Gamsberg Search, Rescue and Translocation Protocol V02

ENDEMIC VISION
ENVIRONMENTAL SERVICES

10 YEAR ANNIVERSARY

SANBI
Biodiversity for Life
South African National Biodiversity Institute

PREPARED FOR:

black mountain

BLACK MOUNTAIN (PTY)LTD

Directive

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Team Member	Qualifications	Experience
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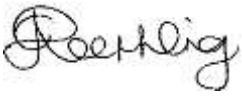
The author of this report, Chrizette Neethling, does hereby declare that she is an independent consultant on behalf of EV and has no business, financial, personal or other interest in the activity, application or appeal in respect of which she was appointed other than fair remuneration for work performed in connection with the activity, application or appeal. There are no circumstances that compromise the objectivity of the specialist performing such work. All opinions expressed in this report are her own.



Signed: **C.D. Neethling**

Dated: 01 April 2020

Document Specifications

1. Document Structure

Specification	Report Details
Title:	Gamsberg Search Rescue and Translocation Protocol V02
Report structure:	Main protocol with Appendices - Pdf Map files: CSV; Shp
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Date:	01 April 2020
Author(s):	C. Neethling; E. Cronje; V. Wilman (SANBI - Millennium seedbank). D. Roets (Consultant) with specialist input from E. van Jaarsveld (Succulent specialist); W. Voigt (SANBI - Karoo Botanical Garden); V. Wilman (SANBI)
Client:	Vedanta Resources; Biodiversity Department
Synopsis:	A definitive and explanatory search, rescue and translocation protocol is required to fulfil the obligations for environmental authorisation to drill Gamsberg East.

2. Document History

Date	Author	Report Name	Status
30/05/2018	D. Roets Botanical Services	Gamsberg Search & Rescue and Transplantation Protocol	Final
01/02/2020	EndemicVision	Gamsberg Search & Rescue and Transplantation Protocol V02	Draft
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3. Document Distribution

Date	Company	Person	Data type
	BMM		
	Vedanta Prospecting		
	SANBI		

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Definitions and Acronyms

Term	Definitions
Listed Species	Species listed in terms of International published species lists (IUCN, CITES), National and/or Provincial legislation.
Offset	Measurable conservation outcomes resulting from actions designed to compensate for significant adverse biodiversity impacts arising from project development and persisting after appropriate avoidance.

Acronyms	Description
BMM	Black Mountain Mining (Pty) Ltd
DENC	Department of Environment and Nature Conservation
MSB	Millennium Seed Bank

1 Background

Black Mountain Mining (Pty) Ltd (BMM) submitted a search and rescue protocol as part of its environmental authorization process to allow for drilling on Gamsberg East. The Department of Mineral Resources granted an Environmental Authorization on 19 February 2019.
Appeal to the authorization was made whereby Black Mountain was requested to update the Search and Rescue protocol.
With reference to section 1.14 and 1.20 of the Appeal decision LSA 182804 dated 05/08/2019 the following amendments are required:

- Condition eight (8) of the Record of Decision recognizes the need for rescue operation of all listed species suitable for translocation;
- Clarity must be provided of the way a similar habitat for flora is to be identified for the purpose of transplantation;
- SANBI should be involved in the protocol compilation and translocation process; and
- Plan must be made subject to public comment (standard stakeholder engagement process).

EndemicVision Environmental Services (Pty) Ltd was appointed to update the search and rescue protocol with the input of SANBI and other relevant specialists.

2 Protocol Objectives

The objective of this protocol is to ensure successful search, rescue, translocation and conservation of listed species affected by prospecting and or mining at BMM operations.

The objective is achieved by fulfilling the following goals that can be measured as key performance indicators:

- Adequate seed harvesting to allow for restoration of the species on site;
- Adequate seed harvesting to allow for enough seed banking at the Millennium seed bank to conserve this species;
- Adequate search, rescue and in-nursery survival of listed species to re-establish on impacted areas;
- Adequate numbers of listed species are provided to recognized nurseries for safe keeping; and
- Listed species are translocated successfully to impacted areas.

3 Limitations and Assumptions

This protocol excludes aspects of nursery management that is part of the nursery management plan.
Nursery propagation, care and monitoring is excluded.
It is assumed that seed collection will take place by trained staff or staff under supervision of trained supervisors.
This plan is part of an adaptive management process and must be updated regularly based on new conditions and information.

4 Safety, legal requirements and risk assessments

Landowner consent is required in writing and should be available while on site.
Flora permit conditions that instruct seed collection or search and rescue as a condition must be used to align collection strategies in order to fulfill the requirements of the permits.

Transportation, collection, cutting or plucking permits must be obtained from the relevant departments before collection commence in terms of national legislation for protected trees and in terms of provincial legislation for protected plants and trees.

Be aware if there are any rare plant or animal species in the area, or anything else that could be damaged by your chosen collecting technique. Modify the collecting methods if necessary.

Identify if there are any specific hazards to the collecting team. Assess the danger and decide how to minimize the risk, work in pairs, carry a first aid kit and maintain visual contact with team members at all time. Compile a Risk Assessment with the competent person on site prior to commencement of any work on a daily basis.

Know your plant material, if the plant is toxic, has an irritant or is very thorny dress appropriately. Always take gloves, tarps and extra clothing with to avoid contact with hazardous material.

5 Herbarium voucher specimens

Herbarium specimens are an important product of the project and represent a valuable biological record in their own right. Herbarium specimens are sent to experts to confirm identification of the species collected. SANBI will collect herbarium samples for the National Herbarium and train BMM employees as part of SLA agreement in place.

Care needs to be taken when selecting herbarium material, it is critical that it fully represents the plant population from which the seed was collected. The voucher will always provide a material link to the collection; if the voucher is renamed in the future the collection can also be kept up to date.

One or more of the collectors should select the herbarium material to be pressed. All plant population exhibit morphological variation; try to select material, which represents the 'typical' individual of the population.

In the case of large plants, remove the required number of specimens from a single representative individual.

- In the case of small plants, and most annuals, you will need a complete individual for each duplicate herbarium specimen.
- If you have collected seed from an extremely variable population, it is good practice to collect additional, separately numbered, herbarium specimens to illustrate the range of material available.
- If you have collected flowering material on a previous visit, you may now have to collect an additional voucher at seed collection time to make absolutely sure of the identification.

5.1 Plant herbarium material should be fertile and representative of:

- Population: collect all or a range of phenotypes;
- Individual: Collect some top, middle and base of plant if not possible to collect whole plant;
- Underground parts if possible: These are particularly needed for restios, bulbs, grasses and sedges;
- Bark/wood;
- Developmental stages (leaf buds, young leaves, flower buds);
- Male and female flowers;
- Different flower forms;
- Points of attachment i.e. preserve arrangement organs; and
- Loose collections of material such as seeds or small flowers. Place in capsule as extra material.

GOLDEN RULE: LOOK AT AND PLAN SAMPLING OF THE PLANT BEFORE CUTTING BITS OFF.

