GENERAL INTRODUCTION

1.1 The genus *Barleria*

Barleria L. (Acanthaceae) is a large, pantropical genus of herbs and shrubs comprising, at a conservative estimate, some 300 species worldwide (Balkwill and Balkwill, 1998). There are approximately 70 species of *Barleria* in southern Africa, 65% of which are endemic to the region (Balkwill and Balkwill, 1997). The genus has been classified into two subgenera (*Barleria* and *Prionitis*) and seven sections (*Barleria*, *Chrysothrix*, *Cavirostrata*, *Fissimura*, *Stellatohirta*, *Somalia* and *Prionitis*). Three *Barleria* species belonging to Section *Barleria* and Subgenus *Barleria* have been selected for this study, *B. argillicola* Oberm. (Obermeyer, 1961), *B. greenii* Balkwill and Balkwill (Balkwill *et al*., 1990) and *B. saxatilis* Oberm. (Obermeyer, 1961). Section *Barleria* is distinguished by its conspicuously scarious calyx and axillary inflorescence based on scorpioid cymes. "This section is the most widespread in the genus, occurring almost throughout the entire geographic range of the genus. It is absent only from the New World and is only weakly represented in the Far East. It is present in all regions in Africa, particularly along the eastern part of the continent. Members of this section predominate in subtropical and temperate southern Africa and in the Zambezian region, where they account for approximately half of the total number of species of *Barleria* present" (Balkwill and Balkwill, 1997).

Barleria argillicola and *B. greenii* are two rare endemics restricted to Estcourt in KwaZulu-Natal. They are sympatric although ecologically separated, occurring on different soil types, and display different population structure. In contrast, *B. saxatilis* is a woody scrambling herb or suffrutex, widespread in dry hot areas of KwaZulu-Natal, Mpumalanga, Limpopo and Swaziland bushveld in sandy, stony soil on moderate south-west facing slopes.

A considerable amount of research has been undertaken on the generic limits (Balkwill, 1993), generic relationships (Balkwill, 1993), infra-generic classification (Balkwill and Balkwill, 1997) and species limits of the genus using morphological data (Makholela, 2000) and isozymes (Otto, 1994, Van der Bank *et al*., 2000). A formal taxonomic revision of the group in South Africa has not been published since the account by Obermeyer (1933), although some new species have been described (Balkwill *et al*., 1990; 1992). Considering the members of the genus *Barleria* as a whole nothing further is known about the biology of the species in this genus. It is for this reason that the red data re-assessment, pollination biology, effects of various management strategies and population genetic structure using allozymes will be studied in selected species in the genus. These will provide insights into the conservation and management of these species since effective management and conservation of rare and threatened species requires understanding of population and genetic structure.

1.2 Conservation and rarity

Rare plants are especially vulnerable to environmental and demographic events, and hence extinction (Lande, 1995). Potential causes of rarity may be small population size, habitat specificity, and/or narrow endemicity (Rabinowitz *et al*., 1986; Prober and Austin, 1990). Knowledge of abundance, spatial distribution patterns, breeding biology, genetics and the effect of management strategies on *B. argillicola* and *B. greenii* might aid in determining the potential causes of rarity in these species and facilitate good management and conservation.

It may be very difficult to devise a viable management plan without fundamental knowledge of the number of populations, number of individuals per population, area of occupancy and extent of occurrence in both *B. argillicola* and *B. greenii*. This information is especially crucial for the informed management of threatened species, in part by providing a continuous update of their population sizes (Menges, 1986; Owen and Rosentreter, 1992; Menges and Gordon, 1996). Through this study, the biology of *B. argillicola* and *B. greenii* will be placed in the context of the population behaviour of other species. By doing so, they contribute to knowledge capital in the dynamics of wild populations and can therefore strengthen the basis for generalisation needed in management for the conservation of all native biota (Keith, 2002). *Barleria argillicola* is listed as data deficient while *B. greenii* is listed as vulnerable (Hilton-Taylor, 1996). However, area of occupancy and the extent of occurrence were not known hence necessitating re-assessment of the red data status of these species.

