 2005), implying that these species do not withstand disturbance. However, some of the protected pristine patches are extensive enough (>5 km²) to accommodate viable populations of *Loudetia camerunensis* and *L. simplex* (Table 5.4). The extensive areas of occupancy, population sizes and numbers of populations therefore make *L. camerunensis* and *L. simplex* not threatened (Table 5.7). *Loudetia filifolia* is confined to rock crevices of quartzite outcrops in the protected Soutpansberg and Blouberg in South Africa, but like *L. camerunensis* and *L. simplex*, it also occurs elsewhere in Africa. The status of locations of *L. filifolia* in Nigeria and Madagascar is not known. Although the species is locally abundant with an estimated greater than 100,000 individuals, its confinement to the rock crevice habitat type makes it vulnerable to drought and *L. filifolia* is therefore considered Vulnerable B1ac(ii,iv) (Table 5.7).

Only 19 mature plants of *Syncolostemon rehmannii* were counted along an extremely small area on the slope of a ridge at Buffelskloof. This species has also been documented at Hautboschdorp and in Polokwane, Limpopo Province at New Agatha and at Wolkberg. Other localities in Mpumalanga are Mac Mac Falls and Tweefontein Experimental area in Sabie. This gives four known localities. A search at Mac Mac Falls and at Tweefontein was not successful and the status of the remaining localities is not known, but it is unlikely that large populations exist. The previous (2001) search for the species at Buffelskloof was not successful, implying that the population is restricted to a very small area or it may be severely fluctuating. Although continuing decline cannot be determined, the extremely small area of occupancy (<50 m²**)**, habitat fragmentation, population and / or range fluctuation and the small number of mature individuals (19 encountered at one locality**)** place *S. rehmannii* as critically endangered: B1ac(i-ii,iv) + 2ac(i-ii,iv); D. About 380 mature individuals of *Syncolostemon albiflorum* are confined to narrow rocky habitats along a stream at Buffelskloof and along the Blyde River at Mac Mac. While the area and extent of occurrence are extremely small and fragmented, it is not known whether the range is declining or the population fluctuates. *Syncolostemon albiflorum* is therefore considered vulnerable: D. *Syncolostemon concinnus* occurs along a seasonal stream at Jericho, Hlangano, Swaziland, surrounded by farming communities. Grazing and crop cultivation are the main threats to the habitat. Pine plantations threaten the population 24 km from Josefsdal Border post with Swaziland on the Piggs Peak to Barberton road. Both locations are small, have small populations occupying restricted areas (Table 5.6), but fluctuations in the area of occupancy or population size are not known. Due to small populations, *S. concinnus* may be vulnerable: D. *Syncolostemon pretoriae, S. subvelutinus, S. thorncroftii* and *S. transvaalensis* cover extensive areas, but fluctuation in range or population sizes are not known. Since population sizes are estimated to be more than 10000 mature plants, these species are considered not threatened.

Table 5.6. Predicted conservation status of species of *Syncolostemon*

Species	1	A2	B1	B2	B3	C	P	1/P	PR	Concl.
S. comosus	1	2	1	1	1	1	2	0.5000	I(A1B1,2,3C)	1st PR
S. gerrardii	1	1	1	1	1	1	1	1.0000	I(A1,2B1,2,3C)	1st PR
S. ornatus	1	2	1	1	1	1	2	0.5000	I(A1,2B1,2,3C)	1st PR
S. oritrephes	1	2	1	1	1	1	2	0.5000	I(A1B1,2,3C)	1st PR
S. comptonii	1	2	2	2	1	1	8	0.1250	II(A1B1,3C)	2nd PR
S. concinnus	1	2	3	1	1	1	6	0.1667	II(A1B1,3C)	2nd PR
S. flabellifolius	1	2	1	2	1	1	4	0.2500	II(A1B1,2,3C)	2nd PR
S. foliosus	1	2	1	3	1	1	6	0.1667	II(A1B1,3C)	2nd PR
S. latidens	1	2	1	3	1	1	6	0.1667	II(A1B1,3C)	2nd PR
S. rehmannii	1	2	2	3	1	2	24	0.0417	IV(A1B1,3C)	2nd PR
S. rotundifolius	2	2	1	2	1	1	8	0.1250	II(A1B3C)	2nd PR
S. rugosifolius	1	2	1	3	1	1	6	0.1667	II(A1B1,2,3C)	2nd PR
S. stalmansii	1	2	1	3	1	1	6	0.1667	II(A1B1,3C)	2nd PR
S. subvelutinus	1	2	1	4	1	1	8	0.1250	II(A1B1,3C)	2nd PR
S. obermeyerae	1	2	2	3	1	1	12	0.0833	III(A1B3C)	3rd PR
S. ramosus	1	2	1	3	1	2	12	0.0833	III(A1B1,3C)	3rd PR
S. ramulosus	1	2	1	3	1	2	12	0.0833	III(A1B1,3C)	3rd PR
S. persimilis	1	2	1	3	2	1	12	0.0833	III(A1B1,3C)	3rd PR
S. albiflorum	2	3	3	2	2	1	72	0.0139	IV(A1B1,3C)	LP
S. argenteus	4	3	3	2	1	2	192	0.0053	IV(B3)	LP
S. bolusii	1	2	1	2	4	1	16	0.0625	1V(A1,2B1,3C)	LP
S. bracteosus	4	4	4	4	3	4	3072	0.0003	IV(A1B1,3C)	LP
S. canescens	3	4	4	3	4	4	2304	0.0004	IV(A1B1,3C)	LP
S. cinereus	1	2	1	3	2	4	48	0.0208	IV(A1B2,3C)	LR
S. densiflorum	2	2	2	3	1	2	48	0.0208	IV(A1B1,3C)	LP
S. elliottii	2	2	4	4	3	4	768	0.0013	IV(A1C)	LP
S. eriocephallus	1	2	1	4	2	1	16	0.0625	IV(A1B1,3C)	LP
S. floccosus	1	2	1	4	3	3	72	0.0139	IV(A1B1,3C)	LP
S. incunus	1	2	2	2	1	2	16	0.0625	IV(A1B1,2,3C)	LP
S. liniaris	1	2	2	2	1	2	16	0.0625	IV(A1B1,2,3C)	LP
S. macranthus	1	2	2	3	1	2	24	0.0417	IV(A1B3C)	LP
S. modestus	3	3	2	3	2	2	216	0.0046	IV(A1B1C)	LP
S. namapensis	2	2	1	3	2	1	24	0.0417	IV(A1B1,3C)	LP
S. parviflorus	4	2	4	3	2	4	768	0.0013	IV(B1,3)	LP
S. petiolatus	2	3	2	2	1	1	24	0.0417	IV(A1B1,3C)	LP
S. pretoriae	3	3	2	3	2	3	324	0.0031	IV(B1,3)	LP
S. punctatus	2	2	1	3	1	2	24	0.0417	IV(B1,3C)	LP
S. stenophyllus	2	2	2	3	1	1	24	0.0417	IV(A1B1,3C)	LP
S. teucriifolius	3	2	1	2	3	2	72	0.0139	IV(A1B1,2,3C)	LP
S. thorncroftii	1	2	2	2	2	3	48	0.0208	IV(A1B1,3C)	LP
S. transvaalensis	4	4	4	4	3	4	3072	0.0003	IV(B1,2,3)	LP
S. welwitschii	2	2	3	3	3	3	324	0.0031	IV(A1B2,3C)	LP

Table 5.7. Estimation of population and range sizes of selected species of *Loudetia* and *Syncolostemon* using field surveys. Population size was estimated by counting the number of mature individuals (mostly a tuft constituting an individual) in two 100 m transects laid 50 m apart in each location studied. In each transect, 10 quadrats each 1 m^2 were laid at 10 m intervals along the transect. An average number of mature individuals per 1 m^2 was calculated and multiplied by the area of occupancy (AOO, being the proportion of the area in square meters a species occupies in 100 m^2). Bold type = totals estimated in one locality.

