

A Handbook for Botanic Gardens on the Reintroduction of Plants to the Wild




Botanic Gardens Conservation International

RTZ

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**A Handbook for Botanic Gardens on the
Reintroduction of Plants to the Wild**

Compiled by
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Prepared by Botanic Gardens Conservation
International, in association with IUCN Species
Survival Commission (Reintroductions Specialist
Group)

Cover Photo
Serpentine vegetation reconstructed within the
Jardín Botánico Nacional de La Habana, Cuba.
Peter Wyse Jackson

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Preface

The increasing loss of plant diversity worldwide has made the development of a variety of new habitat restoration techniques urgent. The successful conservation of many plant communities, plant species and populations will, in future, require increasingly active management. Very few natural areas remain that are not subject to increasing human pressures. The introduction and reintroduction of species to the wild, especially those that have become rare and endangered, has become an important tool for the conservation of biodiversity. Although a large number of institutions in many countries are becoming active in this field, there are few adequate manuals available to guide their work. Consequently it seems that many unsuccessful, ill-advised or uncontrolled plant reintroductions are taking place. This booklet is a first attempt to explore the issues and challenges facing botanic gardens in this field and to provide them with some guidance in activities for which their expertise is increasingly being sought and their living plant collections are being used more and more.

In 1991, Botanic Gardens Conservation International (BGCI) began the preparation of this handbook for botanic gardens. The first draft arose from a meeting on 9-11 April 1991 held in Las Palmas de Gran Canaria, Spain. There BGCI and the Jardín Botánico Canario hosted an international workshop on the special role of botanic gardens in plant reintroductions and habitat restoration. The document was reviewed at a second international workshop on 6-7 September 1991 at the Royal Botanic Gardens, Kew, U.K. The workshop was organized jointly by BGCI, the Reintroductions Specialist Group of the IUCN Species Survival Commission and the Royal Botanic Gardens, Kew, following a conference entitled "From specimen to habitat management" held on September 1-5, 1991. A third workshop to continue work on the project was held in Rio de Janeiro, Brazil, during the 3rd International Botanic Gardens Conservation Congress organized by BGCI and the Jardim

Botânico do Rio de Janeiro '19-25 October 1992'.

Following these workshops' John Akeroyd and Peter Wyse Jackson undertook the task of incorporating the many comments, ideas and information into the present published form.

We are grateful to all those, especially those listed below, who contributed text, gave many suggestions, advice and assistance in the preparation of this booklet: - David Bramwell (Las Palmas, Spain), John Clayton (Palabora, South Africa), Margarita Clemente Muñoz (Córdoba, Spain), Quentin Cronk (Edinburgh, U.K.), Donald A. Falk (Tucson, U.S.), Alan Hamilton (WWF, U.K.), Dorothy Harris (RTZ, U.K.), Esteban Hernández-Bermejo (Córdoba, Spain), Vernon Heywood (Reading, U.K.), Ailene Isaf (BGCI), Manfred Jusaitis (Adelaide, Australia), Angela Leiva (Havana, Cuba), Len E. Marrs (Utah, U.S.) Mike Maunder (Kew, U.K.), Brien Meilleur (St Louis, U.S.), Louis Olivier (Porquerolles, France), Esperanza Peña Carofa (Havana, Cuba), Tânia Sampaio Pereira (Rio de Janeiro, Brazil), Alan Pottinger (Oxford, U.K.), Tammera Race (Florida, U.S.), Mark Richardson (Canberra, Australia), Rebecca Rowe (Oxford, U.K.) and Susan Wallace (Florida, U.S.).

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We hope that this Handbook will be a useful and handy reference text for those working in plant conservation. We welcome feedback from users to help us improve the Handbook for future editions.

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Introduction

Throughout the world, wild plants and their habitats are under increasing threat. Clearly, the conservation of habitats and species *in situ* must be seen to be preferable to *ex situ* measures, but the scale of habitat destruction in many regions has too often denied or reduced the option of extensive *in situ* conservation. Thus the reintroduction of individual species into protected sites in the wild (i.e. native or semi-natural habitats) and, in some cases, the restoration or reconstruction of whole communities, will in the future become essential measures to conserve threatened plants. Maunder (1992) has reviewed this new science of plant reintroduction and habitat restoration, citing published accounts of the relatively few plant reintroductions that have been carried out so far.

There are many causes of depletion and extinction of plants. These have been summarized by Frankel & Soulé (1981), who have emphasized the importance of human impact and reproductive isolation in extinction. Causes include biotic factors such as competition, predation and disease; spatial or reproductive isolation, leading to genetic erosion or drift; and major habitat changes as a result of geological processes, climatic change, natural catastrophe or human activity.

Reintroduction as a means of restoring or increasing the viability of plant populations is not an easy exercise. The procedures are long-term, time-consuming and expensive, and, if their application is to be considered as a valid conservation exercise, they require rigorous control, with the establishment of strict guidelines and appropriate national legislation.

It should be noted at the outset that it is better to conserve existing plant and animal communities *in situ* than to try to recreate them. The ability to reintroduce species and to restore habitats should be used only to protect or enhance diversity. It must never be allowed to provide an excuse to hasten the

destruction of natural habitats or populations of plants or animals.

The reintroduction of plant species to the wild will become an increasingly important tool for the conservation and management of natural and semi-natural habitats and wild populations of plants. There has been some reluctance among botanists to embrace the concept of reintroduction, often on spurious grounds of 'authenticity', although zoologists have achieved successful results in this field, especially with some birds and larger mammals. As wild habitats come under even greater human pressure, the implementation of restocking, reintroduction and restoration programmes will be seen to be both an essential management procedure and a respectable scientific technique.

1.1. Definitions of terms

Definitions of **Introduction**, **Reintroduction** and related terms are given on page 7.

A variety of **Reintroduction** or **Restitution** techniques exist that may be used to restore or establish populations of plants to enable them to become self-sustaining in natural habitats. Such techniques may include direct sowing of seed or the planting of material raised in nurseries, or producing conditions that will encourage the growth of seed already existing in the soil or introduced by birds and other natural means.

These definitions of procedures do not describe the full range of biological or technical considerations necessary for their proper and successful application. For instance, practitioners of **Reinforcement** will have to take into account the geographical origin and genetic make-up of the source material. Botanic gardens and their collaborators should carefully determine the proper

- strategy and procedure from a biological perspective.

The ultimate objective of these restitution techniques is to establish or reinforce viable, self-maintaining populations that will persist for an ecologically significant period of time. The placement of plant material into the landscape is thus undertaken with the goal of integrating these plants and their progeny into a functioning ecosystem. It should be noted that a project may involve the application of several of the procedures. For example, consider a situation commonly encountered in which an existing plant population has declined to an unsustainable fragment. A conservation plan may involve bringing plant material from another site to augment the existing population (**Reinforcement**), restoring its original extent (**Re-Establishment**), and establishing plants in appropriate habitats not known to have been previously colonized (**Introduction**).

Civil engineers and landscape architects increasingly use native plant species for practical, amenity and

- aesthetic planting, especially in Europe, North America and Australasia. Their methods are analogous to those of the conservationist, but they are in general much less particular about the identity and provenance of plant material. Frequently sites for **Revegetation** or **Reclamation** are urban and industrial wasteland or spoil-tips, or landscaped road-verges, where safety and practical considerations have to take priority. In Britain, the use of the seeds of native plants is now an important component of the landscaping of new roads ([U.K.] Department of Transport 1993), and on road verges and elsewhere there exist extensive artificial plant communities analogous to former species-rich grasslands.