5.2 Determining the number of herbarium vouchers needed:

The effort to collect herbarium specimens is time and resource intensive. It must be planned to make sure enough vouchers are collected first time round:

- 2 vouchers need to be taken for the Millennium Seed Bank;
- Where possible enough material to cover 2 x A3 herbarium sheets;
- 1 Voucher is sent for identification and lodged at the herbarium in SA and one is stored with the seed collection; and
- 1 voucher or photographs can be taken if it is a very threatened species with not many plants in the population.

5.3 Data Collection

The collection of data associated with the species or populations from which seeds are collected is a vital contribution to knowledge about these plants. Habitat information such as altitude, slope, landform, aspect, geology and soil characteristics have important implications for restoration ecology applications. Population characteristics such as phenology, number of plants sampled, % population producing seed, climatic conditions and temperature are useful data for conservation authorities and information about the plants themselves (form, height, flower/fruit morphology) is required by taxonomists.

Use a standard herbarium data sheet (Appendix 4) to record all details of the collection pertaining to:

- Locality and date collected;
- Edaphic factors (soil, geology);
- Habitat and vegetation type;
- Sampling information; and
- Include a description of the plant, concentrating on things lost in sampling of herbarium specimens - smell, colours, life form, 3 dimensional structures including: Habit/height/spread, underground parts if not collected, stems and trunks-buttresses, bark, latex etc. Stipules, Fresh size, shape, colour of inflorescence, flowers, fruits and seeds.

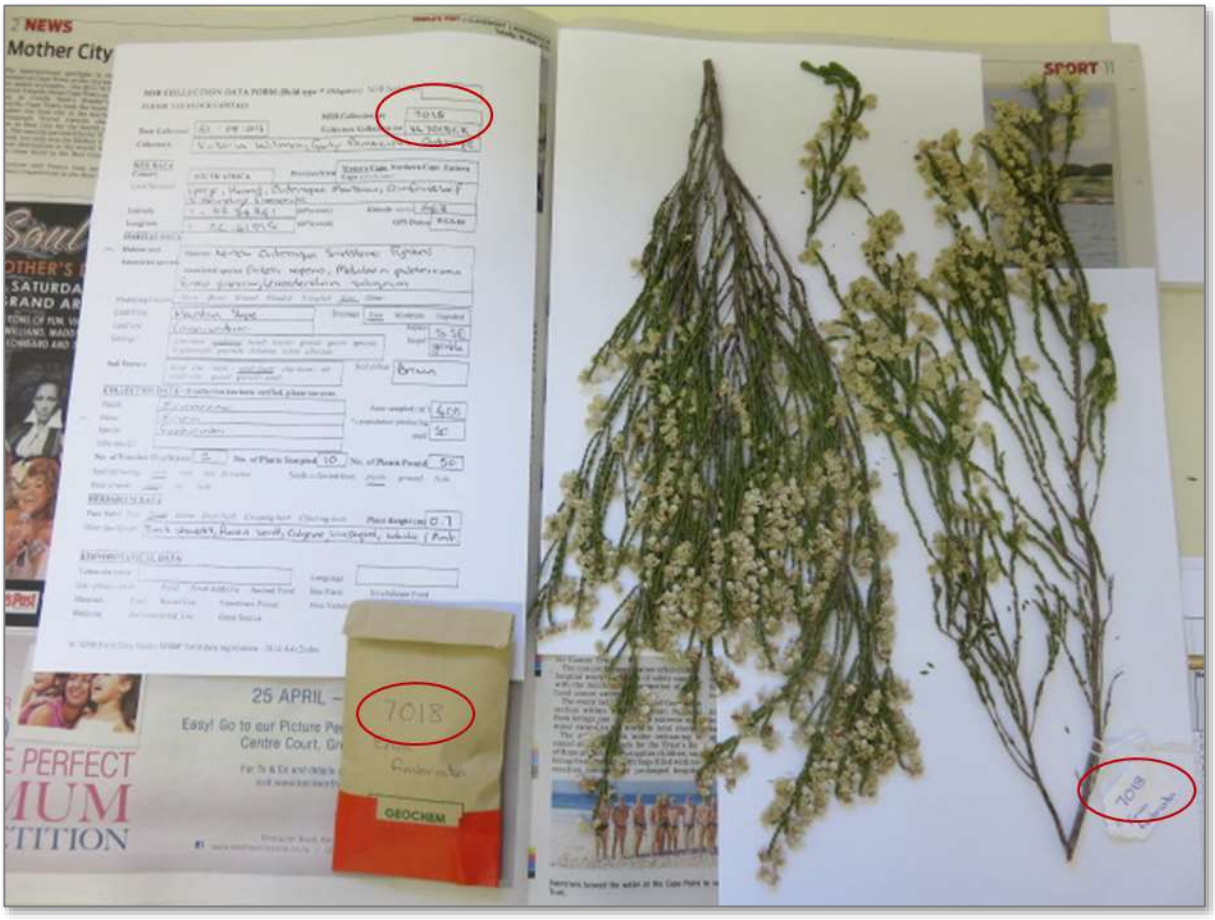

Figure 1: Seeds, herbarium voucher and data linked to unique numbering system *(Wilman, V; Mdayi, 2019)*

Data Management: The collection number is the single most important reference for the material collected. Agree beforehand what numbering system the team will use and keep records extremely carefully to avoid danger of number duplication. Use the same number for herbarium material and associated seed and data collected. The same filing system should be used electronically for herbarium photos and scanned voucher specimens.

6 Species location

Locating species for plant or seed collections are guided by planned developments, rehabilitation requirements, conservation objectives and genetic variations of important species.

BMM is required to ensure harvesting of specific areas according to their planned developments and associated flora permits. A consolidation of all the BMM flora permits were done according to areas where species and number of specimens were logged for search, rescue and restoration before site clearance of these areas commence.