Name	Location / habitat	¼-degree squares	Estimated AOO (m^2)	Average per m^2	Estimated population size
L. simplex			**2,872,250**		**3,567,675**
	Buffelskloof, Grassland (short grasses)	2530BB	600,000	3.21	1,926,000
	Buffelskloof Stream	2530BC	2,250	0.45	9,675
	Buffelskloof Hut and surrounding areas	2530BB	120,000	3.33	399,600
	Buffelskloof High Forest	2530BC	75,000	0.11	8,250
	Lochiel	2531DA	100,000	0.01	1,000
	Immelmansdal	2530DD	10,000	0.10	1,000
	Crystal Springs Mt.	2430DC	25,000	0.01	2,500
	Kalmoesfontein	2530BC	50,000	1.00	50,000
	Uitsoek Plantation	2530AC	50,000	0.01	100
	Three Falls Farm	2530AB	120,000	1.92	230,000
	Longtom Pass	2530AC	50,000	2.0	100,000
	Elandspruit		40,000	2.25	89,200
	Malolotja Nature Reserve, Logwaja Viewpoint, Swaziland	2631AA	20,000	1.03	20,600
	Nkomazi River	2631AA	15,000	0.05	7,500
	24 km from Josefsdal to Barberton	2531DA	50,000	4.67	207,500
	Fairview above Sheba Mine	2531CA	1,050,000	0.42	12,600
	Mac Mac Falls Nature Reserve	2530AD	240,000	0.46	110,400
	Graskopkloof	2430DD	40,000	0.25	10,000
	En route to Pilgrim's Rest from Graskop	2430DD	200,000	1.89	378,000

Name	Location / habitat	¼-degree squares	Estimated AOO (m^2)	Average per m^2	Estimated population size
	Pullen Farm	253CA	15,000	2.25	3,750
L. camerunensis			**1,995,600**		**518,196**
	Buffelskloof Stream	2530BB	10,000	0.10	1,000
	Buffelskloof tributary	2530BC	150,000	1.67	90,000
	Buffelskloof Hut	2530BB	10,000	0.15	150
	Three Falls Farm	2530AB	100,000	2.00	200,000
	Longtom Pass, 0.5 km from the turn-off to Environmental Centre	2530AC	400,000	0.01	4,000
	Sabie, 1 km towards Hazyview	2530AD	10,000	1.33	13,300
	Pilgrim's Rest	2430DD	250,000	0.17	42,500
	23 km from Barberton towards Josefsdal, stretching to 24 km from Josefsdal	2531DA	200,000	0.03	2,400
	Ridge across Nkomazi on the way to Pigg's Peak	2631AA	100,000	0.10	1,000
	Fairview above Sheba Mine nr Barberton	2531CA	40,000	0.09	3,600
	Mac Mac Falls Nature Reserve	2530AD	20,600	0.16	3,296
	Crystal Springs Mt. Along rd from Pilgrim's Rest to Lydenburg	2430DC	200,000	0.03	6,000
	Blyderivierpoort Nature Reserve	2430CD	5,000	0.19	950
	Lajuma Plateau Nature Reserve and surrounds, Soutpansberg	2329AB	500,000	0.30	150,000
L. filifolia			**1,045,000**		**313,500**
	Soutpansberg Mt., Lajuma & surrounds	2329AB	45,000	0.30	13,500
	Blouberg	2329AA	1,000,000	0.30	300,000

Name	Location / habitat	¼-degree squares	Estimated AOO (m^2)	Average per m^2	Estimated population size
S. albiflorum			**5,500**		**380**
	Buffelskloof Stream nr High Forest	2530BC	5,000	0.02	80
	Mac Mac Nature Reserve along a trail	2530AD	500	0.60	300
S. argenteus			**200**		**200**
	Mac Mac Falls Nature Reserve	2530AD	200	1.00	200
S. concinus			**11,000**		**470**
	Nr Jericho High School, Hlangano District	2731AB	10,000	0.03	300
	24 km en route to Barberton from Josefsdal Border Post	2531DA	1,000	0.17	170
S. parvifolius			**20,000**		**600**
	Blyderivierspoort Nature Reserve	2430CC	20,000	0.03	600
S. pretoriae			**45,500**		**4,950**
	Bejisa Village, Hlangano District, Hlatikulu Region	2731AC	500	3.50	1,750
	Nr Jericho High School, Hlangano Masilela Border Post rd	2731AB	6,000	0.20	120
	24 km from Josefdal to Barberton	2531DA	1,000	0.20	500
	Fairview above Sheba Mine	2531CA	35,000	0.06	2100
	Mac Mac Falls Nature Reserve along a nature trail	2530AD	3,000	0.16	480
S. rehmannii			**100**		**19**
	Buffelskloof Nature Reserve	2530BB	100	0.19	19
S. subvelutinus			**150,100**		**9,420**
	Long Tom Pass	2530AC 2530AB	150,000	0.06	9,000

Name	Location / habitat	¼-degree squares	Estimated AOO (m^2)	Average per m^2	Estimated population size
	Mac Mac Falls Nature Reserve	2530AD	100	4.20	420
S. thorncroftii			**102,550**		**32,665**
	Songimvelo Nature Reserve	2530DD	50	0.30	15
	Ridges between Graskop & Pilgrim's Rest	2430DD	100,000	0.30	30,000
	Malolotja Nature Reserve	2631AA	2,500	1.06	2,650
S. transvaalensis			**78,500**		**1460**
	Songimvelo Nature Reserve	2530DD	20,000	0.01	200
	Sabie, 13 km en route to Hazyview	2530AD	500	1.43	714
	Graskop – Pilgrim's Rest rd	2430DD	25,000	<0.01	66
	Blyderivierpoort Nature Reserve	2430CD	30,000	0.02	60
	Mlumati (Lomati) River near Lufafa Mt., Swaziland	2431DB	1,000	0.09	90
	Nkomati River, Magagu Dam, Swaziland	2631AB	1,000	0.03	300
	Peaks Timber Lodge, Phophonyane River nr Piggs Peak	2531DB	1,000	0.03	30

5.5.11 *Confirmation of the new method by between-methods comparison*

Results of the risk-predicting and IUCN SSC (2001) methods were coded and compared (Table 5.8). The coding method was as follows: First Priority (I) was considered equivalent to Critically Endangered (CR), 2nd Priority (II) to Endangered (EN) of the IUCN SSC (2001), 3rd Priority (III) to the IUCN SSC (2001) category of Vulnerable and the Lower Priority (LP, IV) to the Least Concern (LC) IUCN SSC (2001) category. The coding strategy was developed based on results of the comparison between the IUCN SSC (2001) and the Risk Predicting method, in which selected species of *Loudetia* and *Syncolostemon* within the critically endangered, endangered and vulnerable categories were assumed to be of 1st, 2nd and 3rd priority for conservation status assessments, whereas least concern species were placed in the 4th category (low risk).

Table 5.8. Conservation status of selected species of *Loudetia* and *Syncolostemon* based on the IUCN SSC (2001) method.

Species	Estimated population size	Estimated range size	Habitat fragm.	Extreme fluctuation	Conservation status
L. filifolia					
	150,000	<20,000 km²	Severe	Unknown	VU:B1ac(iii,iv)
L. camerunensis	>1,000,000	>20,000 km²	Severe	Unknown	NT
L. simplex	>1,000,000	>20,000 km²	Severe	Unknown	NT
S. rehmannii	<50	<10 km²	Severe	Inferred	CR: B1ac(i-ii,iv) + 2ac(i-ii,iv);D
S. albiflorum	380 counted	<10 km²	Severe	Unknown	VU: D
S. concinnus	About 430	<1 km²	Severe	Unknown	VU: D
S. pretoriae	5,100	>7 km²	Severe	Unknown	NT
S. subvelutinus	10,000	>3 km²	Severe	Unknown	NT
S. thorncroftii	10,000	>8 km²	Severe	Unknown	NT
S. transvaalensis	>1000	>7 km²	Severe	Unknown	NT

Table 5.9. Comparisons between the risk-predicting and IUCN SSC (2001) methods.