These revegetated and reclaimed communities are not designed to match existing wild habitats. Although they do promote some biological diversity, they may pose a threat to native and semi-natural plant communities via competition and hybridization between introduced and native plants. More rigorous planning and selection of material would make such planting schemes important and



*Serpentine
vegetation
reconstructed
within the Jardín
Botánico Nacional
de La Habana,
Cuba.*

*Photo:
Peter Wyse Jackson*

valuable examples of **Restoration**. Such is the case in parts of the North American prairies, where grassland communities have been restored by the use of native species (Howel & Jordan 1991). The techniques of the landscaper are nevertheless often appropriate for reintroductions: for example, the use of exotics as nurse crops in the intermediate stages of a project.

'Corridors' (linear, restored or protected fragments of natural vegetation) are increasingly seen as a solution to the severe fragmentation of habitats by urbanization and intensive agriculture. They may be artificially constructed or follow existing topographical and ecological features such as rivers, streams, roads, railway embankments or shelterbelts. It has been suggested that global climatic changes may make human activity the important agent of species movement, perhaps through the construction or maintenance of corridors to connect natural vegetation fragments (Saunders & Hobbs 1991).

1.2. Integrated Conservation Strategies

The combination of the complementary approaches of *in situ* and *ex situ* conservation strategies to the protection and management of biological diversity has been termed "integrated conservation" (Falk, 1990).

Integrated conservation strategies seek:

- To combine research on individual species (autecology) with the scientific management of communities and ecosystems.
- To capture and maintain the genetic variation present in populations, including distinct ecotypic variants.
- To draw together organizations using different approaches and methods for the conservation of diversity.

As well as basic taxonomic, genetic and ecological research, an integrated conservation strategy must consider the problems of habitat acquisition and management, the legal protection of both land and threatened species, the careful maintenance of *ex situ* plant collections, and the rescue, reintroduction and restoration of threatened plants. A significant proportion of time and effort must be devoted to the active management of habitats, communities and

Definitions of terms

Introduction or Reintroduction

are general terms to describe the controlled placement of plant material into a natural or managed ecological setting. They also have stricter definitions (see below).

Introduction

Establishment of a plant in an area in which it has never been known to occur.

Reintroduction or Restitution

The release and management of a plant into an area in which it formerly occurred, but in which it is now extinct or believed to be extinct - also called **Reinstatement** or **Re-Establishment**.

Translocation

Transfer of material from one part to another of the existing range of a species, either to existing or to new sites.

Reinforcement

A measure to increase population size or diversity by adding individuals to an existing population - also called **Supplementation**, **Enhancement**, **Augmentation** or **Restocking**.

Restoration

Defined by the Society for Ecological Restoration as: "The process of intentionally altering a site to produce a specified historic ecosystem. The intent of the work is to emulate the natural structure, function, diversity and dynamics of a defined, indigenous ecological system." Substantial elements of the original habitat may persist, but **Reconstruction** may be necessary where the habitat has to be reconstituted wholly or from fragments. Where an entirely artificial community or 'facsimile' habitat is created, perhaps for practical (e.g. civil engineering) or aesthetic reasons, the terms **Revegetation** and **Reclamation** apply.

In the commercial sector, for example when land has been restored after mineral extraction, the term **Reinstatement** is used.

populations. Management practices require detailed information about species biology and sources of propagated plant material for use in any reintroduction or restoration project.

Any perceived dichotomy between *in situ* and *ex situ* plant conservation ought thus to be resolved. Integrated conservation strategies are of particular importance where the level of damage to natural and semi-natural plant communities is such that some degree of restoration is necessary, for example in many parts of Europe (Wyse Jackson & Akeroyd 1994).

1.3. The unique role of botanic gardens in plant reintroduction

For several centuries, botanic gardens and arboreta have maintained extensive *ex situ* plant collections. Over the last two decades an increasing number of botanic gardens have turned their attention to the role that these collections may play in practical conservation. Indeed, botanic gardens have become involved to a considerable degree in the conservation of plants *in situ*, through the work of their staff in plant exploration and collection, scientific research and the management of natural areas, frequently within or adjacent to the garden. Recently there has been a trend to combine these activities with their work on the conservation *ex situ* of individual species through the development of Species Recovery Programmes. These may involve plant propagation, reintroduction, restocking and relocation projects or the restoration of destroyed or degraded habitats.

Botanic gardens are uniquely placed to undertake reintroduction projects for plants. They are often the only institutions to hold adequate and accurately named collections of living plant germplasm, including garden and arboreta collections, field gene-banks, seed-banks and micropropagated material. They have the requisite infrastructure and propagation facilities, together with the horticultural and other applied scientific skills of their staff needed to undertake practical aspects of a Species Reintroduction Programme.

Furthermore, an increasing number of botanic gardens worldwide are acquiring natural areas within or adjacent to their institutions for the management and conservation of plant populations and habitats *in situ*. This gains them both resources and experience for the conservation and

management of populations of wild plants. In a 1991 survey, BGCI noted that 394 botanic gardens worldwide include areas of natural or semi-natural vegetation within their boundaries.

Another trend within the botanic garden community has been the creation of native plant nurseries, alongside important protected natural areas, to serve as both resource centres and to provide material for public education.

Whilst botanic gardens can help to alleviate the consequences of habitat destruction, by growing or maintaining plants in a variety of ways, under no circumstances should botanic gardens endorse the initiation of destructive activities. Gardens should avoid contributing to projects that encourage gratuitous habitat destruction, and they should never accept their ability or facilities to grow or reintroduce plants as justification for the destruction of any habitat or naturally occurring population of plants.

Every botanic garden should set out a statement of ethical and professional standards regarding their involvement in reintroduction activities and support for conservation of natural areas and naturally occurring populations.

Botanic gardens should commit themselves to working in close co-operation with the conservation community to ensure that any programme of local or regional reintroduction furthers the ultimate objective of conserving global biological diversity.

1.4. Relevant recommendations and sets of guidelines on reintroductions

The importance of appropriate environmental studies as the basis of reintroduction projects has been emphasised elsewhere, for example in the Council of Europe's Recommendation (R (85) 15) concerning the reintroduction of wildlife species. This document, adopted by the Committee of Ministers in September 1985, sets out guidelines, viz.:

1. Determine the causes of decline or extinction.
2. Analyse past and present ecological characteristics.
3. Submit proposals to remedy any causes of decline or extinction.

4. Delimit and indicate exactly the areas in which introductions are to be carried out.
 5. Enumerate planning, management and supervision measures to be taken before, during and after the implementation of the project.
 6. Evaluate the probability of success and the possible repercussions of reintroduction.
 7. Establish which population or which of any intraspecific taxa of the species should be reintroduced into a given area.
5. Reintroduction of species into the wild is not cheap, but may work out cheaper in the long-term than their permanent maintenance in cultivation. Reintroduction of a plant involves research, planning, aftercare and monitoring, but also envisages the ultimate survival and reproduction of the species in the wild.
 6. Reintroduction is a very positive process. Conservationists frequently emphasise the negative, highlighting loss of species and overall biodiversity. Reintroduction enhances or replaces something that had previously diminished or disappeared - an important consideration for public relations and education policy.

The IUCN Species Survival Commission (Reintroductions Specialist Group) has produced similar guidelines for plant reintroductions worldwide; the Center for Plant Conservation, St Louis, U.S.A., is developing guidelines for reintroductions in the United States.

