Introduction

Generally, the more genetic variation that is captured when collecting material for your species recovery project, the more likely it is that the recovery of the target species will be successful in the long-term. In this brief, we provide guidance on how to sample and collect material to ensure that recovered populations are functioning, self-sustaining and able to survive changing conditions.

Where to source your material?

*Ex situ collections*

Material from *ex situ* collections (material maintained outside the target species’ natural habitat) can sometimes be used for species recovery. However, it is very important to ensure that material used for your recovery project:

1. Is of high quality and purity
2. Is of known origin
3. Contains sufficient genetic variation.
A seed collection for recovery is typically stored short to mid-term before use (direct seeding, growing plants for transplantation, or producing plants to generate larger quantities of seed). Collecting seed well in advance of potential use has several benefits:

- **Knowing the amount and quantity of seed available**
- **Time and availability to confirm species identification using material collected from the field (e.g. botanical vouchers)**
- **Having seed available in seasons or years when low or no wild seed is produced**

### Advice for collecting vegetative material for species recovery

If you are collecting vegetative material, follow the guidance provided below on the number of individuals and populations to sample from. However, fewer cuttings should be taken from each mother plant compared to the number of seeds recommended. The exact number will depend on the type of plant and you should ensure that the collection of material does not significantly harm the mother plant.

### Collecting your own material

It is recommended to collect your own material for recovery programmes, to help ensure meeting the three requirements above. For rare species, seed or plants are unlikely to be available from commercial sources. It is usually best to collect seed rather than vegetative material (e.g. cuttings) where possible, as vegetative cuttings produce plants that are genetically identical to the mother plant and therefore limit the genetic diversity captured. In some cases (e.g. for target species that do not produce viable seed, or highly inbred seed), vegetative propagation will be the only option.

### How many populations?

**A population:** A group of individuals, capable of interbreeding, that occupy a defined geographic area.

You must aim to ensure that the material used for your recovery project is collected from populations that are sufficiently large and contain enough genetic variation to ensure that the newly planted individuals are functional.

---

**TOP TIP**

**Material of known provenance and appropriate genetic diversity may be available from a botanic garden or seed bank.** BGCI’s PlantSearch database (www.bgci.org/plant_search.php) can be used to locate plant species found in living and seed bank collections, including from threatened plants.

**TOP TIP**

**Collecting your own material requires adequate space, personnel, expertise and resources to maintain and grow the seed.** If this is lacking, resources may be initially better spent in solving infrastructure deficiencies, forming partnerships with other institutions with expertise (such as botanic gardens), or making a small scale seed collection and completing research that will give basic knowledge required for the species such as germination requirements.
and resilient. Currently there is not an agreed number of populations to sample from, as it will vary depending on the species and the scale of the recovery programme. However, the following steps can be taken to help you determine how many populations are appropriate:

• **STEP ONE** – A thorough field exploration should be carried out to determine how many populations of the target species exist.

• **STEP TWO** – When several populations of the target species exist, it is best to conduct a genetic analysis to evaluate the genetic variation within each population. Also the biogeographical characteristics of each population should be determined. It is important that chosen populations cover a range of environmental conditions, space and geography to maximise matching the biogeographical conditions at your recovery sites.

• **STEP THREE** – Select populations to collect species recovery material. In cases where there are fewer than five populations, each one should be sampled. For species with broad distributions and multiple populations, recommendations range from five to fifty populations per ecoregion. Ideally, seed source populations should have at least 1000 plants and a history of stable size.

• **STEP FOUR** – A number of resources are available to help you to evaluate the completeness of your collections. Niche models can measure coverage in environmental space, while genetic models can measure coverage in genetic diversity.

**TOP TIP**

If sufficiently large and accessible populations exist, populations much smaller than 100 plants should not usually be sampled for seed due to possible inbreeding and genetic drift.

### Adaptation

Adaptation is a key concern for species recovery. Each recovery site will have its own set of environmental conditions. The available seed source may be many hundreds of kilometers away and/or from a different environment. There is increasing evidence that the success of many recovery actions will depend on using seed that is adapted (or matched) to the local current or future environmental conditions. This does not necessarily require seed to be geographically local but some degree of environmental matching is important.

**Inbreeding depression** is a reduction in biological fitness (vigour) in a given population as a result of breeding between related individuals. It may occur in outcrossing plants with population sizes approximately less than 100 individual adult plants.

**TOP TIP**

Choose seed source sites based on similarity in environmental factors rather than choosing based on simpler metrics such as proximity (e.g. 100 km radius of the recovery site).
Local adaptation is not universal and can be imperfect, in that plants may as well, or even better at sites other than their origin. Local adaptation is a good starting point for choosing source sites, however for many plant species introduction of non-local seed does not necessarily mean it will be maladapted to local conditions. It is particularly important to consider sourcing seed from outside the local site when recovering very small populations to minimise inbreeding depression.