Species	Risk-predicting method		IUCN SSC (2001) method	
	Risk status	Code	Conservation status	Code
S. concinnus	II	2	VU	3
S. subvelutinus	II	4	LC	4
L. filifolia	III	3	VU	3
S. thorncroftii	IV	4	LC	4
S. albiflorum	IV	4	VU	3
L. camerunensis	IV	4	LC	4
S. pretoriae	IV	4	LC	4
S. rehmannii	IV	4	CR	1
L. simplex	IV	4	LC	4
S. transvaalensis	IV	4	LC	4
Association	0.40		Not correlated	
r	0.40		Not correlated	
P-value	0.500 (-)		Not significant	

5.5.12 *Modification of the method*

Although there is no significant difference at 95% confidence limit between the risk predicting method and the IUCN SSC (2001) method (p = 0.500), the correlation coefficient is very low (r = 0.40, Table 5.9), indicating that the risk predicting method is not associated with the IUCN SSC (2001) method despite a lack of significant difference between the two methods (P =0.500, Table 5.9). Therefore the risk predicting method

requires modification to improve its correlation with the IUCN SSC (2001) method. Discrepancies between the risk predicting method and the IUCN SSC (2001) method can be observed in *Syncolostemon albiflorum* (LP versus Vu), *S. rehmannii* (2nd priority versus CR) and *S. subverutinus* (2nd priority versus LC; Table 5.10). Criterion A2 (Table 5.3) has therefore been modified by increasing the range for specimens collected over the past 3 decades as follows: (a) from 0, 1-12, 13- 24, ≥25 to 0-5, 6-19, 20-29 and ≥30, respectively (Table 5.11). Revised risk predictions using the revised criteria for *S. albiflorum, S. rehmannii* and *S. subverutinus* are presented result in an increase in correlation coefficient (corr. = 0.66; Table 5.12.

Table 5.10. Differences between threat assessments based on the IUCN SSC method and priority based on the risk predicting method in species of *Syncolostemon.* PR = predicted risk based on the risk predicting method, TC = threat category based on the IUCN SSC (2001) method. Parameters A1 to C are as in Table 5.3. Subscript = score per parameter based on Table 5.3.

Species	A1	A2	B1	B2	B3	C	PR	TC
S. albiflorum	30_2	17_3	7_3	5_2	13_2	1_1	IV	VU
S. rehmannii	11_1	4_2	5_2	8_1	6_1	1_1	IV	CR
S. subverutinus	4_1	1_2	2_1	10_4	1_1	1_1	II	LC

Table 5.11. Revised risk-predicting parameters derived using herbarium data based on discrepancies between the IUCN SSC (2001) method and risk predicting method.

Criteria	Considerations	Scale of risk indicator		
		1	2	3
A	Number of specimens collected during: 3. the period of high collecting intensities 4. the past 3 decades	 1-15 / decade 0-5	 16-30 / decade 6-19 / decade	 31-45 / dec 20 / 29 dec
B	4. Number of locations of population occurrence	1-2	3-5	6-10
	5. Number of empty grids separating occupied ones indicating the apparent extent of habitat fragmentation	≥6 grids	4-5 grids	2-3 grids
	6. Number of filled contiguous grids indicating the area of occupancy	1-10 grids	11-20 grids	21-40
C	2. Number of habitat types	1	2	3-4

Table 5.12. Revised risk predictions for *Syncolostemon albiflorum, S. rehmannii* and *S. subverutinus* based on Table 5.11.

Species	1	A2	B1	B2	B3	C	P	1/P	PR	Concl.
S. albiflorum	2	2	3	2	2	1	48	0.0208	IV(A1B1,3C)	LP
S. rehmannii	1	1	2	3	1	2	12	0.0833	III(A1B1,3C)	3rd PR
S. subvelutinus	1	1	1	4	1	1	4	0.2500	II(A1B1,3C)	2nd PR
Student's *t-test*	Replacing risk predictions in Table 5.9 by values in Table 5.12									0.339
Correlation	Replacing risk predictions in Table 5.9 by values in Table 5.12									0.66

5.6 Discussion

5.6.1 *Useful parameters in the risk predicting method*

Number of specimens. During the period of high collecting intensity all known species of *Loudetia* were represented in herbaria (Figure 5.5). During this period, species represented by more than 4% of the sample of herbarium specimens examined also have RPI less than 0.1000 and are considered of no priority for detailed conservation status assessments (Table 5.4; Figure 5.7; Criterion A1). On the other hand, species represented by 3% or less of the specimens examined have RPI greater than 0.1000 and they are regarded as falling within the first and second priorities for detailed conservation status assessments.

In spite of the decline in frequencies of specimen accumulations, the recurrent documentation of species during this period (Figures 5.5 & 5.6) presents a track record of their persistence in nature (Criterion A2). Reductions in the number of specimens (Figure 5.5) may indicate that range and / or population sizes are declining, but the randomness of rates of decline may be attributed to the combination of collecting efforts with other factors, including specimen storage policies. Factors which may contribute to the decline in recent collections include the loss of suitable habitats due to changes in land use systems, deforestation, over-utilization, fire and natural phenomena, all of which are known to predispose species to increased risk of extinction (Burkey, 1989; Pimm *et al.,* 1988, 1995; Myers, 1995; Liu, & Bråkenhielm, 1996; Steinlein *et al.,* 1996; Wiegand & Milton, 1996; Purvis, *et al.,* 2000; Jensen *et al.,* 2001; Mazia *et al.,* 2001; Pullin, 2002; Raven, 2002). Apparently, suitable habitats for species of *Loudetia* are diminishing due to the expansion of agriculture and plantation forestry reported by White (1983) and Bredenkamp *et al.* (1996) among other workers. Pine plantations in Mpumalanga and Swaziland and settlement in Swaziland have severely diminished and / or fragmented ranges of *Loudetia simplex* and *L. camerunensis* and of species of *Syncolostemon.* However, the random display of rates of decline in recent collections in *Loudetia* implies that herbarium data cannot be used to infer population or range reductions. In addition, herbarium specimen labels without collecting dates (Figure 5.4) may introduce errors in the comparison of the historic number and rates of decline of specimens (Criterion A; Table 5.3). The lack of indications about population sizes and reductions is consistent with data from well-collected species of *Plectranthus* from eastern and southern Africa (Willis *et al.,* 2003).

The absence of records for 10 to 30 years or 3 generations may be assumed to indicate that local extinctions have occurred (MacDougall *et al.,* 1998; IUCN SSC, 2001). The lack of species representation during the period when collections began to increase may indicate that species have gone locally extinct (MacDougall *et al.,* 1998). There were no missing species of *Loudetia* during the period of high collecting intensity, implying that species known previously had not yet gone locally extinct. However, the intermittent records for the recent 3 decades in *Loudetia pennata* and *L. tisserantii,* 2 decades in *L. angolensis* and *L. coarctata* and one decade in *L. kagerensis, L. phragmitoides* and *L. togoensis* (Figure 5.5) removes the evidence for the persistence of species from herbarium data. This lack of records may therefore indicate that there have been reductions in population and / or range sizes to the extent that collectors have not been able to spot the plants in the wild. Absence of records can also be indicative of a shift in the interest of collectors and / or specimen storage policy or that localities have become unsafe for collectors due to insecurity in some parts of Africa. Alternatively, local extinctions may have occurred at least in some locations, particularly where competitive land use systems such as agriculture and plantation forestry have been extensively implemented. However, the sudden reappearance of *L. demeusei, L. tisserantii* and *L. densispica* after not less than 10 years during which the species had not been represented in collection is indicative of difficulties in interpreting absence of records, thus casting doubt on the occurrence of local extinctions and providing a motivation for checking herbarium records with field surveys. Sometimes collections might not be immediately accessioned, causing a shift in periods of collections, but this is expected to have a negligible overall effect.