1.5. Justification for reintroduction

Reintroduction can be a controversial step, either because of genuine fears of deleterious ecological or genetic consequences, or through dogmatic views on the authenticity of natural communities. Several arguments can be advanced in favour of reintroductions:

1. Conservation *in situ* must always be the preferred policy. Reintroduction should be seen as an acceptable compromise and safety-net.
2. There may be an identified, frequently practical, need for reintroduction by a land-management agency, especially where the species is a dominant or significant element in the habitat. Reintroduction may be part of a large-scale restoration of vegetation or habitats.
3. A successful reintroduction programme requires detailed information on the species and its ecology, providing scientific information in addition to that directly applicable to the project. The ecology of a rare species may indicate environmental conditions that show why a habitat is special, why a rare species occurs there, and why it (and perhaps other species) may have declined.
4. Reintroductions can assist in habitat stabilization and rehabilitation. The study of individual species for reintroduction focuses on all the factors that affect them and how the species interact with their habitat.

1.6. Reintroduction of trees: lessons from forestry

Foresters have applied on a large scale many of the principles of reintroduction adopted by plant conservationists.

Forestry involves the artificial management or replacement of existing plant communities, usually in order to exploit or establish an economically viable yield of timber. The conservation of natural or semi-natural stands of vegetation has often been a secondary consideration of modern forestry practice. Nevertheless, forestry has sought in some areas to restore degraded vegetation, especially where there is a need for shelter or to control soil erosion, and to conserve certain tree species or communities.

This may be achieved (Pottinger 1993) by:

- Management that favours natural regeneration of trees, based on their effective reproduction.
- Enrichment planting, with manipulation of numbers and density of particular species.

From the point of view of conservation, natural regeneration should be encouraged, but may not be feasible where the habitat has been severely degraded. Planting or replanting, perhaps as a complement to natural regeneration, produces a quicker result and enables more manipulation of the age-structure and genetic constitution of the population. It can be achieved either by the use of exotic species, sometimes as a nurse crop, or native species, when it is effectively a conservation exercise. Typically, one or several species are planted

into artificial gaps within the existing community.

Success, as in any transplant or reintroduction scheme, relies on factors such as suitable age of transplanted material, correct shading regimes, genetic provenance and constitution of the trees, and control of weeds and predators. It is expensive, but ensures a good survival rate and helps to stabilize the community. Further details of procedures and examples of forest restoration are given in Pottinger (1993). A useful technical manual for the restoration of native forest has been produced for New Zealand, but with wider practical applications (Porteous 1993).

Reintroduction of forest trees requires:

- Adequate knowledge of the ecology and genetics of the species:
 - the reasons for loss of the species at the site
 - the restoration of as much of the community, both plants and animals, as possible
 - a wide, if possible 'pre-degradation', genetic base for introduced population
 - an adequate size of population based on genetic, breeding and reproductive systems.
- Thorough knowledge of the ecology of the area, notably biotic factors and any history of management.
- Protection or isolation of the site, with long-term monitoring and aftercare.

Brazilian Atlantic Rainforest species in nursery cultivation.

Photo:
Pedro Schuback

- Appropriate modification of the site by landscaping, planting and weed control; measures to combat degradation of soil (especially in the tropics).
- Management of a successional regime in the community, perhaps using nitrogen-fixing legumes or nurse crops.
- Good propagation and nursery facilities, preferably nearby, the site to provide correct climatic regime and ease of translocation.

Foresters are particularly aware of the importance of genetic provenance. The reintroduced population should have a broad genetic base, assessed both on morphological and phenological characters and, where possible, by molecular methods. The material will preferably be of local provenance to ensure adaptation, with the use of *ex situ* stocks if necessary. The genetic history of the species, especially locally, needs to be interpreted where possible, examining population fluctuations (genetic drift) and episodes of contamination by exotic material (including any hybridization).

The involvement of local people is essential for the success of any reintroduction project. Multiple use of the site is one possibility for management.

An exemplary model of restoration of a complete forest ecosystem is the reintroduction of Bermuda Cedar (*Juniperus bermudiana*) to Nonsuch Island off Bermuda (D.B. Wingate, summarized in Pottinger, 1993). The vegetation of Bermuda has been much degraded by centuries of human activity, and this endemic species had been decimated by introduced scale-insects. The reintroduction was carried out as part of a programme of total environmental rehabilitation based on a thorough ecological study. All exotic species were removed (although two others were planted as a nurse crop), and the new population of the cedar, large and with a broad genetic base, protected by virtue of being on an island. Other native species were established before the cedar was reintroduced, a strain tolerant to scale insects was used and biological control of the insects applied. Native birds were encouraged by the provision of artificial habitats. Such a project to restore successfully a whole community provides a model for reintroduction schemes worldwide.

Restoration of the Brazilian Atlantic Rainforest

Just 2% of the original Brazilian Atlantic Rainforest remains intact. It is unlikely that urban growth on the coast will be halted, at least in the short-term, putting continued pressure on the small area of the forest that remains. Genetic erosion will increase as deforestation proceeds and populations of endangered species are reduced in number and size. Despite much scientific study of the Atlantic Rainforest in recent decades, plant collections are still not fully representative and knowledge of species ecology remains negligible. This lack of hard data impairs conservation policy, further threatening the future of the last remnants of this ecosystem.

In order to assemble the necessary knowledge as a basis for effective conservation and management policies, a joint initiative has been set up by Rio de Janeiro Botanic Garden and the private sector. Both believe that this can only be achieved through the training and establishment of a professional team, as well as the acquisition of equipment for biological studies and the development of laboratory and field techniques and methods.

This belief motivated a group of researchers to organise the Atlantic Rainforest Programme for Reforestation. This programme of twelve projects has received U.S. \$530,000 managed through the Rio de Janeiro Botanic Garden for field work, seed collection and storage, and setting up a plant nursery.

The Programme includes:

- Descriptive investigation of the floristic composition and structure of the Atlantic Rainforest, with inventories of species and their distribution in the bottomland and lowland forests near the coast and the highland forests, within the National Reserves.
- Study of plant succession at a degraded site and the influence of surviving forest fragments in this process. This first attempt to understand plant succession in the Atlantic Rainforest will point out potential species for use in reforestation programmes.
- Phenological observations that ensure seed collection at the appropriate time, promoting high germination and viability during storage.
- Animal-plant relationships (feeding, roosting and nesting behaviour), combining field observation and surveys in conjunction with local people.
- Anatomy and ecophysiology of individual tree species, to provide understanding of survival strategies in the low soil-oxygen conditions of the periodically flooded bottomland forests. This will indicate species suitable for planting on these sites.
- Propagation of selected species from seed or vegetative organs, in association with field observations to check ideal growth conditions for each species. Shade species are grown experimentally under shade nets; some sun species are also grown under shade nets which are removed gradually as the plants grow.
- Production of 36,000 saplings per year in a nursery recently established at Poá das Antas Biological Reserve (IBAMA-RJ). The nursery of the Rio de Janeiro Botanic Garden, which raises 600,000 saplings per year, will help to test over 100 species for seed and vegetative propagation.
- Laboratory seed germination tests, which have suggested that dormancy can be broken by soaking seed in warm or cold water following a period of moist heat.

These integrated studies aim to enhance knowledge of succession processes, so that these can be aided by adequate reforestation strategies at degraded sites. During the next three years, the research group will focus on a severely degraded area of low montane forest, swamp forest and grassland. These studies will give a solid scientific basis for the reforestation of National Reserves and also the management of the tree species of the Atlantic Rainforest.