How many individuals?

The theoretical minimum number of individuals required to capture 95% of existing genetic variation from a single population of randomly mating, well distributed plants is to sample 10 to 100 seeds from 50 individual plants collecting a total of 500 to 5000 seeds. If the seed is to be shared with many partners or if germination rates are very low, it may be necessary to increase this number. However, this theoretical number is not always sufficient to ensure that the restored population has the vast majority of genetic material from the source.

Other important factors to consider are provided below.

Spatial distance

Spatial distance is an important consideration at all levels for capturing more genetic diversity. This is because nearby individuals are often more closely related than distant ones (particularly for species with low dispersing seed and pollen). Therefore:

- Sampled populations should be distant
- Sampled plants within a population should be distant
- Sampled seeds should be collected from different parts of the plant (e.g. when collecting from a tree, collect seed throughout the crown). This is particularly important for large plants such as trees that are likely to have been pollinated by different paternal plants.

Sampling strategies

Good spatial coverage can be achieved by sampling:

- Randomly (using random numbers to choose plants)
- In a stratified manner (randomly within a chosen area)
- Systematically (at regular intervals on a grid).

Systematic sampling is recommended in large, uniform landscapes, while stratified sampling is recommended in highly environmentally variable sites. When choosing plants, a collector should include a variety of ages, growth forms and seeds. Do not only sample the plants that appear to be the largest or healthiest. If sampling cannot be made with good spatial coverage (e.g. due to land access limits), the number of plants sampled from should be increased by at least a factor of two, and more if possible.

How seeds are sampled can be as important as how many seeds are collected!

Sample from as much of the population as possible.
How many seeds?

There are several key factors to consider when determining the number of seeds to collect:

| Number of maternal plants to sample | Sample seeds from as many maternal plants as possible. The genetic reward is always improved by sampling from a new plant. For example, sampling 50 seeds from 10 plants captures less genetic diversity than 10 seeds from 50 plants. |
| Number of seeds per plant | Where possible collect the same number of seeds per plant. |
| Over-harvesting | Some seed must be left for natural plant regeneration in the sampled site and forage for wildlife. The level of safe harvest will vary considerably among species, from 10% of available seed to (rarely) over 95%, with 20% recommended if no knowledge of the species is available. Annual plants, especially those without a seed bank, should be harvested below 20%. There are however exceptions. If the species or population is in imminent danger of complete loss, collecting as much seed as possible may be advised. |
| Adjusting seed quantities | Species recovery is only successful when adult plants reproduce. Some authors recommend accounting for at least 90% loss of seed over the recovery process (e.g. seed which is infested or empty, seedlings that die). |
| Species biology | Sampling should be adjusted for a species’ dispersal mode, life history and other aspects, often resulting in larger sample sizes than the theoretical minimum. |
Genetic diversity

High genetic diversity is known to help populations survive future stresses, such as seasonal extremes or gradually increasing temperatures. Two methods which can be used to ensure high genetic diversity of material for restoration include:

- **Composite provenancing** – using predominantly “matched” seed (e.g. similar environmental conditions) but also including a substantial percentage of seed from non-matched locations (different environmental conditions) to facilitate natural selection and future adaptation.

- **Predictive provenancing** – using a seed source selected according to future projected environmental change, e.g. planting southern or low altitude seed sources in more northerly or higher altitude locations in the expectation that the recovery location will experience higher temperatures in the future. This method has rarely been tested and has some dangers, but is sometimes recommended.

Other considerations

<table>
<thead>
<tr>
<th>Additional uses</th>
<th>Additional seed should be collected for backing up the collection, distributing the material for study and monitoring seed viability. All of which are essential for long-term seed storage and are recommended for species recovery.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Costs</td>
<td>To maximise the genetic variation captured, it is more effective to move on to a new plant rather than take more seed from a plant already sampled. Likewise, it is better to move to a new part of the population, a new population, or a new region. However, this must also be balanced by logistical limitations (e.g. time and money).</td>
</tr>
<tr>
<td>Timing of collection</td>
<td>The fruiting season of a given species may be spread over multiple months, and not all individuals will have mature seed simultaneously. It is important to sample multiple times in the fruiting season (only sampling mature seed), and over multiple years to maximise genetic diversity of collected material.</td>
</tr>
<tr>
<td>Associated data</td>
<td>To be useful for a species recovery programme, accompanying data for seed collections must include where, when and how seed was collected. Comprehensive field data (e.g. plant size, health, soil type, associated species, site history and management regime, etc.) will help greatly in finding an environmental site “match”. This data determines where the seed can be reintroduced, what can be mixed together and how the species can be propagated <em>ex situ</em>. Lastly, data should be kept on the use of the seed, including the number of seeds which do not germinate, the number of plants which die, etc.</td>
</tr>
</tbody>
</table>