5.6.2 *Collecting localities*

Counting the number of occupied grids might give a rough, but simple indication of the range size (Schatz, 2002; Willis *et al.,* 2003). Species that are geographically restricted occupy fewer grids than extensive ones and may be predisposed to increased risks of extinction (Figure 5.1). Spatially extensive species of *Loudetia* have been considered of no priority for risk assessment, whereas restricted ones are regarded as belonging to the first, second or third priority for detailed conservation status assessments (Criterion B3; Table 5.4). The number of empty grids separating filled cells has been employed to make inferences about the degree of threat emanating from species range properties. Wide gaps between filled cells indicate that natural mechanisms of pollen transfer from anthers to stigmas between members of separate populations and seed dispersal by wind are ineffective, causing breakdowns in the flow of genes. Distances for effective gene flow to occur through pollination and seed dispersal differ among plant species and agents, but species-specific information is scanty. Wind is the agent of pollen and seed dispersal in *Loudetia* in which seeds may also be carried by animals from one place to another. The mature spikelets attach to fur and hair by calluses until they are removed, which raises a possibility of seed dispersal being aided by animals. Animal dispersal may carry seeds long distances. In an investigation of the adaptation of spikelets to wind dispersal, a desk fan set at high speed was placed one metre from the floor in the laboratory from which spikelets were blown away to within 1.5 m from the fan. This indicates short distance dispersal under normal wind speed, but whirlwinds and gales may transfer spikelets to

distant places. The pattern of distribution for species of *Loudetia* is mostly clumpy, which indicates that short distance dispersal occurs. Short distance dispersal in species of the Arundinelleae has also been reported by Stebbins (1981). Pollen could be blown across longer distances because it is lighter than spikelets. It is assumed that pollen transfer by wind across 24 km (approximate distance across the ¼-degree-square grids used) might be ineffective. Therefore a single unfilled grid may indicate separate populations. However, allowing for uncertainties in the herbarium data and a possibility of long distance dispersal accomplished by animals and high speed winds, more than one unfilled grid has been considered a minimum approximation of the distance between independent populations (Criterion B1 & B2).

Recording the latitude and longitude of collecting localities on specimen labels has become common only recently (from 1970s) with the use of GPS's. Maps accompanying a few labels dated earlier than 1970s simplify the task of obtaining latitudes and longitudes of collecting localities that have been illustrated. However, most old specimens often do not have latitude and longitude records, necessitating the use of gazetteers. Latitudes and longitudes obtained from gazetteers do not represent precise localities because collecting localities are often referenced to the nearest named geographical features (Rhoads & Thompson, 1992; MacDougall *et al.,* 1998; Willis *et al.,* 2003). Imprecise localities and untraceable geo-references are common in specimen labels of old collections of *Loudetia* (Figure 5.4). The resultant plots might distort the spatial pattern of species distributions in *Loudetia* by having unfilled grids where the species has been documented and filling the grids that only represent the referenced geographic feature rather than collecting localities. In turn, herbarium records may display incorrect indications of the size and number of locations (Criterion B). In addition, locality data based on cumulating records of specimens may swamp recent changes in range sizes because points generated from them represent fixed historic records, making recent habitat fragmentations and range expansion or shrinkage indiscernible.

Temporal and spatial fluctuations in the number of herbarium records (Figures 5.5 & 5.8) may be attributable to various reasons associated with research and collector interest. For example, unusually low densities of points in *L. camerunensis* (see Chapter 3) may be attributed to the political turmoil which persisted during the period of increased collecting intensities (Figure 5.5) in some parts of Africa, including Mozambique and Angola, but some records from Tanzania may have been lost when the museum in Berlin was bombed during the Second World War. The generally more sparse distribution of filled grids north of 9°S may indicate differences in collecting intensities across Africa, in which collecting intensities are higher southwards than northwards, but it could also be due to limited access to herbarium specimens from that part of Africa or both. On the other hand, the small proportion of specimens of *L. filifolia* (Figure 5.7) may reflect difficulties associated with access to mountainous environments to which the species is restricted. Its diminutive size may also elude the attention of non-specific collectors. For example, a farmer at Lajuma Plateau, Soutpansberg guided the location of *Loudetia camerunensis,* but he had not noticed the occurrence of *L. filifolia* growing less than 30 m away – on low-lying rock outcrops by the roadside in his farm. This dependence of the accumulation

of herbarium specimens on extrinsic factors thus introduces uncertainties in interpreting the number of localities and habitat fragmentation inferred from herbarium speciemens.

One of the factors that are not readily discernible in herbarium specimens is the abundance level within a particular spatial pattern of species distribution. The information conveyed by distribution maps can thus easily be interpreted as if a particular species is evenly distributed over its entire range. However, some species are rare along the entire or part of their ranges. For example, *Pelargonium tomentosa* is rare on Hottentots Holland Mountains and abundant on the Langeberg range in South Africa (van der Walt, 1981). *Pelargonium pillansii* has a wide distribution in the Western Cape Region, but it is rare in the vicinity of the Cape Peninsula, which may indicate that a species has reached the periphery of its distribution area (Maggs *et al.,* 1999). The change from abundant to rare along the range of a particular species may have a profound effect on the gene pool of the population and consequently the choice of a site for conservation efforts. On the other hand, some species may have restricted distributions, but face no immediate threat. These may not be priority species for conservation considerations, but this knowledge can only be obtained through field surveys.

5.6.3 *Habitat types*

An assumption was made that species occurring in more than one habitat type are more likely to survive localized stochastic events or habitat-specific anthropogenic pressure than restricted ones (Figure 5.1; Criterion C). However, the assessment of the vulnerability of species occupying more than one habitat type is confounded by the limited knowledge about habitat specificity, rate of destruction and the variation of human disturbance in position, frequency, intensity and extent (White, 1983; MacDougall *et al.,* 1998).

Habitat types are not always recorded on specimen labels and, where the information is available, terms used are not standardized. The absence of reliable data on habitat types and / or confusing terms used to describe habitat types by different collectors make the discerning of affinities of species with the vegetation and habitat types difficult. Obtaining information on habitat types from vegetation maps is only informative when mapping scales are small. Large scales tend to obscure vegetation types of limited extent, with which species of *Loudetia* may be associated. For example, *Loudetia camerunensis* grows mainly in low-lying and relatively poor drainage areas, with *L. simplex* occurring in shallow, stony soils. Such habitat preferences are obscured on distribution maps that incorrectly illustrate these species as occurring sympatrically.

5.6.4 *Within- and between-method comparisons*

The low associations and correlation coefficients between pairs of criteria and sub-criteria (Table 5.5) imply that risks perceived to predispose species to extinction do not necessarily have to be inter-dependent. Therefore, each parameter may be sensitive to a particular stress independent of other parameters. For example, *Loudetia annua* has fewer specimens than *L. phragmitoides* (Figure 5.7) and is assigned a higher risk indicator than the latter with respect to Criterion A1 (Table 5.4). Since *L. phragmitoides* has not been represented in recent collections whereas *L. annua* is represented (Figure 5.5), it is

assigned a higher risk indicator than the latter with respect to Criterion A2. Similar trends can be seen in *Syncolostemon* (Table 5.4) in which most species appear to be confined to one location (high risk indicator with respect to the number of locations; Criterion B1) with no evidence of range fragmentation (low risk indicator with respect to Criterion B2). Because of these discordances, each of the criteria may perform significantly differently to the predicted risk (Tables 5.4 & 5.5). One of the effects of independent risk indicators may be smothering the predicted risk in the case of a species that should have been considered to be of high priority, but advantages include increasing the sphere for capturing stresses, which might otherwise be obscured. Listing these stresses along with the predicted risk may help if it is necessary to verify them in the field.