Source: Tânia Sampaio Pereira, *Programa Mata Atlântica, Jardim Botânico do Rio de Janeiro, Brazil.*

Species Recovery Programmes

It is important at the outset of a species reintroduction project to develop a comprehensive, practical plan of action (Species Recovery Plan). This will cover detailed research on the taxonomy, genetics, reproduction, distribution and ecology of the species, and practical aspects of propagation, reintroduction and aftercare, together with administration, financial considerations and the allocation of staff to the project. It must be stressed that, although there are well in excess of 20,000 threatened plant species worldwide, practical and financial constraints have limited, and in the foreseeable future will continue to limit, Species Recovery Programmes to just a small proportion of them.

Microcycas calocoma
cultivated at the
National Botanic
Garden, La
Habana, Cuba

Photo:
Peter Wyse Jackson

Nevertheless, regional co-ordinated actions, assessing the highest priorities for plant conservation, may help to highlight the most urgent target species. For example, the Center for Plant Conservation in St Louis, Missouri, USA, has developed regional task forces to plan, co-ordinate and implement co-operative strategies between organizations and agencies for the conservation of plants in regions of high species diversity within the United States.

Species Recovery Programmes are being developed throughout Europe, North America and Australasia, and new individual species conservation and reintroduction projects are increasingly being initiated and implemented in developing countries.

Points to remember:

- Botanic gardens should undertake a Species Recovery Programme in co-operation with appropriate land-management agencies and conservation bodies. Whereas the botanic garden partner may initiate the programme, it should be emphasized that the primary responsibility for its implementation and long-term management should fall to the land-management agency.
- Institutions that undertake a Species Recovery Programme should recognize that such work may take many years and may be expensive. It is a fundamental requirement that they consult and work closely with other national and local organizations and institutions, such as conservation and land-management agencies, central government ministries (who may be able to provide some funding), local authorities and non-governmental organisations, at each stage in the initiation and implementation of a Species Recovery Plan. The value of a botanic garden creating formal links with National Parks and other Protected Areas in their region should be emphasized.
- In many cases, especially in tropical or other regions of high biological diversity, a pragmatic and practical



approach will have to be undertaken by the small number of institutions with the resources to devote to extensive plant conservation. They should aim to protect the greatest amount of plant diversity possible within the limits of their resources and not to feel bound by the rigid procedures and sometimes impractical demands of a formal Species Recovery Programme.

- Botanic gardens should have a clear reintroduction policy, linking projects to existing collections, research, horticultural expertise and contacts with other bodies or individuals. Species of local interest that are already grown in a botanic garden are, as a general rule, that institution's most suitable subjects for reintroduction.

A useful CSIRO text on the theory and practice of the conservation and management of endangered plants in Australia, with valuable case histories, illustrates many of the principles of Species Recovery Programmes (Cropper 1993).

2.1. Structure of Species Recovery Plan

A Species Recovery Programme needs first to be set out as a comprehensive and carefully considered Species Recovery Plan to document, rescue and restore a particular species, including an outline of steps that may be necessary to maximise, if necessary, impoverished genetic variation. The Plan will include some or all of the following headings:

1. Description of the species or taxon
2. Taxonomy, morphological and, where possible, genetic variation of the species
3. Present and past distribution, as far as is known
4. Current numbers and status
5. Population and reproductive biology/life-history
6. Habitat description and ecology
7. Limiting factors
8. Actual and potential threats
9. Conservation measures and actions required
10. Recovery objectives and scale of the project
11. Criteria to measure success of recovery
12. Implementation schedule
13. Horticultural and propagation techniques and feasibility
14. Aftercare and monitoring
15. Staff requirements and work-plan
16. Costs

It is not just the suitability of the plant and the chances of its survival, the planning and the cost of

the exercise that need to be considered. It is important to have tight project management and adequate long-term staffing. The responsibility for the various tasks should be given to individual members of staff of the institution, ensuring continuity over the whole period of the project. Staff should also be involved in collaborative effort.

2.2. Cultivation and propagation considerations

A major consideration in any Species Recovery Programme is the practical work entailed for the botanic garden.

- A botanic garden that intends to carry out reintroductions should have the necessary horticultural and nursery techniques and expertise available.
- Projects should be tailored to fit within the space available at the garden - large amounts of plant material may have to be propagated and maintained over a long period.
- Bear in mind that it may be necessary to protect plants after reintroduction and perhaps remove or reduce the original causes of rarity.
- A botanic garden should involve its horticultural staff at all stages of the project, including the preparation of a Species Recovery Plan.
- Good labelling and documentation is of special importance in reintroduction projects, because the use of mixed or unsuitable material will jeopardise or at least diminish the value of the project and may effect the genetic integrity of existing populations.
- Staff should be available at all stages of the work to carry out practical aspects of introduction: preparing and planting the site, aftercare and short- and long- term monitoring.
- It will be essential to prepare a list of resources required and available, such as any new equipment needed or the recruitment of staff or voluntary helpers.

Useful information on practical aspects of the restoration and aftercare of plant communities can be found in *The wildflower handbook* ([UK] Department of Transport 1993) and *Native forest restoration* (Porteous 1993).

Species Recovery Plan for a rare Cuban cycad

The geographically and taxonomically isolated *Microcycas calocoma* (Miq.) A.DC. (Zamiaceae) is a jewel of the Cuban flora and has been declared a National Natural Monument. Research on this rare and threatened plant was initiated at the National Botanic Garden, La Habana, in 1980. This has involved:

- Compilation and evaluation of all available information on its biology.
- Development of a strategy for its *in situ* and *ex situ* conservation.
- Application of techniques required for a successful propagation programme to guarantee adequate germplasm in *ex situ* collections and its reintroduction to the wild.
- Submission of these data to the Ministry of Agriculture, the agency responsible for land management in Cuba, in order to co-ordinate a multidisciplinary group, involving national and regional institutions, to design and carry out practical aspects of a Species Recovery Programme.

Development of the Plan

The research has confirmed that *Microcycas* occurs in a limited area within the Pinar del Rio and that some previously reported populations have disappeared. This represents reduction of a formerly more extensive distribution, primarily as a result of the degradation of the original vegetation. Nevertheless, the species has the capacity to adapt to different edaphic and topographic conditions.

- Numbers are diminished and the trees are mostly old. At almost half the sites, one sex is absent or trees lack reproductive structures, with consequent lack of natural regeneration. Less than 20% of trees produce reproductive structures.
- Natural pollination seldom occurs because of the irregular distribution of the sexes within dense vegetation or perhaps because of the extinction of a specific pollination vector (if the plant is zoogamous as reported in other cycads). Artificial pollination offers the possibility of achieving successful fertilization.
- Seed production is rare, but can be increased by artificial procedures.
- Germination in the wild takes place between mid-September and the end of October under appropriate moisture conditions. At other times, excised embryos can be grown successfully *in vitro*.
- In the wild more than 90% of juvenile plants die during their first year; seeds and juvenile plants are eaten by hutias, a rodent. In the nursery all survive and are healthy.
- The main risks to the species' survival are the extension of arable cultivation, cattle-ranching, reforestation, quarrying and the over-collection of specimens.

The evidence suggests the need to re-evaluate protected areas and for research and development of restitution techniques, aftercare and monitoring of populations *in situ*, together with the maintenance of the species *ex situ* in botanic gardens and other institutions.

A conservation strategy has been produced, with provisions for the *in situ* conservation of *Microcycas* in the protected area of Mil Cumbree and the remainder of the province of Pinar del Rio. It is proposed that the species be represented in the field and display collections of the National Botanic Garden, with germplasm stored and propagated in the garden's *In Vitro* Culture Unit and seed bank.

Techniques have been developed to increase numbers by artificial pollination, enhanced seed germination and *in vitro* culture of embryos and efficient nursery cultivation.

All the accumulated evidence and experience has been submitted to the Ministry of Agriculture, with a timetable of action to be taken. Meanwhile, restitution techniques for the species are being evaluated.