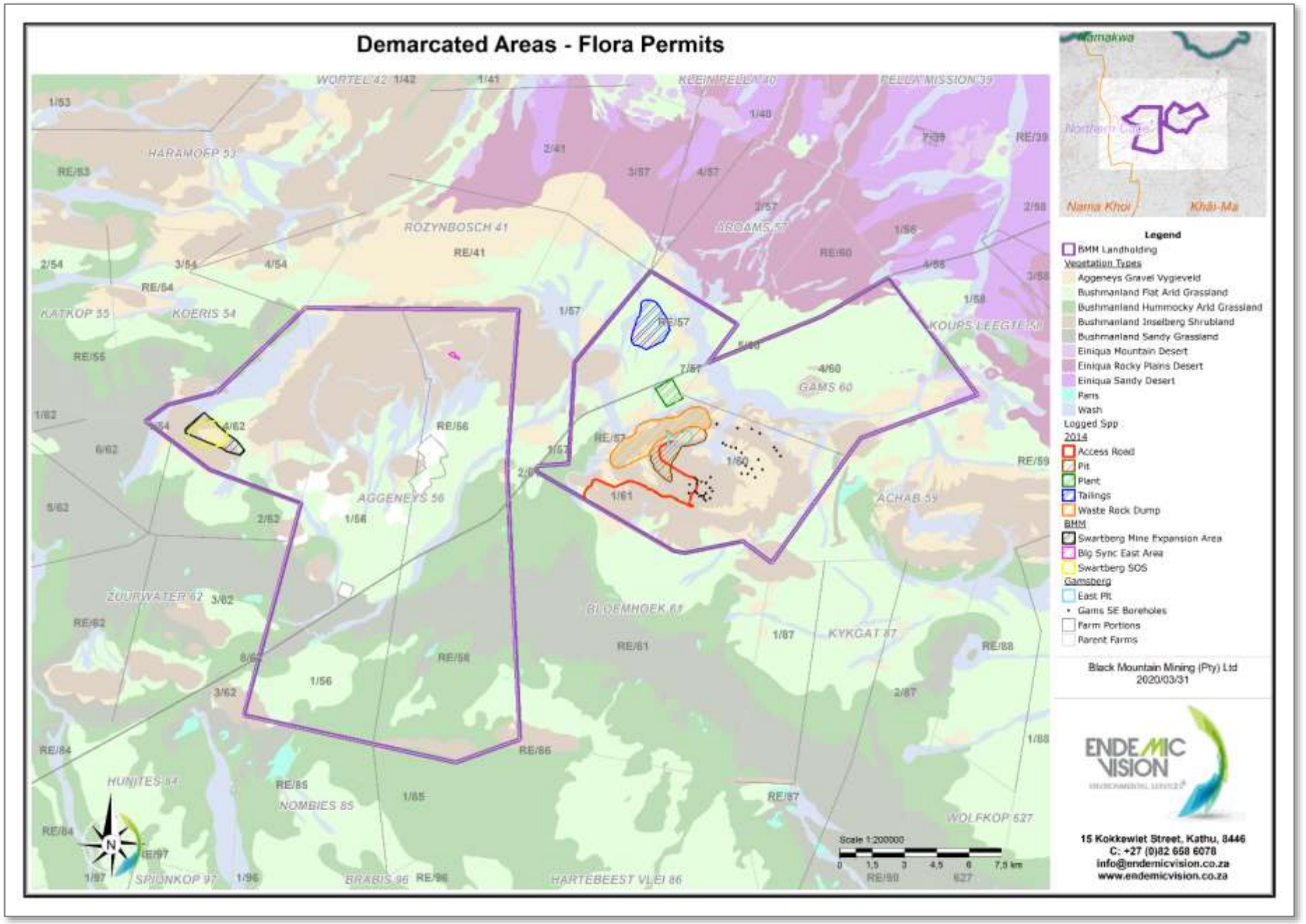

Figure 2: Vegetation harvesting areas as required by BMM flora permits

In order to comply with protected flora legislation, the impacted area must be demarcated and the number of listed species that will be affected recorded.

Species and estimated populations recorded in terms of flora permits should be used as the minimum scope of harvesting for a specific area.

1. Succulent shrub and open habitats should be demarcated into drive count strips. The width of one drive count is the number of persons available for the drive count x 500 cm. (5 persons can conduct a drive count of 25 meters wide)
2. Species that are harder to spot and those which may be invisible by the time of collection must be flagged by identifying and marking preferred habitats for these species in 10m² areas. (for example, quartz patches).
3. A drive count methodology should be applied to find specimens before collecting with persons 0.5 meters apart for succulent shrub and open habitats to identify and mark specimens for collection.
4. Succulent and Quartz patches require persons to walk directly next to each other and to overlap each person's view-frame in order not to miss specimens.
5. Colour markers (wire pegs with tape) should be used according to plant type to assist in grouping plants for "similar" handling and transplantation.
6. Plant functional groups are referenced from the Flora monitoring protocol – habitat complexity determination as:

Table 1: Plant functional groups

1	Lichens, Ferns and Mosses
2	Forbs
3	Geophytes
4	Grass
4a	Dwarf perennial tuft grass (*E. meluiria*)
4b	Annual
4c	Perennial tuft grass
5	Succulent
5a	Stone plants
5b	Weedy succulents
5c	Leave succulents (woody stem)
5d	Stem succulents
6	Shrubs
6a	Dwarf shrub (<30cm)
6b	Shrub (>30cm)
7	Trees

7. After each patch / drive count strip is assumed to be cleared of species of interest, a single person is to zigzag the patch to ensure no material was overlooked.

The scanning and logging of an area for plant or seed collection can also use the BMM pre-clearance report template that is required in terms of the BMM ISO14001 system for Black Mountain Mining. The pre-clearance report requires the logging of species that must be collected before the contractor may access the site. After contractor plant collecting the site clearance contractor receive the go-ahead (site clearance approval) to clear the site (remove topsoil and construct).

7 Collection of Plants

Succulents is relatively easy to harvest and transplant. The flora data sheet, as adapted from Millennium Seedbank, KEW Royal Botanical Gardens, UK should be used for all plant/ cutting or seed collection taking place (Appendix 2).

The optimum time is before plant hibernation; when ambient temperatures decrease and evaporation / trans evaporation reduces.

For this site, Autumn is the best harvesting and translocation time. Harvesting and relocation can continue into winter with awareness about potential frost that could kill the plants. (Transplanted plants may have higher moisture levels at this time as appose to veld plants that have retracted moisture levels).

The optimum approach is to harvest the plant with its root structure in-tact and the soil pocket within which the roots are situated and transplant them directly to similar surrounding habitat. This is relatively easy considering the shallow root system of the plants.

Some succulents are seated between pebbles or rock crevices. A small fork or hand spade should be used to carefully loose them.

It is important to note the aspect and local environment within which succulents are located in crevice's in order to try and replicate this environmental condition at transplantation.

Some succulent and/or indigenous plants cannot be harvested / transplanted successfully. These plants must rely on sufficient seed harvested to make out part of the restored area diversity.

Geophyte collection steps

Some bulbs may retract deep into the soil, in which case it may be necessary to pierce a spade as deep as possible before wedging. If the soil does not accommodate a spade, one could make use of a pickaxe or similar implement to remove soil around the bulb. Collect in tubs with water or wet bags in an upright position.
Cut back a third of the leaf material (if any present).

Succulents (stone, weedy, leave, stem) collection steps

1. Dig all around the root ball area;
2. Deep enough as far as the trowel will go before carefully wedging the plant out of the soil from four positions around the plant. If the plant only has a single taproot, then care should be taken to continue digging deeper until the whole root can be safely removed without breaking any part of it;
3. The root ball should be intact and with as much soil clinging to it as possible;
4. For sensitive species, such as *Conophytum angelicae subsp. angelicae,* the process is the same with the exception of applying more care and patience. In this case, the prime concern is not to disturb the junction of the central root and the base of the plant;
5. Move the plant over to a tub while still resting on the trowel and kept in position with the other hand;
6. The tub is to contain some hydrogel in which the plant is pressed into as to cover the root ball but not the rest of the body and leaves. This will keep the soil on the roots moist and prevent it from dropping off;
7. Remove a bit of extra soil from the hole and reserve in another tub to use at receptor site or nursery;
8. For larger plants removed by spade, it may be necessary to wet the root ball before removing it from its hole; and
9. Place inside a tub or wrap with a medium weave hessian material.