The low association implies that there are disparities between the risk-predicting and IUCN SSC (2001) methods, although there is no significant difference between results obtained by either method ($P = 0.296$; Table 5.9). The main sources of error include the geographical distribution and size of the sample, unsystematic collections in herbarium data already referred to and problems in determining cut-off values between categories within each, but especially in the risk-predicting method. It is imperative that samples of herbarium specimens be representative of species distributions to avoid masking the number of locations and relative range size, which may affect the predicted status of the risk. For example, *Syncolostemon subvelutinus* is not perceived to be threatened using the IUCN SSC (2001) methods, but it is under-represented in herbarium data, resulting in its placement within the second priority for detailed conservation status assessment. On the other hand, the critically endangered *S. rehmannii* is well-represented in herbaria and would appear to fall within the low priority (Table 5.10). Many species are however under-represented in herbarium records and may appear to be of high priority for risk assessment. Over-representation in herbarium records may be misleading when compiling a list of species requiring further studies and eventually when setting priorities for conservation efforts. On the other hand, under-representation may be favoured as precautionary, but it might increase the number of species earmarked for detailed field assessments, thereby increasing the time and expenses required to complete detailed assessment exercises. The risk-predicting method has, however, been sensitive in discerning species for which detailed assessments may not be necessary because they are considered not threatened using the IUCN SSC (2001) method.

5.6.5 *Linkage between herbarium collections and field data*

Area of occupancy. The extent of occurrence and area of occupancy (AOO) for species can be estimated by joining at least three non-linear points on a map using GIS and determining the area of the polygon so produced (Willis *et al.* 2003). However, densities of most species differ markedly across the landscape, often being higher in some patches within one locality than in other patches (Maggs *et al.*, 1999). The distribution of *Loudetia simplex* along stony hill slopes and *L. camerunensis* along poorly-drained sandy-loamy soils at Pullen Farm, Nelspruit already referred to (this study) implies that micro-habitats are important in determining the dispersal of a species across the landscape. Herbarium data do not capture differences in micro-habitats and therefore estimations of the extent of occurrence and area of occupancy based on polygons or a count of contiguous occupied cells (Schatz, 2000) generated by locality records cannot be

accurate. Field surveys in Mpumalanga & Limpopo provinces of South Africa and in Swaziland revealed varying estimations of AOO and average number of plants per unit area (Table 5.7), which provide reasonable estimations of the range of species. Therefore field data provided the estimation of AOO, which was useful in the testing of the risk-predicting method by using the IUCN SSC (2001) method.

5.6.6 *Status of localities*

Habitats within a given locality can be modified or lost due to agricultural activities, plantation forestry or housing developments. The extent of habitat modification, with the entire or part of the habitat modified, may affect the quality of the locality to the extent that species may be represented by fewer numbers of individuals or go locally extinct (Pimm *et al.* 1988, 1995). Herbarium data do not provide indications of any changes in the use of landscapes since the specimens were first collected. During the field survey, pine plantation in Mpumalanga, with settlement and subsistence farming in Swaziland were noted as major threats to habitats for species of *Loudetia* and *Syncolostemon,* in which localities have been diminished in size or lost. Expansion of pine plantations will result in the loss of even already fragmented habitats, which might drive species of *Loudetia* and *Syncolostemon* to local extinction. Field-based assessments of threat thus provided a parameter useful in assessing the conservation status of species using the IUCN SSC (2001) method.

5.6.7 *Requirement for modification of the new method*

Field data revealed that the risk predicted from herbarium records can be under- or over-estimated (Table 5.10). Of concern are species that are threatened, but appear to be of low priority in herbaria as in *Syncolostemon rehmannii* (Table 5.10). This implies that there are discrepancies between the risk predicting method and IUCN SSC (2001) method, resulting in low correlation coefficient (corr. = 0.40, Table5.9). The under-estimation of risk would result in incorrect listing of priorities, in which threatened species appear in under the low priority list, which might misguide conservation assessments. Adjusting classes of risk indicators to accommodate *S. rehmannii* among priority species has limitations, including the placement of most unthreatened species in the priority ranks. Therefore simulations of risk indicators are required to accommodate most of threatened species while excluding as many of unthreatened species as possible improvement.

5.7 Conclusions

The number of specimens and locations, evidence of recent collections, habitat fragmentations, range sizes and habitat types obtained or inferred from herbarium records are useful indicators of distribution patterns of species, which may be informative in predicting relative risks of species extinction. These factors are therefore useful components of the risk-predicting method using data gleaned from herbaria. Relative risks predicted from herbarium records are designed to help with making recommendations for detailed conservation status assessments. Although contradictions leading to lower predicted priorities for detailed conservation status assessments may be prevalent, the combined use of all of these risk indicators is recommended in order to ascertain that as many sources of stress as possible are captured and errors due to collection patterns are minimized. Thus, each of these indicators measures a particular

source of stresses believed to drive species towards extinction, such as small population sizes and range restrictions.

The accuracy of the risk-predicting method depends on the relationships between herbarium records on one hand and relative abundances, range sizes and patterns of species distributions in nature on the other. Thus, systematic collections are required, in which specimen collecting efforts reflect relative temporal and spatial distributions of species. However, specimen-collecting efforts are not systematic because of the combination with research biases and specimen storing policies. As a result, some rare or endangered species have been over-represented whereas those that are not threatened may be under-represented in herbaria, leading to inaccurate risk predictions. However, a preliminary test using field data of selected species of *Loudetia* and *Syncolostemon* shows some harmony between species considered to be of priority and threatened categories and those under no priority with species considered to be Least Concern using the risk-predicting and IUCN SSC (2001) methods, respectively. This agreement implies that the proposed exclusion of non-priority species from detailed assessments can be undertaken while accounting for most threatened species. In this way, expenses can be reduced by dedicating time and money required for detailed conservation status assessments only on species that have already been prioritized using herbarium records. In addition, progress in detailed conservation status assessments can be accelerated, thus providing data on threatened species to users. However, some threatened species are predicted as being of low priority, which might undermine assessments. Attempt to modify the method has improved correlation coefficient between the risk method and IUCN SSC (2001) method (corr. Coefficient = 0.66 versus 0.40). However, simulations of risk indicators are required to identify ranges that might include as many threatened species as in the priority list while placing unthreatened species in the low risk category.

While ages of collections and numbers of localities, habitat types and specimens have been used to assess rarity and the conservation status of species before (MacDougall *et al.*, 1998; Golding, 2002; Willis *et al.* 2003; Victor & Keith, 2004), using herbarium records to predict risk and therefore prioritize species for detailed conservation status assessment presents a new method. In this new method, comprehensive analyses of ages of collections and examinations of locality data and habitat types obtained from specimens belonging to species of *Loudetia* have been provided. The new method is designed to concentrate time and funding on species already prioritized for detailed assessments while avoiding inconsistencies in herbarium data by making a provision for verifying the risk with field data. The new method thus represents a departure from regarding provisional assessments based on herbarium data as final.

5.8 *Suggestions for improvement of the risk predicting method*

Aspect of change in range size. The absence of indications of changes in population or range size of species from herbarium records poses a problem in predicting risk. A species could be well represented in herbaria, giving an indication that it may be of lower priority for assessment while its population or range size is diminishing, necessitating a change in priority ranking to high priority. While assessing population size without field data may be difficult, a reasonable estimation about changes in range size can be obtained

from different sources, including GIS maps and knowledge of localities from forestry personnel or personal experiences. Incorporation of change in range size as a parameter in the method would greatly improve results, particularly for well-collected species that may be threatened by habitat distruction. In addition, simulations of classes of risk indicators are required in order to accommodate threatened species that are over-represented in herbaria.

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Appendix 5.1. Threshold values for categories of threat. After IUCN (2001). Abbreviations used: CR = critically endangered, EN = endangered and VU = vulnerable.