Source: Prof. Káparanza Peña Garoia, National Botanic Garden, La Habana, Cuba.



The endangered Dragon Tree, Dracaena draco, native to Gran Canaria, Spain has been extensively propagated and reintroduced to the wild by the Jardín Botánico Canario, Viera y Clavijo, Las Palmas and used extensively in its public education programmes.

*Photo:
Peter Wyse Jackson*

2.3 Choosing species for reintroduction

Priority must be given to endangered species, especially those that are of global rarity and are of economic, cultural or biological importance. This usually means:

- Species exploited for agricultural, medicinal, commercial or industrial purposes, or closely related species and intraspecific taxa.
- Wild populations of species formerly or currently exploited in a sustainable manner by rural or traditional communities; also plants of cultural, religious and historical significance.
- Species of significant biological value: for example, morphologically or genetically isolated species, monospecific genera and relict populations.
- Plants listed in international conventions, national or local legislation; plants which have special propagation problems; local endemic or relict species; species at the limit of their natural distribution or threatened outside region.

Wallace (1992) has suggested a series of **Practical Criteria** that should be applied before any reintroduction programme is initiated. These are based for the most part on important questions of horticultural and management feasibility rather than botanical or threat assessments. They are:

Criterion 1

- Is an introduction really necessary for the survival of the species?

Criterion 2

- Is an introduction horticulturally feasible?

Criterion 3

- Is an appropriate recipient site available?

Criterion 4

- Are the goals and methods of the introduction clearly spelled out?

Criterion 5

- Is the paperwork in order?
 - Are effective administrative procedures in place?
 - Are all the responsibilities clearly spelled out?
 - Is there enough funding available?
 - Is there a long-term institutional commitment to the care of the introduced population (ie. is the project viable in practical terms)?

"If I had known how many would survive, I wouldn't have planted them in rows", joked Doria Gordon, Nature Conservancy ecologist. Actually, maturity of plants and a prescribed fire management regime are breaking down this pattern as new seedlings appear randomly around the experiment.

Photo: Doria Gordon, Nature Conservancy



In each case the particular need for species introduction should be identified in conjunction with the land-management agency or landowner, the partner responsible for the management of the reintroduced populations. There should be long-term commitment to security of land-tenure and funding for monitoring and management.

Broadly speaking, when undertaking a species reintroduction, at least 50 years security of tenure is advisable, and considerably more with species such as forest trees or desert perennials that have a long life-history or reproductive cycle. This must be discussed at the earliest possible stage with the land-management agency.

Consideration should also to be given to the response and involvement of local people. For example:

- Has the plant a local economic use?
- Has it cultural or religious significance?
- Is it poisonous or in any way detrimental to health?
- Has it been recognized as a noxious weed?

In the case of the reintroduction of forest species, either trees or herbaceous plants, especially in tropical regions, Criterion 3 is of particular significance. Appropriate shade and soil factors must be re-established or conserved, perhaps by means of nurse species, before any successful reintroduction programme can be achieved.



Back into the wild: commonly called Apalachicola Rosemary, this rare Florida endemic has aromatic foliage and typical mint family flowers (see opposite):

Photo: Gary Knight, Florida Natural Areas Inventory

A rare Florida mint settles into a new home

Much of northern Florida, U.S.A., appears to be wild and natural, with long drives down rural highways flanked by pine forests. A closer look, however, reveals that these are not natural forests, but first or second generation pine plantations. Periodic summer fires formerly kept these woods open and sunny, with a rich mix of grasses and wildflowers on the forest floor. Today the dense stands of monoculture pines and the exclusion of fire have diminished this diversity and many endemic species have dwindled to low numbers, hanging on along fences and roads.

Apalachicola Rosemary (*Conradina glabra*) is one of these rare endemic species. It is a semi-woody subshrub of the mint family with aromatic, needle-like foliage and pale blue flowers. The common name aptly suggests its resemblance to the common garden herb Rosemary and its native range on high ground near the Apalachicola River. Florida has many endemics in the mint family, most of them critically endangered, including three other species of *Conradina*, which remain today only in small, isolated populations.

The few surviving populations of *C. glabra* grow on land owned by a paper company. Florida's weak plant protection laws do not compel private landowners to preserve rare species. Fortunately, however, this plant is an ideal candidate for introduction.

Firstly, it has proved remarkably easy to propagate. Shoot-tip cuttings root readily and a collection of 48 clones was established at Bok Tower Gardens. From these, more plants were readily produced for an introduction project of 1,300 individuals.

The ideal recipient site was only 3 km away, on land owned by the Nature Conservancy. The site had been clear-felled and severely degraded, but was now part of a comprehensive restoration project. Nature Conservancy officials agreed to introduce *C. glabra* and to provide long-term monitoring and site management. The introduction was considered from the outset to be an experimental project, but from its scale it was hoped to reproduce a sustainable population.

Another fortuitous factor was the region's cool winter climate and frequent natural rainfall, which resulted in a 95% survival rate. The plants more than doubled in size during the first summer. After the first flowering, only nine seedlings were found, but by the second season many hundreds had appeared. This robust natural reproduction is a reassuring signal that the plants have established successfully.

Source: Susan Wallace, formerly Bok Tower Gardens, Florida, now Director of Horticulture, VanBloem Inc., Georgia, U.S.A.

2.4. The economics of reintroduction

To prevent reintroduction projects becoming a financial liability to a botanic garden, it is important to consider the following:

- How many species can be managed adequately by a single institution?
- How many species should be the target of reintroduction programmes nationally and internationally?
- Is the reintroduction of a particular species cheaper, and safer, than the maintenance of material *ex situ*?
- What are the precise costs of reintroduction?
- Are funds available from collaborating agencies, government departments or interested societies and individuals?

- How will the other partners in the Species Recovery Programme be involved in the reintroduction experiment, either through practical measures or through advice and consultation?
- Have studies been undertaken to determine the genetic variation of surviving wild populations and cultivated material?
- What is the provenance and genetic constitution of available wild-collected material? Is there any evidence for inbreeding depression or genetic erosion?
- Is adequate accession information available on cultivated holdings of the species being reintroduced? If not, then is better documented material available elsewhere?
- What is the likelihood that genetic erosion, inbreeding, hybridization, disease or other loss or damage to the variation of the material has occurred in cultivation?
- If wild material is to be used for reintroduction, will it be from the same population or locality as the proposed reintroduction? If not, are there, or are there likely to be, significant differences between the plant material earmarked for reintroduction and any surviving populations at the intended site of reintroduction?

Photo: Narcissus longispatus, a rare Andalusian plant that is being restored to the wild (see p.22).

Photo: Jardín Botánico de Córdoba



It will be useful to create a checklist of such possible problems for each accession being considered for reintroduction to assess what is the 'best quality' material available.

4.2. Staff, resources and organisation

One member of staff should have ultimate responsibility for running a reintroduction project.

- What staff resources will be required during the various phases of the project?
- Which member of staff will be responsible for the day-to-day management and administration of the project? Will this individual be in a position to direct other staff?
- What is the budget for the reintroduction project? What will be the financial requirements and liabilities for maintaining, continuing or monitoring the project in subsequent years?
- From which source(s) will funding be obtained? If special funds are obtained from a third-party agency, what special requirements or reporting procedures are specified?
- What equipment will be required? Is it already available for use within the institution? If not, what are the financial implications, or where else may equipment be obtained or borrowed?
- Will voluntary labour be used and thus what special conditions or obligations may arise? These may include:
 - training needs of volunteers
 - technical ability of volunteers
 - security, insurance or supervisory considerations.