1.3 Pollination biology, breeding systems and genetic structure

Pollination biology, breeding systems and genetic structure are common concerns when considering threats to conservation of rare species. Genetic or ecological factors can influence reproductive success at any or all stages of plant reproduction: pollination, fertilization and seed maturation. Plant breeding system has a major effect on genetic variation and genetic structure within and among populations (Rymer *et al*., 2002). Outcrossing species tend to be more genetically variable and have less genetic differentiation among populations, whereas selfing species are less genetically variable, but have more local genetic differentiation and divergence among populations (Hamrick and Godt, 1996). Breeding systems in plants directly affect gene flow, through pollen movements and pollen specificity. Plant breeding systems may be obligate selfing, facultative selfing, or out-crossing with various levels of self-incompatibility. Theoretically, species will be predominantly outcrossing or selfing to avoid inbreeding and outcrossing depression, respectively. However, many plant species have a facultative breeding system (Schemske and Lande, 1985; Lundvuist, 1991). The type of breeding system is not known in *B. argillicola* and *B. greenii*.

Plant breeding systems and pollinator behaviour are influenced by a variety of plant traits, including floral morphology and phenology, self-incompatibility and inflorescence architecture (Wyatt, 1982; Richards, 1986; Harder and Barrett, 1996). In turn, the breeding system is a primary determinant of plant population structure. Inbreeding species are expected to have less genetic diversity and heterozygosity within populations and more genetic differentiation among populations than outcrossing species (Wright, 1921; 1951; Allard *et al*., 1968; Jain, 1976). Surveys comparing allozyme and quantitative genetic variation across a wide range of taxa generally support these theoretical predictions (Brown, 1979; Schoen and Brown, 1991; Charlesworth and Charlesworth, 1995; Hamrick and Godt, 1996). However, interspecific comparisons of many unrelated taxa may be confounded by the effects of correlated ecological traits or phylogeny in producing the observed pattern of genetic structure (Williams *et al*., 2001). In addition, comparisons of rare vs. widespread species indicate that measures of genetic diversity are strongly correlated between congeneric species (Gitzendanner and Soltis, 2000). Comparative studies of closely related taxa have the advantage of being able to better isolate the effects of variation in single traits. To this end, numerous intrageneric and intraspecific comparisons have examined the relationship between the breeding system and allozyme diversity and heterozygosity within populations (Williams *et al*., 2001). It is for this reason that intraspecific allozyme comparison was made between the two endemics (*B. argillicola* and *B. greenii*) and their widespread congener (*B. saxatilis*) in this study.

In conservation biology, ecological factors, including the availability of appropriate habitat, intra and inter-specific competition, herbivory and seed predation, and the availability of mutualists such as pollinators and seed dispersers, are generally viewed as most critical in regulating short term population survival rates (Aigner, 2004). On the other hand genetic factors, particularly the loss of allelic diversity through random genetic drift, are considered most critical to the ability to adapt to long term environmental changes (Lande, 1988; Caro and Laurenson, 1994). A consequence of this view is that many empirical studies of rare plant species have focused on either ecological or genetic factors individually and hence have ignored the potential for ecological and genetic factors to operate jointly or interact (Schemske *et al*., 1994).

1.4 Management strategies

Fire, grazing and browsing are among important management issues. The effectiveness of fires depends on a range of factors that can be managed to achieve the most desirable result. These include the frequency and intensity of fire, the amount of fuel available and the nature of grazing/browsing both before and after burns. The frequency and intensity of biotic and abiotic disturbances strongly affect the structure of many communities (Sousa, 1984; Petraitis *et al*., 1989; Hobbs and Hueneke, 1992). Despite the relatively well-known effects of different management regimes and disturbance on community organisation and species diversity (Bullock *et al.*, 2001; Huber 1994; Schlapfer *et al*., 1998; Landsberg *et al*., 2003), their effects on individual plant populations have received far less attention (Byrs *et al*., 2004). Both rare

Barleria species (*B. argillicola* and *B. greenii*) are found on privately owned land and in the Weenen Game Reserve. In the former area there is cattle grazing and a two year burning cycle whereas, in the reserve, grazing pressure is very low and there is a four year burning cycle. However, the management regime that better suits the long term fitness and thus conservation of these species is unknown.

1.5 Aim

The aim of the proposed research will be to elucidate the population structures of the rare and locally endemic taxa by re-assessing their Red Data Status, establishing their pollination and breeding biology and determining their population genetic structure using allozymes, comparing their population genetic structures with that of a more widespread congener. The management strategy appropriate for each of the two endemics will also be determined. This will be achieved by addressing the following subsidiary aims:

1. To establish the pollinating agents of the endemic species;
2. To determine the nature of the breeding system expressed by the two endemics;
3. To determine the reproductive capacity of the two endemic species;
4. To determine the population genetic structure of the two endemic species and a congener;
5. To establish whether there are differences in population genetic structure and breeding systems between a widespread and two closely related rare and locally endemic species and assess whether these factors might be responsible for rarity; and
6. To determine which management strategy, in terms of fire regime and grazing level is most beneficial for each of the endemics.