Criterion	Considerations	Category of threat		
		CR	EN	VU
A	Reduction in population size over 10 years or 3 generations:	≥90%	≥70%	≥50%
	1. Observed, estimated, inferred, suspected (where causes are reversible and understood and have ceased, based on			
	1. direct observation	≥80%	≥50%	≥30%
	2. an index of abundance appropriate to the taxon			
	3. a decline in area of occupancy, extent of occurrence and/or quality of habitat	≥80%	≥50%	≥30%
	4. actual or potential level of exploitation	≥80%	≥50%	≥30%
	5. the effect of introduced taxa, hybridization, competitors			
	6. Observed, estimated, inferred, suspected (where causes are not reversible or known or have not ceased) based on a-e in A1 above			
	3. Population size reduction within the next 10 years based on b-e in A1 above			
	4. Observed, estimated, inferred, projected, suspected population size reduction (past and future) where causes are not reversible or unknown or have not ceased based on a-e in A1 above			
B	Geographical range in the form of either B1 (extent of occurrence) or B2 (area of occupancy) or both			
	1. Extent of occurrence, indicating at least 2 of a-c below:	<100k m²	<5000 km²	<20000 km²
	a) Severely fragmented or known to exist at only a single location			
	b) Continuing decline, observed, inferred or projected in:			
	(i) Extent of occurrence			
	(ii) Area of occupancy			
	(iii) Area, extent and/or quality of habitat			
	(iv) Number of locations or subpopulations			
	(v) Number of mature individuals	<10 km²	<5000 km²	<2000 km²
	c) Extreme fluctuation in any of the			

Criterion	Considerations	Category of threat		
		CR	EN	VU
	following: (i) Extent of occurrence (ii) Area of occupancy (iii) Number of locations or subpopulations (iv) Number of mature individuals. 2. Estimation of area of occupancy and indicating 2 of a-c below: a. Severe fragmentation or known to exists at 1 location b. Continuing decline as in B1b c. Extreme fluctuation as in B1c			
C	Estimated population size in either: 1. Estimated continuing decline within 1 generation or 10 years (up to 100 years) 2. Continuing decline, observed, projected or inferred in numbers of mature plants and at least 1 of a-b: (a) Population structure in the form of the following: (ii) Estimation of mature plants in any subpopulation: (iii) Distribution of mature plants in 1 subpopulation (b) Extreme fluctuation in number of mature plants	<250 25% ≤50 90%	<2500 20% <250 95%	<10000 10% <1000 100%
D	Population size of mature plants estimated to number	<50	<250	<1000
E	Quantitative analysis showing the probability of extinction in the wild in 3 generations or 10 years (up to 100 years)	50%	20%	10%

Appendix 5.2. Risk-predicting parameters for species of *Loudetia* derived using herbarium data with standardised ranges. Risk indicator: 1 = 1st Priority with respect to a particular parameter, 2 = 2nd Priority, 3 = 3rd Priority and 4 Lower Priority with respect to a particular parameter. These may be equivalent to Critically Endangered, Endangered, Vulnerable and Least Concern of the IUCN SSC (2001) method, respectively. Conservation status assessment might be required for risk categories 1 to 3 and apparently no need for field-based conservation status assessments for category 4.

Criteria	Considerations	Scale of risk indicator		
		1	2	3
A	Number of specimens collected during: 5. the period of high collecting intensities 6. the past 3 decades	0.02-0.33 / decade 0.00-0.17	0.34-0.65 / decade 0.0.18-0.0.63 / decade	0.6 0.0
B	7. Number of locations of population occurrence 8. Number of empty grids separating occupied ones indicating the apparent extent of habitat fragmentation 9. Number of filled contiguous grids indicating the area of occupancy	0.09-0.18 ≥1.00 grids 0.00-0.24 grids	0.19-0.45 0.51-0.99 grids 0.25-0.49 grids	0.4 0.3 0.5
C	3. Number of habitat types	0.00-0.20	0.21-0.40	0.4

Appendix 5.3. Revised risk-predicting parameters with standardised ranges derived using herbarium data based on discrepancies between the IUCN SSC (2001) method and risk predicting method.

Criteria	Considerations	Scale of risk indicator		
		1	2	3
A	Number of specimens collected during: 7. the period of high collecting intensities 8. the past 3 decades	0.02-0.33 / decade 0	0.34-0.65 / decade 0.04-0.48 / decade	0.6 0.5
B	10. Number of locations of population occurrence	0.09-0.18	0.19-0.45	0.4
	11. Number of empty grids separating occupied ones indicating the apparent extent of habitat fragmentation	≥1.00 grids	0.51-0.99 grids	0.3
	12. Number of filled contiguous grids indicating the area of occupancy	0.00-0.24 grids	0.25-0.49 grids	0.5
C	4. Number of habitat types	0.00-0.20	0.21-0.40	0.4

Appendix 5.4. Abbreviations used in this thesis.

ABBREVIATION	FULL NAME	SOURCE
IUCN	Nature Conservancy (formally International Union for the Conservation of Nature: an organization which deals with the conservation of natural resources worldwide	IUCN SSC (2001)
SSC	Species Survival Commission: One of the specialist groups under IUCN	IUCN SSC (2001)
GPWG	Grass Phylogeny Working Group: a consortium of researchers in grass systematics	Ann. Missouri Bot. Gard. **88(3)**: 373–475 (2001)
AOO	Area of occupancy: IUCN parameter of measuring risk of extinction, referring to the area a species occupies	IUCN SSC (2001)
EOO	Extent of occurrence: IUCN parameter of measuring risk of extinction, referring to the entire geographical range a species can be found	IUCN SSC (2001)
RPI	Risk-predicting index: defined by	This thesis
LR	Lower Risk: where a species may not be in danger of extinction	This thesis
PR	Predicted risk: defined by	This thesis
A1	Criterion A(1): an element of estimating the risk of extinction	This thesis
A2	Criterion A(2): an element of estimating the risk of extinction	This thesis
B1	Criterion B(1): an element of estimating the risk of extinction	This thesis
B2	Criterion B(2): an element of estimating the risk of extinction	This thesis
B3	Criterion B(3): an element of estimating the risk of extinction	This thesis
C	Criterion C: an element of estimating the risk of extinction	This thesis

TC	Threat category: refers to a system in which species are classified as being threatened with extinction, including VU, EN & CR	This thesis
CR	Critically Endangered: a situation in which a species is at risk of extermination in the next 10 years	IUCN SSC (2001)
EN	Endangered: a situation in which a species is at risk of extermination in the next 50 years	IUCN SSC (2001)
VU	Vulnerable: a situation in which a species may be facing risk of extinction in the near future, but it does not qualify to be classified as endangered at present	IUCN SSC (2001)
LC	Least concern: A species is facing no foreseeable risk of extinction	IUCN SSC (2001)
OL	Overlapping: referring to plots of ranges of measurement data which exhibit continuous variation, indicating no statistical difference between them.	This thesis

CHAPTER 6

DISCUSSION & CONCLUSIONS

6.0 **Aim**

The aims of this thesis comprised clarifying the taxonomy of the *Loudetia simplex* complex, investigating the generic circumscriptions of *Loudetia* and *Loudetiopsis,* updating the species enumeration in *Loudetia* and formulating a new method for predicting the risk of extinction using herbarium records. A summary of the results and conclusions is presented below.

6.1 *Objective 1: To provide a taxonomic clarification of the* Loudetia simplex *complex*

Attempts have been made before to recognize distinct taxonomic groups as belonging to different species, subspecies and varieties (Stapf, 1898; Hubbard, 1934; Conert, 1957). However, these have been sunk because of the perceived morphological intergradations when the entire distribution area is considered (Clayton, 1974). The resultant group, the *L. simplex* complex, is a morphologically highly variable entity. A morphometric study was therefore conducted in order to clarify the taxonomy of this complex group. The following questions were addressed:

6.1.1 Can distinct taxonomic groups be identified within the *Loudetia simplex* complex?

6.1.2 At what taxonomic level is it appropriate to recognize such groups, if they exist?

Two distinct taxonomic groups have been identified within the *Loudetia simplex* complex (see Chapter 2). The two groups can be distinguished primarily by the presence of tubercule-based hairs on glumes and the lower lemma, but also by the shape of the apex of the lower glume and the relative lengths of the teeth of the callus of the upper floret, with teeth equal in length in the non-tuberculate form and slightly unequal in the tuberculate form – which are all stable characters. The two forms exhibit different habitat requirements, in which the tuberculate form grows in well-drained, shallow, stony soils mainly along hill slopes while the non-tuberculate form occurs in seasonally waterlogged sandy soils. Thus, *L. simplex* and *L. camerunensis* exhibit habitat specificity even where they co-occur. In addition, the flowering period is slightly different, with the tuberculate form flowering earlier. With reference to the point at which a well-defined species, *L. demeusei,* clusters in the dendrogram, the two groups can be recognized at specific level. Because the name *L. camerunensis* was the first to be applied for

the non-tuberculate form, it will be resurrected. *Loudetia simplex* is therefore restricted to the tuberculate form.