4.3. Site selection and preparation

- What primary and secondary field research will be needed to identify a suitable site?
- What are the criteria that you are using to determine a suitable site?

What threats to any existing or reintroduced population are present at the proposed site? For example, if the plants are to be reintroduced to a diminished or extinct population, what was the

cause of decline or local extinction of the species?

- from human pressure (collecting, development, agriculture)
 - from other plants (exotics or vigorous natives)
 - from animals (grazing, disturbance, trampling)
 - from pests and diseases.
- What impact might the introduced plants have on other plants or animals at the site?
 - Which management and conservation authorities and agencies must be approached for involvement or permission? Do licences or permits need to be sought and obtained?
 - What is the long-term security of the site and current ownership?
 - What is the access to the site for those involved in the reintroduction, for on-going monitoring (presenting practical difficulties) or for the general public (perhaps presenting further conservation problems)?

4.4. Other aspects

- Is the material correctly identified and labelled?
- Which other botanic gardens (locally, nationally, regionally and internationally) will provide any extra expertise, plant material or other resources needed?
- How does the reintroduction project fit in with the overall Species Recovery Programme or other action plan for the conservation or recovery of the species?
- What implications are there, or experiences to be gained, which may be significant for the long-term management of the species, both at re-established sites and within surviving natural populations?
- An agenda of related activities that will contribute to the potential success of reintroduction programmes might include the following topics:
 - Education and public awareness
 - Funding for the institution and for each Species Recovery Programme
 - Efficient use of appropriate staff resources (eg. ecologists, conservation officers) and additional staff training
 - Data management system development
 - Efficient involvement of biological and horticultural research capacity of collaborating institution(s).

4.5. Horticultural activities

- How many plants will be required for the reintroduction project and how will these plants be propagated, raised and maintained?
- What is the expected germination of seeds, the time cuttings will take to root and the growth rate of young plants?
- Will it be more efficient or feasible to consider direct sowing of seeds at the reintroduction site rather than to use whole plants or 'plugs'?
- At what phase of the project, at what size and stage of life-history and during which season, will plants be introduced?
- What nursery or garden space and facilities (e.g. glasshouses or standing-out areas) are required or available for the project?
- What horticultural supplies will be needed for the practical reintroduction work?
 - fertilizers, soils and composts
 - tools and pots
 - stakes and ties
 - fencing materials
 - transport
 - watering apparatus in garden and field.

4.6. Monitoring after reintroduction

- What degree and frequency of monitoring will be necessary?
- Who will undertake the monitoring and how will it be resourced?
- What provision will be necessary, feasible or desirable for the replacement of any plants that fail to establish?
- What sort of data collection and analysis, publication or other forms of information dissemination will be desirable or necessary?
- What evaluation of methodology and overall review of the Species Recovery Plan may be necessary or desirable to improve future projects?

The Córdoba Botanic Garden and its conservation programme

The Córdoba Botanic Garden was established in 1981 as an initiative of a research team with an established conservation programme for endangered Iberian plants (germplasm banks and reintroduction techniques). Thus, from the outset, the design, facilities and functions of the Botanic Garden were geared to the protection of the plant genetic resources of its own region, Andalusia.

In 1982, the Andalusian Environmental Agency (AMA) was created, in line with the progressive autonomy of the region, becoming the official institution responsible for nature conservation in Andalusia. At an early stage the Córdoba Botanic Garden signed an agreement with this Agency to develop joint conservation plans. As a result, eight specific collaborative programmes were carried out during the period 1988-94. One of them made possible the creation of the Andalusian Germ Plasm Bank, a seed-bank whose efforts have been focused on the conservation of the Andalusian flora. Other programmes allowed the application of *ex situ* and integrated conservation strategies, including reinforcement and reintroduction techniques for the restitution of endangered species in the wild. A specific agreement also permitted a study of the possibilities of increasing the network of natural areas in Andalusia based on their botanical value.

One of the most remarkable collaborative projects has been the elaboration of an overall strategy for conservation of the Andalusian flora, published under the title "Protection of the Andalusian Flora" (Hernández Bermejo, Clemente Muñoz et al. 1994). In this work, out of an estimated total of 4,000 taxa in the Andalusian flora, 1,053 species and subspecies have been selected for special conservation on the basis of rarity, endemism or risk of extinction; 463 of them are endemic to the region. The most urgent action is being directed towards the restitution of 72 taxa considered to be at greatest risk, and specific criteria and appropriate techniques are required for each. Finally, all international conventions, as well as the European Union, Spanish and Andalusian legal framework concerning the conservation of regional floras, are set out in this work.

The Córdoba Botanic Garden is taking action towards the restitution of 21 of the 72 taxa at greatest risk. Research is being carried out to determine their exact distribution and numbers, their biology, together with information on propagation (including *in vitro* culture techniques), germplasm conservation, obtaining cultivated material and field sampling of taxa that could be potentially cultivated. Finally, their restitution in the wild, through introduction, reintroduction or reinforcement, is carried out.

Some of the species currently in the most advanced phase of study are:

Antirrhinum charidemi, *Aquilegia pyrenaica* subsp. *cazorlensis*, *Artemisia granatensis*, *Betula fontqueri*, *Cneorum tricoccum*, *Cytisus moleroi*, *Narcissus longispathus*, *N. nevadensis*, *N. tortifolius* and *Rosmarinus tomentosus*. The greatest success so far has been achieved in the restitution of *Antirrhinum charidemi* and *Artemisia granatensis*.

The Córdoba Botanic Garden has co-operated on several programmes to protect endangered plants with other institutions in Spain and abroad. A project on aromatic and medicinal plants of the Mediterranean region has been carried out jointly with the botanic gardens of Porquerolles (France) and Pisa (Italy); another, on the propagation of several orchids threatened with extinction, was carried out jointly with the National Botanic Garden in Cuba. The successful restoration of an endangered species of the Balearic flora, *Lysimachia minoricensis*, was achieved on Minorca in 1990, in collaboration with the Balearic Autonomous Government and the botanic garden of Brest (France).

Source: Esteban Hernández Bermejo, Director, Jardín Botánico de Córdoba, Spain

5

Sampling Procedures for *Ex Situ* Conservation and Reintroduction

Adequate sampling is the cornerstone of sound conservation and reintroduction policy. The narrow genetic base of many botanic garden collections can lead to genetic erosion through sampling error, bottleneck effects (genetic drift), repeated selfing or cloning, and poor maintenance or documentation. Such holdings are rarely of value for conservation projects, except as educational exhibits.

Sampling procedures for the conservation and reintroduction of rare plants should aim to maintain and promote existing genetic variation rather than to erode it any further. In theory, at least 95% of the genetic variation of a population or populations, or as wide a range of genotypes as possible, should be captured to provide adequate material for reintroduction. However, practical reality, especially in a tropical region of high diversity, will often make this difficult to achieve.

5.1. Suitability of material available or assembled for reintroduction

It is important to assess the whole range of genetic variation of a species for reintroduction, both in the wild and in cultivation. Botanic garden stocks are especially important where a species, or one or more populations, have become extinct in the wild, and several examples of this sort of rescue have been documented.

At an early stage of any reintroduction project, it is important to check:

- What stocks are already held by your institution, other botanic gardens and scientific institutions or private individuals?
- Is the material derived from seed or is it clonal? Seed should be used whenever available, representing as it does a broader sample of genetic diversity.
- What is its level of genetic variation?
- What is the genetic diversity of the surviving wild populations?
- Taking the above into account, is there need for further collection of material from the wild? Is this feasible, ethical or sensible?