Shrub and Tree collection steps

Large succulent shrubs and trees (like quiver trees) should be loosened at the base of the plant. Note the tree orientation (aspect, what is the front facing part of the tree, which part is facing South) for translocation replication. Be aware that shrub / tree succulent branches are weak and fragile and need to be supported by persons or ropes when laying the plant down. A number of persons are required to lay the plant down after excavation to limit damage / bending of the main trunk or branches. Large specimens should be stored under a moist cloth in shady areas until it can be transplanted. Large succulents are known to successfully store for more than one year in shade/ moist cloth covering. It is very important to transplant the plant at the same

aspect; orientation, habitat and depth as the original plant (may need to mark base of plant upon excavation). Ropes or stones must be used to support the plant that cannot stand upright directly after transplantation.

Succulent shrubs can be considered for collecting cuttings. Allow the cuttings to rest in ambient conditions until a callus has formed at the cut, otherwise a root would not be able to grow from it. Propagate with 'seed sowing mix' (as seen in the Nursery Management Plan) in a multi-cell tray.

Collect succulent stemmed plants or from shrubs with woody stems but with succulent leaves, such as some *Ruschia* shrub species. However, the cut needs to be treated with hormone powder before placing in the growth medium. Make sure to collect from a variety of shrubs within the same species so as to ensure enough genetic variety. This is because all cuttings will be clones of the source plants.

Boscia albitrunca trees cannot be relocated due to its deep tap root, but grow well from seed or cuttings. Root and shoot cuttings (1cm x 15cm) and even truncheons (10cm x 2-3m large branches) can be collected.

Transportation of collected specimens

Succulent shrubs and trees should be transported in cool, shaded environments with moist cloth covering them at all times. Smaller succulent specimens should be wrapped in newspaper to serve as branch/leaf stabilization and to insulate them. Succulents should not be transported on open vehicle with direct sunlight; wind heat/chill factor accelerating both plant metabolism and desiccation while uprooted.

8 Collection of Seeds

8.1 Identifying areas and species for seed collection

In identifying and recording species for seed collection, the first step is to determine how much seed is required on site in terms of flora permits and for rehabilitation; what species are required from SANBI and at KEW millennium seedbank.

Visit potential locations early in the season when the plants are in flower to make herbarium vouchers and to mark the plants and make plans for collection strategy (time, logistics, resources, data management).

As far as possible, try to identify the species that will be collected. Where species names are not immediately known, use a pseudo name for that specific species and replace this name with the species name once identification has been confirmed.

It is important to understand what species are collected, but in some cases accurate identification may be difficult. This can be substituted by accurately supplying the following information:

1. The first step is to identify the plant functional group (see table 1).
2. Acquire photos or photocopies on prepared herbarium specimens for targeted species. These can be obtained from Compton herbarium in Cape Town or some online sources. Compare these with the site specimens and complete draft identification of species.
3. A close-up, scaled photograph illustrating clearly the key identification features e.g. number of stamens, leaf venation, stipule shape, etc.
4. Comprehensive identification notes entered on the field data form, with information about the presence of closely related species/any risk of hybridization.
5. A representative herbarium specimen. Ideally include flower, fruiting structure and vegetative parts. This specimen will be accessioned into SANBI and or RBG Kew's Herbarium.
6. Prepare herbarium specimens of targets if possible.
7. Quality photographs illustrating the plant and its habitat is valuable reference material.
8. Copyright is retained by the photographer (or the photographer's employer) and material will not be used in publications without permission. Please indicate whether permission is granted if photographs are sent.
9. Mark your speedbags upfront using the above data + flora data sheet before harvesting.

Evaluate the area and record:

- How big is the population?
- How many in the population?
- Is it damaged? Parasitic, burnt?
- Are the majority of the plants in seed? (Stages of phenology)
- Sub-populations? Can these be collected separately?

Data management: Set-up organized herbaria collection of voucher specimens for species that will be harvested in-situ or ex-situ. The species list of logged species that must be rescued / collected according to flora permits must be used as basis to decide which species will be investigated for harvesting.

8.2 Check seed readiness

It is recommended that seed maturation is monitored if possible. Seeds should be harvested as close to natural dispersal as possible to achieve maximum longevity in storage. Levels of insect or other damage within the seeds can be checked prior to collection. A cut test is the best way to assess this.

Look out for readiness of seed through plant assessment from flowering time on a regular basis (preferably once a week, depending on species). Any seeds found as ready to harvest, should be harvested according to permit conditions.

Seeds collected should be of high quality with 80 – 90 % viable and healthy, they should be full and not be insect-damaged.
Wild seeds sometimes show a high level of unviability. This could be due to the flowers not correctly pollinated or some seeds do not fully develop, seeds can also be insect-damaged.

Seed readiness indicators:

- Changes in fruit colour;
- Changes in seed coat colour;
- Fruits splitting and/or breaking open;
- Rattling seeds;
- Seeds hard and dry; and
- Some seeds already dispersed.

To confirm seed readiness or indicate some manner of fecundity a cut test can be performed on the seed itself. For mesembs the cut test would involve wetting of the seed container so that it opens for one to count the amount of seeds present in the locules (The Royal Botanic Gardens Kew, 2001).

Perform the cut test. Sample at least 10 seeds over well-spaced sample population by sectioning to determine a viable quantity.

A cut test is done by sectioning the seed using secateurs or a blade and using a hand lens (x10 or x20) to examine the cut seed (Wilman, V; Mdayi, 2019).

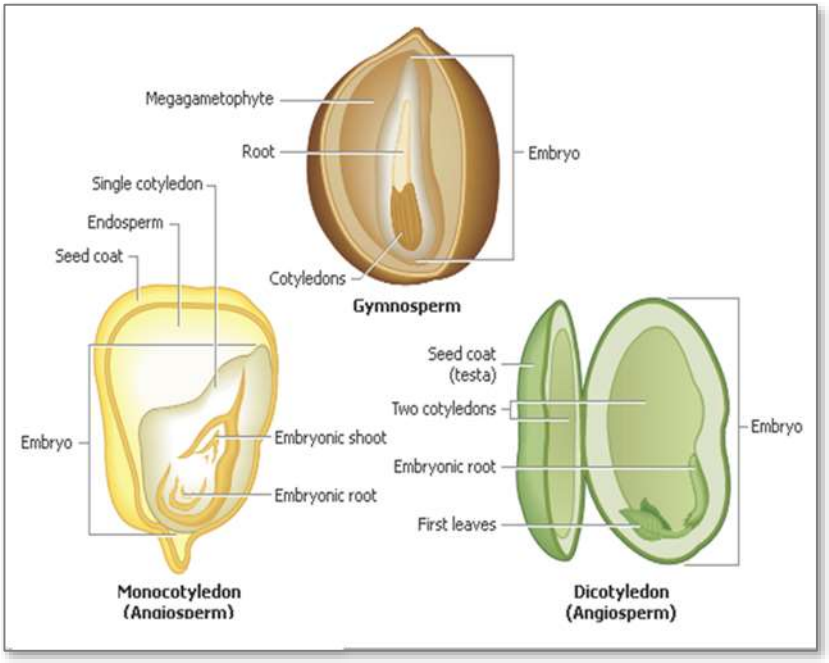

Figure 3: Demonstrating a cut test of seed indicating signs of a healthy seed (seed purity)

Be aware, look and record frequency that defunct seeds are found to give indication of that population predation. Some may be empty, undeveloped, or infested.