1.6 Research questions

To achieve these aims the following research questions have been addressed:

1. What organisms are visiting the flowers of the two rare and endemic species? Do their activities bring them in contact with anthers and stigmas?
2. What is the reward and at which time of the day is the reward produced?
3. Are the plants self-compatible or not? If not, could this be attributed to pollen viability, incompatible pollen deposition or stigma receptivity?
4. How much fruit set takes place when pollinators are excluded, i.e. does pollinator visitation increase fruit set?
5. What are the levels of inbreeding and outbreeding?
6. What extent and pattern of gene flow exists between populations?
7. Do the results show any significant difference in levels of inbreeding and gene flow between rare and localized versus common and widespread species?
8. Is there any association between geographical distance and gene flow?

9. Do the two restricted endemics do well under frequent/infrequent fire regimes and light/moderate grazing?

1.7 Structure of the thesis

The thesis comprises seven chapters. The first chapter presents the background to the study and outlines the genus *Barleria*, conservation and rarity, pollination, breeding biology and genetic structure, management strategies, aims and research questions. Subsequent chapters each address a specified goal with Chapters 2 and 5 already published and Chapters 3 and 4 being drafts to be submitted for publication. The final chapter is the general discussion and conclusion. The conclusion considers the results in the light of the original aim of the thesis.

References

Aigner P. A. (2004). Ecological and genetic effects on demographic processes: pollination, clonality and seed production in *Dithyrea maritima*. Biological Conservation 116: 27-34.

Allard R. W., Jain S. K., Workman P. L. (1968). The genetics of inbreeding populations. Advances in Genetics 14: 55-131.

Balkwill M. J. (1993). A reappraisal of the generic limits and subdivision of *Barleria* L. (Acanthaceae). PhD thesis. University of the Witwatersrand. Johannesburg.

Balkwill M. J., Balkwill K. (1997). Delimitation and infrageneric classification of of *Barleria* (Acanthaceae). Kew Bulletin 52(3): 535-573.

Balkwill M. J., Balkwill K. (1998). A preliminary analysis of distribution patterns in a large pantropical genus, *Barleria* L. (Acanthaceae). Journal of Biogeography 25: 95-110.

Balkwill M. J., Balkwill K., Vincent P. L. D. (1990). Systematic studies in the Acanthaceae: A new species of *Barleria* from Natal. South African Journal of Botany 56(5): 571-576.

Balkwill M. J., Stalmans M., Balkwill K. (1992). Systematic studies in the Acanthaceae: A new species of *Barleria* from the eastern Transvaal, with some notes on its ecology. South African Journal of Botany 58(4): 286-291.

Brown A. H. D. (1979). Enzyme polymorphism in plant populations. Theoretical Population Biology 15: 1-42.

Bullock J. M., Franklin J., Stevenson M. J., Silvertown J., Coulson S. J., Gregory S. J., Tofts R. (2001). A plant trait analysis of responses to grazing in a long term experiment. Journal of Applied Ecology 38: 253-267.

Byrs R., Jacquemyn H., Endels P., De Blust G., Hermy M. (2004). The effects of grassland management on plant performance and demography in the perennial herb *Primula veris*. Journal of Applied Ecology 41: 1080-1091.

Caro T. M., Laurenson M. K. (1994). Ecological and genetic factors in conservation: a cautionary tale. Science 263: 485-486.

Charlesworth D., Charlesworth B. (1995). Quantitative genetics in plants: the effect of breeding system on genetic viability. Evolution 49: 911-920.

Gitzendanner M. A., Soltis P. S. (2000). Patterns of genetic variation in rare and widespread plant congeners. American Journal of Botany 87: 783-792.

Hamrick J. L., Godt M. J. (1996). Effects of life history traits on genetic diversity in plant species. Philosophical Transactions of the Royal Society: Biological Sciences 351: 1291-1298.

Harder L. D., Barrett S. C. H. (1996). Pollen dispersal and breeding patterns in animal pollinated plants. In: Lloyd D. G., Barrett S. C. H. (Eds.), Floral biology: studies on floral evolution in animal pollinated plants. Chapman and Hall, New York, USA.

Hilton-Taylor C. (1996). Red data list of southern African plants. National Botanical Institute, Pretoria.

Hobbs R. J., Hueneke L. F. (1992). Disturbance, diversity and invasion: implications for conservation. Conservation Biology 6: 324-337.

Huber R. (1994). Changes in plant species-richness in a calcareous grassland following changes in environmental conditions. Folia Geobotanica 29: 469-482.