Proper assessment of genetic variation not only ensures that existing stocks are adequate, but will guide further sampling procedures.

5.2. Collection of material from the wild

Sampling should follow the practices and techniques outlined in sections 5.3-5.6 (below). The background principles for sampling natural populations are given in detail in Falk and Holsinger (1991). Points to consider when sampling:

- Samples should try to capture 95% of the genetic variation of a rare plant, which is usually possible where populations are small and propagules easily selected.
- Sampling needs to take into account any data that may be available on breeding systems, genetic variation and reproductive biology.
- Samples should be collected from across the whole range of the species, although, wherever possible, reintroduction should be based on genetic stock
- from the same site or nearby.
- Priority should be given to populations most threatened by human or natural factors.
- Avoid the collection of material showing evidence of infertility or infection with pathogens, especially viruses.

- Samples in storage or cultivation should be kept separate from one another.
- Documentation must be meticulous.

5.3. Ecological and geographical survey

As full as possible a study should be made of variation across the geographical and ecological range of a rare species. This will involve literature, garden, laboratory and field studies, and will provide data on which to base sampling procedures and number of samples.

Geographical distribution and genetic variation

Collectors should consult available monographic and floristic literature, herbarium specimens and known experts to determine the distribution of a species. Particular attention should be paid to any morphological differences within or between populations as evidence of genetic diversity, especially where it can be related to ecological gradients or discontinuities.

Where possible, an assessment can be made of genetic variation on the basis of molecular techniques, making full use of all material that is

available. A botanic garden that is considering a plant reintroduction programme should:

- Investigate whether facilities are available locally, nationally or regionally, for screening the genetic variation of the species concerned.
- Obtain the support or co-operation of institutes maintaining these facilities.
- Consider whether it is appropriate or possible for such facilities to be established at your own institute.

Electrophoretic starch- or polyacrylamide-gel analysis of isozymes (variants of the same enzyme) is a cheap and relatively rapid procedure for assessing genetic variation (Soltis & Soltis 1990). Isozymes move at different rates across an electric field, due to minute differences in electric charge, and are then detected by the staining of a substrate that they catalyse. This technique gives a measure of both the numbers of polymorphic loci and their degree of heterozygosity. Analysis of random amplified polymorphic DNA (RAPD), a technique that screens DNA nucleotide sequences as they are amplified by DNA-polymerase, provides a more direct assessment of genetic polymorphism (Williams *et al.* 1990). It is quicker, but does not reveal heterozygosity, as the RAPD is usually a dominant marker.



Terraces behind High Peak, St Helena, planted with St Helena Ebony (*Trochetiopsis melanoxylon*) and a hybrid.

Photo: Rebecca Rowe

Genetic screening to conserve *Trochetiopsis* species (Sterculiaceae) on St Helena

Trochetiopsis is an endangered woody genus endemic to the South Atlantic island of St Helena. There are two species: St Helena Redwood (*T. erythroxylo* (Forst.) Marais), extinct in the wild, and St Helena Ebony (*T. melanoxylo* (Ait.f.) Marais), thought to be extinct until 1980, when two plants were found on a cliff inaccessible to goats.

All Redwood plants now known are descended from a single individual and have been multiplied through selfed seed. By 1993, 86 individuals had been reintroduced to six sites on St Helena.

Ebony has been propagated from cuttings taken in 1980 from the upper of the two plants and in 1982 from the lower plant. By 1993, 2,000 plants of mixed seed and vegetative origin had been reintroduced to one site and introduced to three others on the island. The lower plant genotype is probably under-represented in the plantings.

Hybrids were produced when the two species, previously isolated geographically, were brought together in the nursery. Four F1 hybrids have provided seed for introductions at three sites, where in 1993 about 800 hybrids had been planted with either Redwood or Ebony.

Trochetiopsis is represented in the collections of at least seven botanic gardens. Confusion of records during transfer of seed and cuttings between institutions has resulted in loss of provenance and collection data. It is known that Ebony was represented in botanic gardens only by the upper plant until 1993.

The conservation of *Trochetiopsis* has until recently been carried out without any genetic information. Current research is examining genetic diversity by means of isozyme and RAPD analysis, in order to answer the following questions:

1. What is the present level of genetic variation within and between the two species?
2. What genotypes are represented in botanic garden collections; and can botanic gardens provide an additional source of genetic variation in the case of Redwood?
3. To what degree does introgressive hybridization, which may hybridize the pure species out of existence, occur in mixed planted populations?
4. Is representative sampling being carried out for the benefit of future reintroductions?

Lack of variability in *Trochetiopsis* has already been shown at isozyme loci, and unique isozyme markers have been found in both species. Preliminary RAPD data for 20 arbitrary primers (10-base nucleotide sequences) has detected several differences between the two original Ebony plants. This information will be useful in analysing the degree to which each genotype is represented in the plantings on the island. A greater level of genetic polymorphism has been detected using RAPD compared to isozymes. It is likely that RAPD may prove more useful in the study of the degree of introgressive hybridization that is occurring at the planting sites.

Source: Rebecca Rowe and Quentin Cronk, Plant Sciences Department, University of Oxford, UK.



*St Helena
Redwood
(Trochetiopsis
erythroxylon)
planted at
Scotland,
St Helena*

*Photo:
Rebecca Rowe*

5.4. Sampling

Number of populations sampled

The number of surviving populations of a species should be estimated as a basis of sampling policy, with procedures influenced by consideration of factors such as genetic variation, reproductive biology and life-history (Falk and Holsinger 1991). The sampling of many populations is to be encouraged, although for most rare plants five populations probably represents an adequate sample.

There is debate among conservationists and population biologists as to the number of individuals that need to be sampled if material from a population is to maintain its genetic integrity in cultivation. A minimum of 500 individuals to maintain long-term fitness has been suggested, but many endangered species do not exist in nature in such large populations.

In the case of wider-ranging populations with numerous individuals, biological commonsense and practical and economic resources will determine how large a sample can be taken.

Sample size

If the population contains more than 50 individuals, seed should be sampled randomly from 10 to 50 or more plants, depending on seed yield per plant. The number of seeds collected will influence whether or not material is to be held for purely conservation purposes or if it can also be made available for other purposes without the need for costly multiplication in cultivation.

If the population contains fewer than 50 individuals, seed should be harvested, so far as is possible, equally from all fruiting plants. It is important not to damage small populations, and no more than 20% of the available seed should be harvested unless that population is threatened with immediate destruction (e.g. road construction). Where the species is represented by a few scattered populations, it is recommended that all be sampled. Where the species is widely distributed, 10-20 samples, reflecting ecological and geographical variation, should be sufficient. Each sample should be accessioned and held separately.

Multiplication of small samples

A population sample of 50-100 plants, perhaps 5,000 seeds in all, constitutes an adequate germplasm collection, but if the population or the yield of seed per plant is small, samples will require multiplication. In such cases collectors should harvest a few seeds from each individual and keep samples separate to be grown on as 'families', to represent a maximum number of maternal genotypes in the multiplication phase. Not all seed should be sacrificed in one attempt, and if necessary the genetic integrity of the seed lot should be subordinated to its overall security.

The problems of small population size can be overcome by making more than one collection from the original population, for example one of seed and

one of pollen, to increase the genetic variation of the sample. Sampling over a period of time may also be appropriate for a population with an extended flowering period.