Assume low viability for POACEAE and ASTERACEAE but plan for extensive collecting. All species need to be well represented in the seed bank for future restoration and research efforts.

Data management in terms of identifying species for seed collection includes:
Mapping areas investigated and the results from site inspections according to date. This data can be read with rainfall data and become a valuable source of plant biology information.

8.3 Determine Seed quotas

According to the Millennium seedbank, an ideal seed collection will be from many individuals (>50) and will contain between 10,000 and 20,000 seeds(UK Seed Collecting, 2019).

Assess seed availability. 100% of seeds of planned disturbance/ construction areas should be harvested. Aim to collect 20% of seed per population, per season for natural or conservation areas. Up to 50% of seed can be collected of road verges and higher production areas. 100% seed should be collected of in-situ collections.

The collection should not be detrimental to the population; therefore, this is only a guideline and the collector must use their own initiative in the field.

Aim to collect at least 20,000 seeds per species over the project period. Recent and further drought due to possible effects of climate change may require long-term planning and many seed sources to ensure this.

To determine seed quotas, count estimated seed per seed head and number of seed heads x number of branches and work out how much to collect to obtain quota.

Seed quotas are determined by:

- Permit conditions stipulating number of seeds required;
- Long term conservation objectives for the area / species – require a minimum of 500 seeds;
- Maintenance of populations (germination protocols, viability monitoring) – require a minimum of 750 seeds;
- Duplication of collections in terms of genetic / gene bank items – require a minimum of 1150 seeds;
- Redistribution of a species – require a minimum of 5000 seeds; and
- A minimum of 10 000 seeds are required for initial establishment of bare areas.

8.4 Harvest Seed

8.4.1 In situ seed collection

Seed should be harvested in pre-determined areas (decide boundaries of the harvest block). Pre-determined harvest areas should be mapped, or the track of seed harvesters can be saved and stored on the GPS for later download per list of spp harvested in that area and on that date.

Table 2: Seed harvesting methods

Plucking	Picking of whole fruits and contained in a rigid plastic container for later cleaning. This is the most basic and simple of collecting techniques, it is suitable for the following situations: • Target fruits can be easily selected by eye. • Target and non-target (ripe and immature fruits) cannot be separately collected by another method. • Fruits are easily accessible to the collector. • Seed are from fleshy fruits or many-seeded indehiscent fruits.
Shaking	Covering a branch with a cloth bag and shaking seed into it. Careful shaking of a branch will dislodge the best available seeds, which can be captured in buckets or on a tarpaulin held or laid beneath the plant. The advantage of this is that the poorly developed seed will not dislodge with light shaking. Start with a gentle tap and check each fraction carefully before shaking more vigorously, do so until the optimum 'shake' has been identified. Heavy beating of branches causes damage to the tree and resident insects may also fall into the containers. This technique is not recommended for wind-dispersed seed as it blows away with even the lightest of breezes. It can be used in the following situations: • Dry indehiscent fruit.

	• Dehiscent fruit, which are liberating medium-large seeds. • Seed that have irritant plumes (Rosaceae) or spiny trees.
Bagging	Covering a seed head with a bag so the seed could drop into it for collection on a later day. This methodology is more intensive, but allow for seed protection from predation and prevent dispersal of seed before collection can take place.
Stripping	Wearing gloves and then stripping the seed from the base of an inflorescence towards the tip into a cloth bag. This is a popular technique when collecting grasses and from grass-like plants, a panicle is grasped at the base and pulled upwards dislodging many or all of the seeds. However, this does not give the collector much opportunity to select panicles for harvesting, the seeds may vary in maturity. This technique is best suited for: • Grasses in dense stands where there are no weeds or other species present. • The entire inflorescence is at the natural dispersal age. • Where the whole calm cannot be removed for drying and cleaning.
Pruning	Use of secateurs to prune a branch containing a bunch of fruits to be processed later. Whole culms or branches can be pruned; each fruit must be individually assessed before adding to the collection. Care must be taken not to damage the plant. Pruning can be a very effective technique where: • Fruit is clustered at the terminal part of the branch. • The species is abundant and a small loss of branches or foliage can be tolerated. • Fruit is out of reach and has to be pruned to get it down.
Ground Collect	Collecting seed fallen to the ground from below the plant. Seeds can often be found below a tree or bush in abundant quantities, but is inadvisable to collect such seeds for long-term conservation. Seed lying on the ground are prone to damage from pests and pathogens. Some seeds may also have been on the ground for a long period of time and even be from the previous season's seed production. Collect only from the ground when: • The mother tree can be identified without doubt. • It is certain that you can collect seeds that have dispersed recently. • Seeds are substantially free from damage by pests and pathogens. • Other techniques or collecting options are unsuitable.

Table 3: List of Plant families and preferred seed collection methods

Family	Method
DICOTYLEDONS	
AIZOACEAE (MESEMBRYANTHEMACEAE)	Plucking (from dry pods)
ANACARDIACEAE	Plucking
APOCYNACEAE	Shaking, Bagging
ASTERACEAE	Shaking, Bagging
CAPPARACEAE	Plucking
CRASSULACEAE	Shaking, Bagging
EUPHORBIACEAE	Bagging
FABACEAE	Shaking, Bagging
GERANIACEAE	Bagging or mature seed in ambient conditions.
LAMIACEAE	Stripping
LOGANIACEAE	Pruning
MORACEAE	Stripping
PORTULACACEAE	Shaking, Bagging
SALVADORACEAE	Pruning
SAPINDACEAE	Plucking
MONOCOTYLEONS	
AMARYLLIDACEAE	Pruning
ANTHERICACEAE	Pruning
ASPHODELACEAE	Shaking
COLCHICACEAE	Stripping, Pruning
DRACAENACEAE	Pruning
HYACINTHACEAE	Shaking
IRIDACEAE	Shaking
POACEAE	Stripping

Seed harvesting require good material and data management. It is important that a workflow that will be most efficient for the contractor be set up. The data capturing requires a unique numbering / referencing system to link spp, location, date, seed collector and seed packets for future reference.

Always keep containers of collected seeds out of direct sunlight. Store in a cool and dry place before and during transit to seed bank processing. Cooler bags should be considered in very hot circumstances, separating the seed from ice packs to prevent burning. Paper bags should be used. Plastic bags will collect excessive moisture and heat.
All containers should be labelled inside and out with the appropriate collector/collection number, and securely sealed. Label containers and bags with a unique number linked to collection forms.

Data management: for each seed collection, a unique collection number should be assigned to the Seed collection. The number assigned for the collection should be the same for the seed and its associated voucher and data. Maintain a record of the number of individual plants sampled per area to produce the quota of seeds collected.

8.4.2 Ex-situ seed collection

Ex-situ seed collection as well as vegetative propagation is important to supplement seed banks for rehabilitation as well as seed banking.

The minimum standards required for seed collection by the Millennium Seed Bank is applied whether seed is collected for internal use, South African banking or Millennium Seed banking.