Jain S. K. (1976). The evolution of inbreeding in plants. Annual Review of Ecology and Systematics 7: 469-495.

Keith D. A. (2002). Population dynamics of an endangered heathland shrub, *Epacris stuartii* (Epacridaceae): Recruitment, establishment and survival. Austral Ecology 27: 67-76.

Lande R. (1988). Genetics and demography in biological conservation. Science 241: 1455-1460.

Lande R. (1995). Mutation and conservation. Conservation Biology 9: 782-791.

Landsberg J., James C. D., Morton S. R., Muller W. J., Stol J. (2003). Abundance and composition of plant species along grazing gradients in Australian rangelands. Journal of Applied Ecology 40: 1008-1024.

Lundqvist A. (1991). The self-incompatibility system in *Lotus tenuis* (Fabaceae). Heriditas 119: 59-66.

Makholela T. M. (2000). Variation in the *Barleria affinis* species complex. Unpublished Honours dissertation, University of the Witwatersrand, Johannesburg.

Menges E. S. (1986). Predicting the future of rare plant populations: Demographic monitoring and modelling. Natural Areas Journal 6: 13-26.

Menges E. S., Gordon D. R. (1996). Three levels of monitoring intensity for rare plant species. Natural Areas Journal 16: 227-237.

Obermeyer A. A. (1933). A revision of the South African species of *Barleria*. Annals of Transvaal Museum 15(2): 123-180.

Obermeyer A. A. (1961). Notes and records of African Plants. In: Dyer R. A. (Ed.) Bothalia. Government Printer, Pretoria.

Otto G. P. (1994). Morphological, karyological and electrophoretic studies on the hybrid origin of a new species of *Barleria* L. Unpublished Honours dissertation, University of the Witwatersrand, Johannesburg.

Owen W. R., Rosentreter R. (1992). Monitoring rare perennial plants: Techniques for demographic studies. Natural Areas Journal 12: 32-38.

Petraitis P. S., Latham R. E., Niesenbaum R. A. (1989). The maintenance of species diversity by disturbance. Quartenary Review of Biology 64: 393-418.

Prober S., Austin M. P. (1990). Habitat peculiarity as a cause of rarity in *Eucalyptus paliformis*. Australian Journal of Ecology 16: 189-205.

Rabinowitz D., Cairns S., Dillion T. (1986). Seven forms of rarity and their frequency in the flora of the British Isles. In: Soule M. E. (Ed.), Conservation biology: the science of scarcity and diversity. Sinauer, Sunderland, Massachussets.

Richards A. J. (1986). Plant breeding systems. Allen and Unwin, London, UK.

Rymer P. D., Morris E. C., Richardson B. J. (2002). Breeding and population genetics of the vulnerable plant *Dillwynia tenuifolia* (Fabaceae). Austral Ecology 27: 241-248.

Schemske D. W., Husband B. C., Ruckelshaus M. H., Goodwillie C., Parker I. M., Bishop J. G. (1994). Evaluating approaches to the conservation of rare endangered plants. Ecology 75: 584-606.

Schemske D. W., Lande R. (1985). The evolution of self-fertilization and inbreeding depression in plants ii. Empirical observations. Evolution 39: 41-52.

Schlapfer M., Zoller H., Korner C. (1998). Influences of mowing and grazing on plant species composition in calcareous grassland. Botanica Helvetica 108: 57-76.

Schoen D. J., Brown A. D. H. (1991). Intraspecific variation in population gene diversity and effective population size correlates with breeding system in plants. Proceedings of the National Academy of Sciences of the United States of America 88: 4494-4497.

Sousa W. P. (1984). The role of disturbance in natural communities. Annual Review of Ecology and Systematics 15: 353-391.

Van der Bank F.H., Van der Bank M., Balkwill M.J., Balkwill K. (2000). Electrophoretic evidence for an undescribed species of *Barleria* L. South African Journal of Botany 66(1): 22-27.

Williams C. F., Ruvinsky J., Scott P. E., Hews D. K. (2001). Pollination, breeding system and genetic structure in two sympatric *Delphinium* (Ranunculaceae) species. American Journal of Botany 88: 1623-1633.

Wright S. (1921). Systems of breeding. Genetics 6: 111-178.

Wright S. (1951). The genetical structure of populations. Annals of Eugenics 15: 323-354.

Wyatt R. (1982). Inflorescence architecture: how flower number, arrangement, and phenology affect pollination and fruit-set. American Journal of Botany 69: 585-594.