6.2 *Objective 2: To determine if* Loudetia *is monophyletic*

Tristachya pedicellata Stent was transferred to *Loudetia* (Chippindall, 1955) even though the lengths of the callus of the upper floret and glumes suggest that the species is closely related to *Tristachya.* Although the name *L. pedicellata* has not been widely applied, it has been used in southern Africa (Anderson, 1990). It was therefore necessary to clarify the taxonomic position of *L. pedicellata.*

Loudetiopsis was created partly from *Loudetia* (Conert, 1957). The genus has been controversial because of the perceived lack of distinction from *Loudetia*. In a phylogenetic hypothesis based on intuition, *Loudetiopsis* was presented as a grade within the *Loudetia* clade (Phipps, 1967), implying that the two genera were inseparable. Similar results were obtained by a phenetic analysis of the Arundinelleae, in which species of *Loudetiopsis* clustered together with species of *Loudetia* (Phipps, 1972; Clayton, 1972). Nevertheless, the generic name is still being applied to about 9 species. *Loudetia pedicellata* and all species of *Loudetiopsis* were included in a cladistic analysis based on the combined morphological and anatomical data set in an attempt to answer the following questions:

6.2.1 Is *Loudetia* monophyletic?

6.2.2 What are the taxonomic positions of *Loudetia pedicellata* and *Loudetiopsis?*

In a cladistic analysis based on the combined morphological and anatomical characters, *Loudetia pedicellata* was sister to *Tristachya bequaertii* and *T. leucothrix* while species of *Loudetiopsis* are more closely related to the *Loudetia* clade. Three species of *Loudetiopsis* do not form a clade, one species is imbedded into the *Loudetia* clade and one group comprised 6 species (see Chapter 3). This implies that *Loudetia,* as circumscribed by Hubbard (1934, 1936, 1937) is paraphyletic with the exclusion of species of *Loudetiopsis.* On the other hand, *Loudetiopsis* appear to be polyphyletic. Chippendall's (1955) inclusion of *L. pedicellata* in the genus makes *Loudetia* polyphyletic. A proposal is therefore made to transfer *L. pedicellata* to *Tristachya* and species of *Loudetiopsis* to *Loudetia.*

6.3 *Objective 3: To provide a hypothesis of species relationships based on the combined morphological and anatomical data set*

Although Hubbard (1934) arranged groups in a perceived evolutionary order, it was not until Phipps (1967) that the first explicit phylogeny, based on intuition, was published. Knowledge of the genera and their constituent species has since changed. Taxonomic changes may alter species relationships (Bisby & Nicholls, 1977; Sanderson & Donogue, 1989). In addition, species relationships are now inferred using modern cladistic methods. Thus, changes in the taxonomic grouping have necessitated that the circumscription of genera and relationships of species in *Loudetia* be tested using the modern cladistic method. By using the cladogram (see Chapter 3) this study sought to infer species relationships in *Loudetia* and answer the following question:

6.3.1 What are the species relationships as elucidated by anatomical and morphological characters?

The genus cannot be subdivided into subgenera or sections based on the structure of the cladogram because clades are weakly supported. However, a molecular study will be undertaken, results of which will be compared with the present study in order to confirm if subdividing *Loudetia* is warranted. The structure of the tree indicates that recognizing species of *Lodetiopsis* as belonging to three or more genera, including *Loudetia* (*Loudetiopsis ambiens*), will necessitate creating at least 9 genera or more from *Loudetia* (see Chapter 3). Therefore a proposal is made to transfer species of *Loudetiopsis* to *Loudetia*. The lack of internal branch support for the *Loudetia clade* is due to extensive homoplasy, which necessitates that groups are defined by a suite of characters, which, individually, are also shared with members of other clades as suggested by Phipps (1964).

6.4 *Objective 4: To infer a classification from the cladogram.*

Results of the present phylogenetic analysis were used to infer a classification scheme of *Loudetia* in an attempt to answer the following question:

6.4.1 How does a classification inferred from the cladogram compare with previous classification schemes?

The cladogram (see Chapter 3) is similar to the phylogenetic hypothesis based on intuition and a morphometric study of the Arundinelleae in suggesting that species of *Loudetia* and *Loudetiopsis* are indistinguishable (Phipps, 1967; Clayton, 1967). This agreement necessitates that the circumscription of the genus *Loudetia* be expanded to include species once treated under *Loudetiopsis.* The earlier classification recognized 5 sections: sect. (I) *Loudetia*, sect. (II) *Pleioneura*, sect. (III), *Pseudotristachya,* sect. (IV) *Paratristachya* and sect. (V) *Lophathera* (Hubbard, 1934). This classification was adopted by Phipps (1964, 1967) and

later modified by Conert (1957) and Clayton (1967). However, the subdivision of the genus into sections as previously recognized is not supported (Figures 3.14 & 3.16).

6.5 *Objective 5: To determine the phylogenetic contribution of quantitative characters in* Loudetia

Quantitative characters are easily subjected to mathematical calculations, thus, the boundary between character states can be determined objectively. Plotting ranges and standard deviations of metric characters on graphs enables non-overlapping characters to be determined objectively (Almeida & Bisby, 1984; Swiderski *et al.* 1998). A debate on whether quantitative characters should be used in cladistic analyses is inconclusive. Some workers recommend that quantitative characters should not be used in cladistic analysis because they doubt the merit of metric data in principle, especially methods of deriving discrete character states and the significance of means (Stuessy, 1979; Pimentel & Riggins, 1987; Farris, 1990; Zelditch *et al.,* 1995). However, evidence from genetic and population studies suggest that most expressions of height, weight and shapes are correlated with evolutionary transformations (Mickevich & Weller, 1990; Lawrence, 2004; Vázquez & Barkworth, 2004). Therefore ignoring quantitative characters may lead to loss of phylogenetic signal. In this study, discrete character states were defined from quantitative data by the graph method (Almeida & Bisby, 1984). The following questions were dealt with:

6.5.1 Are quantitative characters valuable in the cladistic analysis of species of *Loudetia* and *Loudetiopsis*?

6.5.2 Can determining character boundaries quantitatively shed light on whether homoplasy in *Loudetia* is due to error in character formulation and coding or the evolutionary history of the group?

Defining characters quantitatively has revealed overlapping ranges in most morphological characters in *Loudetia* (see Chapter 3) implying that the variation between character states is continuous (Swiderski *et al.* 1998). These overlapping characters can therefore be rejected because the resultant character states are regarded as being phylogenetically uninformative (Almeida & Bisby, 1984; Humphries & Funk, 1984; Baum, 1988; Seitz *et al.,* 2000). Among characters with non-overlapping ranges, only the length of the awn of the upper lemma is potentially phylogenetically informative. The small number of discrete character states from morphometric data indicates that the phylogenetic signal is stored in only very few quantitative characters in *Loudetia.* The limited number of potentially informative phylogenetic characters in *Loudetia* is consistent with the

distribution of qualitative character states in the Arundinelleae, in which most of the characters are homoplasious (see Chapter 3). Thus, error in character formulation and coding most likely has only limited effect on the level of homoplasy, while the occurrence of hybridization during the evolutionary history of the Arundinelleae, as hypothesized by Phipps (1967), and Clayton (1972) appears to be supported and needs to be investigated.

Species with overlapping ranges are placed in one class whereas ranges that are separated by a gap are perceived to belong to different classes and therefore assigned single and different ordinal codes, respectively (Almeida & Bisby, 1984). Assignment of the same and different ordinal codes assumes that distinct taxa have attained similar and different evolutionary steps, respectively (Swiderski *et al.,* 1998). However, species with ranges which do not normally overlap with each other may overlap with intermediates and therefore a similar code can be assigned (Almeida & Bisby, 1984). Figures 3.6 and 3.7 show that species of *Loudetia* with ranges which would otherwise not overlap are connected by intermediates and therefore these have been assigned the same code. Placing in the same class species which exhibit seemingly minor evolutionary steps represents the loss of phylogenetic information (Baum, 1988; Swiderski *et al.,* 1998). The loss of evolutionary signals may distort phylogenetic relationships, implying that gap-dependent methods produce coarse-grained results. Therefore there is need to improve the method of determining discrete states from quantitative characters (Liu *et al.,* 2003).