As a minimum seed collected must be:

1. Good quality seed of sufficient numbers
2. 2 herbarium vouchers
3. Correctly entered data entered on MSBP datasheets if possible.

Seed collection methodologies mentioned and applicable to in-situ seed collection is not repeated in the ex-situ seed collection section.

8.4.3 Selecting specific seed types

Mesemb seed can be collected by harvesting the whole sitting capsule; placing it into separate envelopes (per species, per area, per date) and placed in a dry cool place. Mesemb capsules are long-lived. Aloe seed are short-medium lived seed (1-2 years).

Unless completely certain, one must assume plant parts to be toxic and wear gloves; and wear a mask for dusty cleaning operations. Sterilize preparation surfaces and equipment after use to prevent contamination between seed batches being processed.

Secure collections of seed into cloth or paper bags for transit as soon as the collection has been completed. Any awned seed or hooked fruit that would damage or get stuck in cotton should be stored in cardboard boxes or strong paper bags.

Collect fleshy fruits directly into plastic bags and allow them to aerate. Fleshy fruits can decompose rapidly and poor storage can lead to mold-infested seeds. – These seeds should be cleaned immediately if ripe.

B. albitrunca fruits starts changing from green to yellow when it is ready to be harvested. Seeds that have fallen to the ground will mostly be predated and can't be used. The fleshy part of the fruit must be removed as soon as possible after harvesting since it is a growth inhibitor. Only the flesh should be removed, be careful to not remove the shell of the seeds. After removal of the flesh the seeds should be washed to protect them from insects attracted to the fruity smell. Seeds need to be dried out in the sun for a few hours until they start turning white. Cleaned seeds should not be exposed to any moisture and can be stored in an airtight container in a fridge for up to six months. For the best results seed should be stored and sown after winter in late autumn or early spring (Mans, 2018).

8.5 Cleaning and Drying Seed

The correct post-harvest handling techniques should be applied - immature collections ripened before drying while mature collections are placed in dry rooms which maintain15% eRH and 15°C, lowering the seed moisture content to ~ 3-7% eRH or laid out to dry and in a dry cool area. Once the collections are dried, they are cleaned to reduce bulk, reduce disease risk, remove debris and any empty, poorly developed or infested seeds are removed. Collections are initially assessed via x-ray or cut-test to ensure viability and maturity.
A "safe" moisture level for collections in the field is around 50% equilibrium relative humidity (eRH). Collectors need to take measures to promote drying of damp collections and ensure that moisture levels of drier collections do not rise. Seed life span approximately doubles for every 10% reduction in seed eRH.

Even if you are not able to measure seed moisture status or ambient RH and temperature you can still make informed decisions about postharvest handling:

1. Take note of the prevailing weather conditions - if it has recently rained, seeds may be wetter than expected.
2. Observe seed and fruit morphology - seeds enclosed within indehiscent fruits may be at a high moisture level.

3. Seeds which have only just reached the point of natural dispersal may be at a high moisture level, even if ambient conditions are dry and warm.
4. Depending on seed moisture status, ambient conditions and seed maturity, handle collections as follows:

    a. Spread seeds out in a thin layer on newspaper, in partial shade.
    b. Raise seeds off the ground if possible, to allow air circulation.
    c. Re-pack seeds at night and place them within the collecting vehicle to minimise moisture absorption as ambient RH rises.

Make sure that the correct label remains with the collection during drying and re-packing. If ambient conditions are not suitable for drying, use a desiccant such as silica gel (see box, right). Alternatively, spread collections in a thin layer in an air-conditioned room. If none of these options is feasible, get the seeds back to the seed bank as soon as possible, where they can be dried to safe moisture levels.

Seed must be cleaned as soon as possible after harvesting and stored in separate packets to prevent predation, fungi or disease to spread from one batch of seed to the next.

Table 4: Seed cleaning methods

Sieving	Stack a series of sieves with various mesh sizes and place the largest mesh size on top. Place the seed and debris mixture on the top and crush with a rubber bung. If seed is too sensitive, use rubber gloves instead. Continue with each sieve until the seed collects further down and the 'dust' falls right through to the bottom.
Rubber mat rolling	When rubber on sieve method proves too destructive for the seeds, use rubber gloves to crush and separate on a rubber mat.
Bag crushing	Best for pod-like fruit, could be used for mesemb capsules.
Capsule shaking	Shaking capsules that are open enough for seed to easily fall out.
Fleshy fruits	Needs to be cleaned as soon as possible. Section the fruit with a scalpel, scrape out seed and place on a sieve with a mesh size that is smaller than the seed. If necessary, run cool water over it to help remove the flesh. Finally, rinse seeds with warm water, but never hot. Rest seed in ambient conditions for 2 weeks before moving to drying conditions.

Seed should be dried for local long-term storage to increase longevity. After drying seed can be stored in fridges or freezers for storage. Seeds that will be dispatched follow partial cleaning and drying before dispatching (see below).

Table 5: Seed drying methods

Ambient condition drying	Seeds obtained from fleshy fruits or those that are still immature are to be kept in cloth bags in ambient conditions for 2 weeks. This is in order for them to finish ripening and drying slowly before moving to the drying room conditions. A special cage should be constructed for this, located on the southern wall of the seed bank.
'Wet' seeds	Very fresh seeds or those collected close to rainy conditions need to be dried as quickly as possible. This is done by placing them in a folded sheet of newspaper for the day and then transferring them to an airtight container of silica gel.
Resting	Seeds that are already quite dry at the time of collecting can be kept in their cloth bags and spaced in crates in the drying room.
Moisture monitoring	An inexpensive method to monitor seed moisture is by keeping the cloth bag in an open silica gel container with a moisture indicating sachet, which changes colour as it dries. These indicators test for moisture content between 20 to 25%. A hygrometer should be used to test to 15% moisture content, which is what it would need to be for freezing.

Figure 4: Collections are laid out in trays and placed in cool/ambient drying conditions

Immature seeds are ripened before drying. To ripen the seeds, note the following:

- Don't remove seeds from fruits, or fruits from branches or stems;
- Dry them slowly for 1-2 weeks, under natural conditions; and
- If ambient conditions are particularly dry or hot, you may need to slow the drying rate down, for example by enclosing the fruits in a permeable bag or ventilated container.

Figure 5: Immature seed being ripened before drying (ex-situ)

To clean the seed, remove empty, poorly developed and insect-infested seeds and debris. If you are unable to clean the collection they can be dried and dispatched as is.

Figure 6: Cleaning seed

8.6 Seed Dispatching

It is critical to the health and longevity of the seed that it is dispatched to the seed bank within a few days of collection, together with the completed field data forms.

Voucher specimens, photos and any other additional information may be sent at the same time or later quoting the collector's name and the number given to the seed collection.

It is important to confirm with the seed bank (SANBI / RBG) what cleaning and drying is required by you before dispatching, in general, keep the seed collections in a cool, dry place prior to dispatch but please do not refrigerate or freeze them. In some cases, Kew processing staff will be responsible for cleaning the collections on arrival at the seed bank.