Extent of populations to be sampled

Each population should be assessed for size, density and extent, although precise definition of a population may not be easy. In some situations, such as tropical forests, populations may be impossible to delimit, so a radius of, say, 1 km might be used. This assessment should include observations along the following lines:

- A study of ecological and environmental conditions will identify populations particularly at risk from degradation or changes in the environment. Climatic data will also help to decide methods of collection and transport and the need for any additional drying in the field.
- The general health and vigour of individuals in the field should be noted, including seed-set, any pathogens or parasites, insect damage or grazing by herbivores. Malformed flowers or other organs and few or empty seeds may be evidence of inbreeding depression, especially in smaller populations.
- Vegetatively propagated clones of species, especially in smaller populations, may result in the repeated collection of genetically identical material. In outbreeding species a representative sample of the range of genetic diversity will be contained in a much smaller seed sample than an inbreeding species made up of numerous reproductively isolated, small populations.
- Information about pollinators, dispersal agents and levels of inbreeding will be useful for the later maintenance of the material in cultivation or storage.
- Flowering and fruiting periods, seed dispersal mechanisms and levels of seed predation will decide the right time to collect propagules.

5.5. Documentation of collections

Rigorous documentation is essential for all plant conservation collections, particularly of data gathered in the field at the time of sampling. This should include:

- Voucher herbarium specimens, where possible

- Collector(s), number and institute, date
- Geographical and topographical details (latitude, longitude, map references & satellite position, etc.)
- Full habitat notes
- Characteristics and numbers of populations and individuals
- Any threats.

5.6. Collecting techniques

Containers and bags

Seeds should be collected into permeable cloth or non-glossy paper bags, with plastic bags used only to collect fleshy fruits. If seed cannot be cleaned soon after collection, it will be necessary to aerate the bagged fruit regularly. If the plant has shed its seed and it has been collected from the ground, this should be recorded.

Cleaning and drying seed in the field

Seed should be cleaned as fully as possible in the field, and insects and fleshy material that may rot should be removed. If seed samples need to be dried, they can be placed inside a sealed container with blue silica gel (ratio of 2:3 seed to fresh silica gel with minimal air) or dried rice, for example, as an effective substitute. It is not advisable to dry in direct sunlight, as the heat may affect long-term seed viability.

Temporary storage

Seed should be kept dry and cool (but not below 10°C) and should be transported to the seedbank as soon as possible. Simple methods such as keeping seed in the shade and away from the direct sunlight of windows in a vehicle will enhance the possibility of maintaining long-term viability. If there are considerable diurnal humidity and temperature fluctuations, placing collecting bags into a large plastic bag at night will help maintain an even temperature and relative humidity.

Vegetative and clonal material

If species are largely propagated vegetatively or seed is not available, propagules may be collected in the form of bulbs, bulbils, corms, tubers, rhizomes, cuttings, scions, etc. Samples should be taken from at least 10-15 individuals, as a bulk sample from an area of up to 100 x 100m or less. It is often difficult to determine the extent of clone size, so samples should be widely spaced.

National and International Communication

6.1 Links with other institutions

Links are to be encouraged between botanic gardens involved in plant introductions and Species Recovery Programmes. A link should aim:

- To aid in the movement, exchange and, in some cases, repatriation of germplasm and to ensure co-ordination of reserves of germplasm as an insurance against loss.
- To divide the tasks involved in the conservation and reintroduction of individual species or populations in relation to the different skills and resources of the participating institutions.
- To develop institutional capacities for plant conservation through help with training (managerial, technical, scientific, horticultural and educational), strategic planning and project development; together with the transfer of information and experience through staff exchanges.
- To encourage the work of conservation-orientated smaller botanic gardens over larger, often older institutions.

Choice of institutions for linkages

(a) National

Links with land management and development agencies

- Establish agreement for the reintroduction and long-term management of threatened plant populations.
- Liaise with local communities around the proposed sites of reintroduction or existing populations.
- Publicise the project, emphasising its practical, economic and ethical importance.

Scientific links with other institutes or universities

- Sharing of propagation and other conservation and horticultural techniques.
- Exchange of field and experimental, biological and ecological information.
- Advice on taxonomy and distribution, past and present.
- Help with the identification of plants that are priority targets for reintroduction.

Links with other botanic gardens

- Exchange of plant material or propagules.
 - Exchange of horticultural information, techniques and practical expertise.
 - Definition of geographical areas of mutual or exclusive interest to maximize efficient use of resources.
 - Sharing information, staff and technical resources such as seed-bank facilities to avoid expensive duplication.
 - Emphasis of cooperation in publicity and education to raise public awareness of the conservation issues.
 - Funding and implementing training programmes for staff.
- (b) International**

'Sister' or 'twinned' botanic gardens (these can also operate nationally) can locate and share plant, garden, human and technical resources and funding.

International co-ordination can be achieved through

- The IUCN Species Survival Commission (Reintroductions Specialist Group), c/o Threatened Species and Habitat Unit, Royal Botanic Gardens, Kew, U.K. - holds an archive and database on plant reintroduction projects.
 - Botanic Gardens Conservation International (BGCI), Kew, U.K.;
 - World Conservation Monitoring Centre (WCMC), Cambridge, UK;
 - World Wide Fund for Nature (WWF-International), Genève, Switzerland;
 - International Association of Botanic Gardens (IABG), Córdoba, Spain;
 - Center for Plant Conservation, St Louis, U.S.A.
- Exchange or help with resources such as equipment and technology (e.g. database development).
 - Commit financial resources in each institution to the sole purpose of funding links, for travel, exchange of data, equipment or plant material. Any approach to a third party for funding will be enhanced by a co-operative application. Funding for long-term management and monitoring of project sites may also require inter-institutional agreement and commitment.
 - Ensure mutual agreement at an early stage on possible legal or ethical restriction of the availability of germplasm or its transfer; also on the exchange of related information and other data. This should extend to ethical agreement on conservation practices and the division of income in commercial developments.

International co-operation between nations, as part of friendship treaties or aid agreements.

These links will assist with:

- The identification of priority species for reintroduction at global, regional, national and local levels.
- To identify and foster 'sister' links between institutions.
- To identify other resources available, including the location of suitable plant material from the wild or cultivation.
- To raise awareness of conservation issues at international and, therefore, governmental levels.
- The development and maintenance of suitable databases to help co-ordinate worldwide efforts in reintroduction and species recovery.

Practical considerations to ensure success of a link

- Define the purpose of the link, with a jointly prepared, written statement and an agreement or 'Memorandum of Understanding'.
- Define the period of time of the link, normally long-term, related to its purpose.
- Develop good personal relationships between the key staff members in each participating institution. Personal commitments from the staff to such linkages are essential.
- Planning a schedule of visits and exchanges of staff is important, especially when a link is international.

Ensure that participating institutes are fully briefed on the working of CITES and other international agreements on plant trade.

6.2 Network links

The establishment of national and regional plant conservation networks are an especially valuable resource. They can highlight priorities for reintroduction, arrange training, share resources and data, and foster many activities between twinned botanic gardens. They also provide a forum and an 'umbrella' to bring together expertise in plant conservation from different sectors: botanic gardens and other scientific institutions, private and commercial horticulture, NGOs and land-management agencies.

A wide range of national and regional botanic garden networks has been established in all parts of the world (Wyse Jackson 1993). Some of these are national or regional organizations or groups closely associated with Botanic Gardens Conservation International, or regional sections of the International Association of Botanic Gardens such as the Asociación Latinoamericana y del Caribe de Jardines Botánicos. Others are independent national and international NGOs, such as the American Association of Botanical Gardens and Arboreta, the [U.S.] Centre for Plant Conservation, and the Asociación Mexicana de Jardines Botánicos, or government sponsored organizations, such as the National Botanical Institute of South Africa or the Rede Cubana de Jardines Botánicos. Recently