6.6 *Objective 6: To determine if morphological and anatomical data sets give similar or aberrant phylogenetic relationships*.

Studies have reported incongruent estimates of relationships between molecular and morphological data sets or between different genetic data sets (de Queiroz, 1993; Soltis & Kuzoff, 1995; Hedges & Maxson, 1996; Miyamoto, 1996; Normack & Lanteri, 1998; Wiens & Hollingsworth, 2000; Yoder *et al.,* 2001). Aberrant relationships between data sets may indicate that phylogenetic analyses based on only one data set may produce well-supported trees, but incorrect genealogical relationships (Wiens & Hollingsworth, 2000). Aberrant relationships may indicate different evolutionary pathways between data sets, including the occurrence of hybridization in which case tracing the phylogeny between lineages may be complicated. In plants, aberrant relationships between data sets may indicate the possibility that hybridization occurred during the evolution of the group (Soltis & Kuzoff 1995; Schilling & Panero, 1996). Therefore analyses were conducted on separate data sets of morphological, anatomical, leaf surface and the

combined morphological and anatomical data set in order to investigate if these data sets suggest the same or different phylogenetic relationships and to address the following question:

6.6.1 Do morphological data give the same species relationships as anatomical data in the Arundinelleae?

There were significant differences, indicating incongruence between anatomical data set and leaf surface data set, morphological data set and leaf surface data set and the combined morphological and anatomical data set and leaf surface data set. Therefore there is a possibility that hybridization has occurred in the evolution of *Loudetia* and other members of the Arundinelleae. No incongruence was detected between morphological data set and anatomical data set, implying that these data sets can be combined.

6.7 Objective 7: Investigating the effect of omitting one character at a time from the data matrix on species relationships.

It has long been recognized that changing the number of characters or character combinations alters tree topologies (Sanderson & Donoghue, 1989). The effect of changing character combinations on major genera, *Loudetia, Loudetiopsis, Danthoniopsis* and *Tristachya* was investigated by omitting one character at a time in cladistic analyses based on the combined morphological and anatomical data set. In doing so, the following question was addressed:

6.7.1 Are trees stable when the combined morphological and anatomical data set is altered in the Arundinelleae by excluding one character at a time?

Results showed that the placement of major genera in the analyses was altered, placing *Loudetia / Loudetiopsis, Danthoniopsis* or *Tristachya* on basal or terminal position between analyses without necessarily changing species relationships within each genus. This implies that a stable generic classification cannot be achieved with the combined morphological and anatomical data set used. However, the placement of major genera was similar to the effect of separate analyses based on morphological data set, anatomical data set, leaf surface data set and the combined morphological and anatomical data set. Therefore, caution must be exercised when interpreting incongruence in data sets.

6.8 *Objective 8: To estimate the age of the genus and its chaotic character state distributions from the inferred biogeographical evidence.*

The position of sister species *L. flammida* and *L. phragmitoides* on the cladogram (Chapter 3) was used to infer the estimated age of the genus. The following question was dealt with:

6.8.1 Can biogeography offer clues about the estimated age of the genus *Loudetia* and its chaotic character distributions?

The occurrence of sister species in Africa and South America can suggest one of three hypotheses, (1) ancient origin dating back to when Africa and South America were still one continent, (2) introduction by humans and (3) long distance dispersal, perhaps aided by birds (Stebbins, 1981; Goldblatt, 1993, 1994; Daniel, 1995; Balkwill & Balkwill, 1998). *Barleria oenotheroides* Dum. Cours. occurs in disturbed areas in both West Africa and Central America (Daniel, 1995; Balkwill & Balkwill, 1998). Its occurrence in disturbed areas suggests that the species has been introduced by humans (Daniel, 1995; Balkwill & Balkwill, 1998). Unlike *B. oenotheroides, L. flammida* occurs in pristine environments, which suggests that the species was unlikely introduced by humans. Therefore *L. flammida* most likely reached South America through long distance dispersal, perhaps aided by birds. Thus, more information is required to estimate the age of the genus and its chaotic character distributions.

6.9 *Objective 9: To determine the number of species in* Loudetia

The genus *Loudetia* is known from piecemeal publications (see Chapter 1) and determining the number of species is difficult. In order to compile a list of species in *Loudetia,* an enumeration of species has been provided with updated species descriptions and anidentification key (see Chapter 4). This study sought to answer the following question:

6.9.1 How many species are there in the genus?

The genus *Loudetia* is now believed to be represented by 30 species, including a previously undiscovered group recently collected from Ezemvelo Nature Reserve in South Africa and species once regarded as belonging to *Loudetiopsis,* but excluding *Loudetia pedicellata* (= *Tristachya pedicellata*).

6.10 *Objective 10: To determine useful parameters for predicting the risk of extinction using herbarium specimens.*

Numbers of herbarium records, habitat types and collecting localities have been used to provide indications of the degree of threat and / or rarity rating for a particular species (Prance, 1984; MacDougall *et al.,* 1998; Willis *et al.,* 2003; Victor & Keith, 2004). When processing data from herbaria, assumptions are made, including (1) the known collections and localities for a given species are a valid reflection of its abundance and distribution (Schatz, 2002); (2) an abundant and / or widespread species has more chance of being encountered and collected

than a rare and / or restricted one (Prance, 1984; Purvis *et al.,* 2000) and (3) a species occurring in more than one location and / or habitat type has a greater chance of surviving a localized threat than one found in a single location and / or habitat type (Prance, 1984; MacDougall *et al.,* 1998). A comprehensive herbarium collection is required to determine the spread of species. However, collecting records are not comprehensive for many species in tropical Africa (Rhoads & Thompson, 1992; Donoghue & Alverson, 2000). Collecting efforts often reflect current research interest, funding opportunities and availability of botanists among other factors. Thus, abundant and widespread species may be under-represented whereas rare and restricted ones may be over-represented in herbaria (Stern & Eriksson, 2000). This unsystematic nature of records represents a departure from the first two assumptions thereby undermining the information content of herbarium specimens with regard to the determination of the conservation status of species. In the light of this problem, it was felt that herbarium data may better be used to predict species which may require detailed conservation status assessments (see Chapter 5). This study attempts to answer the following question:

6.10.1 What parameters are useful in predicting the risk of extinction using herbarium records?

The number of specimens, age of specimens and habitat types appear to be useful in predicting which species might be threatened. Species represented by few specimens or recorded from few habitat types and those that have not been collected in the recent three decades were placed in a High Priority for detailed conservation status assessment. Species that occur widely and have been collected in the past three decades were regarded as being of Low Priority for conservation status assessment. Testing the method with field based data using the IUCN SSC (2001) method showed that species which were regarded as being of priority for detailed assessment were either endangered or vulnerable, whereas species that were regarded as being of low priority were not threatened. Therefore the new method can be used to reduce expenses and time by concentrating efforts on species that have already been prioritize.

6.11 Future studies

6.11.1 *Hybridization*

The occurrence of hybridization has been hypothesized (Clayton, 1967). To date, no test for hybridization has been conducted in the tribe. The distribution of character states, in which potential diagnostic characters shared by members of one genus are also shared

with members of other genera (Phipps, 1964), strongly indicates that hybridization has occurred during the evolutionary history of the Arundinelleae. It is believed that any cladistic analysis must be interpreted with caution if hybridization has occurred (Vázquez & Barkworth, 2004). Therefore there is need to investigate the extent of hybridization, if any, in the Arundinelleae using molecular markers or allozyme tests.

6.11.2 *Molecular phylogeny*

The hypothesis of relationships based on morphological and anatomical characters does not provide a clear indication about the basal taxa in the Arundinelleae (Figure 3.16). Therefore other sources of phylogenetic evidence, including molecules are required.

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