Damp collections should, as soon as possible after harvest, be spread out on newspaper to dry naturally, either outside in the shade or in a well-ventilated room, before dispatch.

Fleshy fruits may require careful handling, partial cleaning and rapid dispatch to the seed bank: contact the MSB as soon as possible for advice.

Seed, herbarium specimens and Data are sent to MSBP Cape Town. Package them in labelled paper or cloth bags, and together with pressed and dried herbarium specimens and datasheets contact MSBP in Cape Town (see below) to arrange for shipment to the seed processing unit at Kirstenbosch National Botanical Garden.

Figure 7: Dispatching harvested seed to banks

9 Transplantation areas

Areas for transplanting collected specimens and seeds are prioritised according to available rehabilitation areas of the same vegetation and habitat type.

It is recommended that a detail rehabilitation design be made for each rehabilitation area based on soil sampling of the site and the reference site to ensure vegetation establishment and persistence will be successful for the transplanted / sown species.

A mapping overlay was prepared of available rehabilitation areas in terms of the BMM complexes, including prospecting, mining, farming areas owned by BMM.

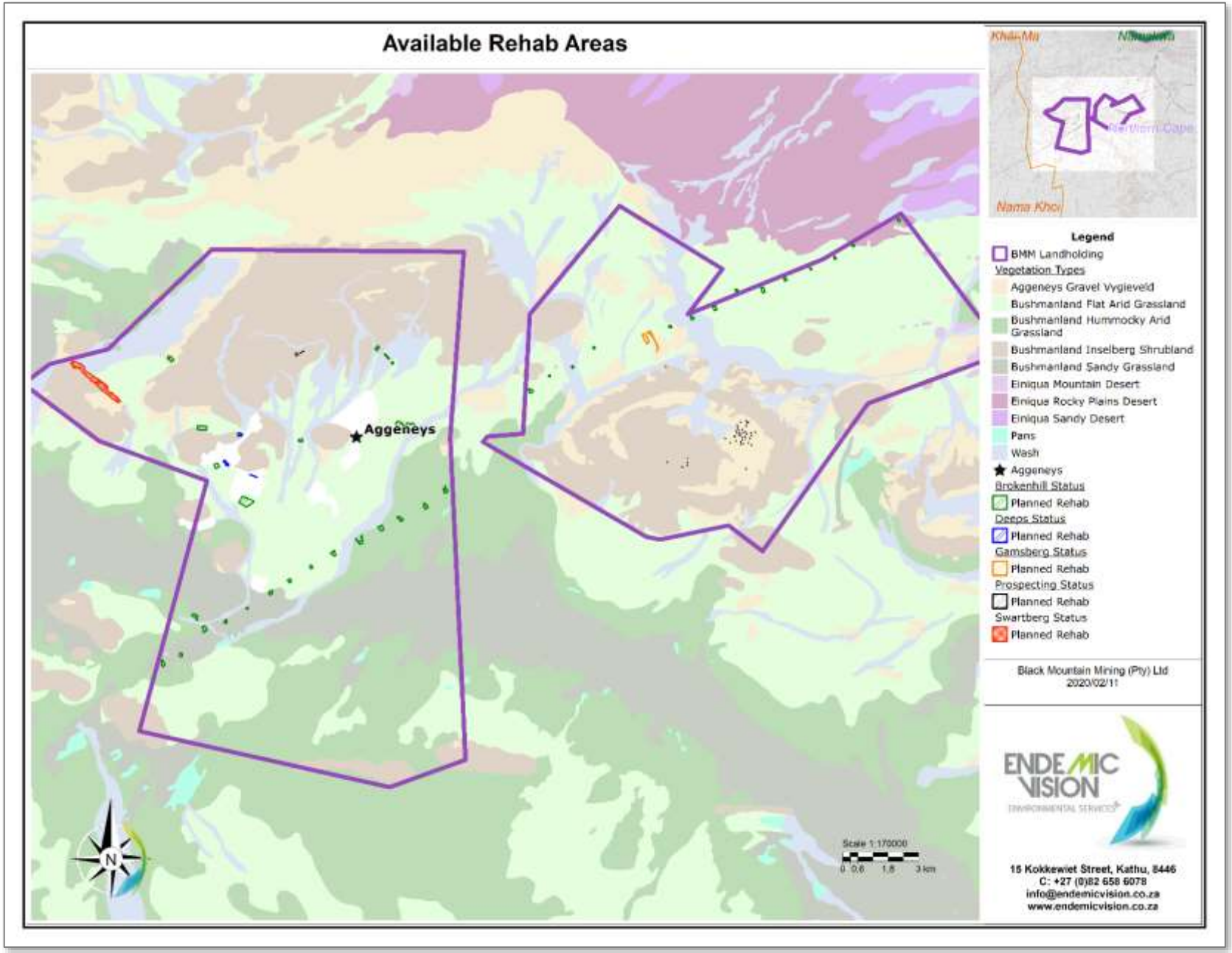

Figure 8: Areas available for rehabilitation and translocation

The maps indicate that the following hectares of the different vegetation types are available for rehabilitation:

Table 6: Areas available for rehabilitation according to vegetation type

Vegetation Type	Area size (ha)
Aggeneys Gravel Vygieveld	11,68
Bushmanland Flat Arid Grassland	58,46
Bushmanland Hummocky Arid Grassland	12,15
Bushmanland Inselberg Shrubland	32,11
Bushmanland Sandy Grassland	11,83
Einiqua Rocky Plains Desert	1,52
Transformed	4,35
Wash	8,80
Grand Total	140,90

The species list of all specimens harvested are included as appendix 3 of this protocol.

The areas where translocation have taken place and is presented below and indicate important monitoring areas. Transplanted areas should be monitored for survival; plant production; plant community development of the different transplanted species and methods applied to increase future success of transplantation projects.

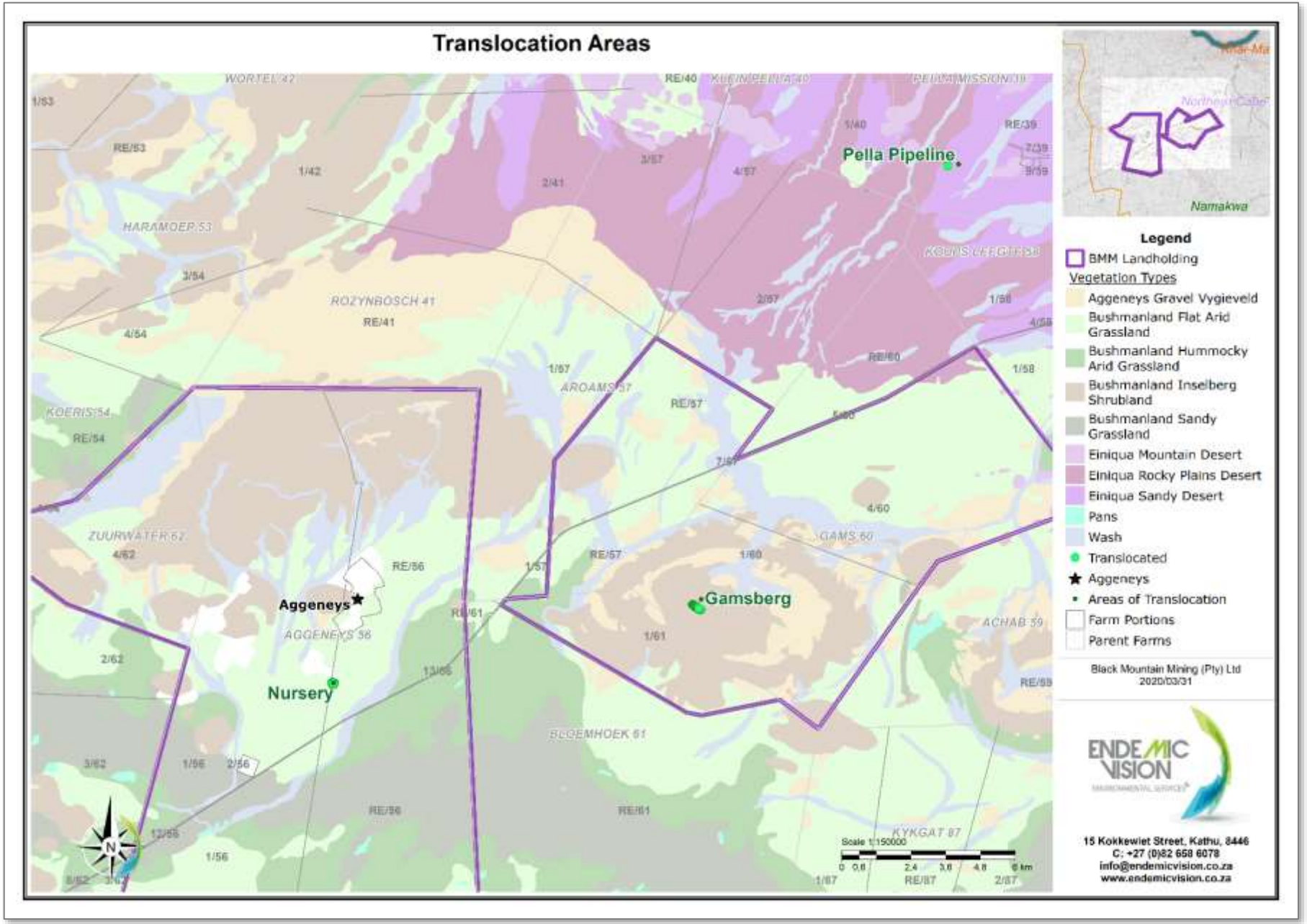

Figure 9: Areas where transplantation has been completed

10 Transplantation

10.1 Transplanting bulbs

Be sure to place the bulb in its hole with growth point of the bulb pointing upwards.

10.2 Transplantation of succulents

Succulents can be transplanted to the nursery for the following reasons before translocation into disturbed habitats:

1. Time of year not suitable for direct transplantation (high environmental stress, low survival);
2. Area for relocation is not ready for transplantation yet;
3. Too few specimens are collected and vegetative / seed collection to increase number of specimens must be done before transplantation;
4. Plants are already under stress (drought, disturbance, predation) when collected and need to be cared for before transplantation (stressed plants will not survive); and
5. Plants are in full production (plants should be removed only after seed collection).

In this case, succulents should be transplanted into sandy growth medium consisting of 2 parts loam soil (or topsoil) / one-part sand or quarts / calcrete or calcine pebbles / one-part organic compost. Initial transplanted plants should be shaded to reduce heat/ sunlight stress. Initial watering is necessary to buffer and establish transplanted plants. Once it is determined that the plants have established (is not 100% dependent on its own succulent reserves) watering can be delayed for a couple of weeks. Watering will then take place as plants indicate water stress.

10.3 Transplanting succulent trees and shrubs

Small to medium succulent shrubs and/or indigenous shrubs should be transplanted into pots or plant bags to aid movement; facilitate translocation with sufficient plant material and to reduce root exposure during transplantation.

Larger specimens such as Aloes and Tylecodons can be planted directly into the ground in the nursery unless a suitable permanent site(s) have been identified for their immediate relocation.

To ensure best possible survival the following steps are recommended:

1. Prepare receptor site with a minimum hole size of 0.5m x 0.5m x 0.5m.
2. Prime the bottom of the hole with 5cm layer of stones (Optional if additional drainage is required).
3. Mix removed soil, compost and bone meal and fill hole 70% with this material.
4. Fill the hole completely with water.
5. Orientate the individual plant by correctly placing the marked northern side on the trunk to face north.
6. Repair damaged trunk / roots immediately before transplanting - make a clean cut to damaged roots and apply fungicide.
7. Position the tree on top of the mixed soil and set slings and ropes for anchoring. Anchor the tree before final soil covering (tree must stand and not be supported by soil covering as this will not last)
8. After 48 hours, test anchors and fill hole with remaining sand.
9. Make sure the filled hole retains a depression (bucket) after filling to collect water in the landscape.
10. Make sure the tree is not planted deeper or shallower than its original planting depth.
11. Compact the surface soils to ensure no air pockets remain on the roots.
12. Water generously.
13. Initial watering will be required to facilitate the plant through transplantation stress.
14. Soil moisture meters should be used to determine watering frequencies.
15. After 6 months the tree should be adapted to its new location and watering may stop.

10.4 Transplanting *B. albitrunca* saplings

Sites chosen for transplantation should have access for watering and follow up watering for optimal survival. Planting must take place in the Winter when the production requirements of the plant is less and the chance of survival is higher. Saplings should not be kept in potting bags for too long as this stunts their growth. Saplings should be brush packed to protect them from predation.

The following steps must be taken for transplantation to ensure optimal survival:

1. Holes must be at least 5m apart.
2. Holes must be twice as wide as the bagged sapling for optimal growth.
3. Holes must be deeper than the bagged sapling to allow for root growth.
4. Loose soil must be placed back in the hole to reach the required depth for the bag (roots will grow easier in loosened soil)

5. The sapling should be deeper than the surrounding soil when planted, to create a "bucket" for water catchment.
6. Fill the hole with water prior to planting and allow the water to seep into the soil.

Figure 10: Prep hole with water, bone meal and compost

1. Add 100g bone meal to the hole, for root production.
2. Add 150g compost for growth.
3. All saplings must be given water prior to planting.

Figure 11: Carefully remove sapling without disturbing the roots

4. Care must be taken when transplanting the saplings to avoid any root damage.
5. Carefully cut the pot bag on both sides and pull the bag down.
6. Place one hand underneath and one on the side of the sapling and do not let the soil around the sapling fall apart.
7. Place the sapling in the hole and fill the hole with the removed soil until level with the planted sapling.

Figure 12: Compress sapling

8. Compress the planted sapling by pressing firmly on top of it and by stepping on the loose soil around it, ensuring that the sapling will not collapse and that there is no air trapped within the root system.

Figure 13: Add compost and mulching on top

9. Add a thin layer of compost on top.
10. Fill the hole with water once again.
11. Fill the rest of the "bucket" with mulching to aid in keeping water from evaporating.
12. Brush pack the holes to prevent predation of the saplings.
13. Follow up watering is recommended.

11 References

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