# Wild Honeybush (Cyclopia intermedia) Augmentation

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By

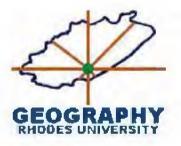
Paul-Luc Michau

G16m3658

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Supervisor G.K McGregor





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## Thesis Abstract

Honeybush (*Cyclopia* spp.) is an endemic legume to the fynbos region of South Africa, and certain species of the genus are used to make health tonics. There are growing international markets for such products and, currently, wild populations of the species *Cyclopia intermedia* are disproportionately relied upon to support this increase. Due to the sensitivity and ecological complexity of the resource, serious concerns about sustainability and regulation have arisen. The social landscape of the resource is equally as complex, and solutions for alleviating pressure on wild populations, such as reducing or banning harvesting, and switching to commercial cultivation are untenable in every case. A possible option, which has been used in the wildflower industry, is to augment (increase) wild populations by planting seeds or seedlings directly into the veld.

The main focus of this research was to explore the viability of using augmentation to boost wild populations of *C. intermedia* through field trials. In support of this broad aim, research objectives addressed understanding the ecological habitat conditions of the species, quantifying the economic potential of augmentation, characterizing wild populations and conducting seed germination tests.

GIS methods were used to locate sites most representative of natural conditions on different post-fire aged veld on a farm in the Kouga mountains. The study characterized habitat conditions by collecting soil and vegetation samples at plots where honeybush was present and absent within close proximity. Vegetation diversity was only slightly higher in presence plots, and no clear pattern between absence and presence plots was observed for soil properties. However, post-fire site age did significantly influence certain nutrient levels.

The augmentation trial results indicate that seedlings have a much higher survival rate than seeds on average. Survival success varied unpredictably between plots where wild honeybush was already present and adjacent plots where it was absent. Post-fire site age influenced seedling growth and seed survival positively with the newly burnt plots showing the best growth rates overall. The cost-benefit analysis reveals that seeds are a better option as they require low-cost inputs and carry less risk, unlike seedlings which may have higher survival but are more expensive. The study recommends using trials to assess survival before choosing an option.

Out of five natural populations of honeybush that were assessed only one showed seedling recruitment and three young plants were found between two of the other sites. Growth characteristics of populations showed expected variability based on site conditions and disturbance history. Growth appears to be rapid in the first-year post disturbance, thereafter slowing down. Average stem numbers and basal circumference increased concurrently for both harvested and unharvested populations. Due to the influence of natural conditions such as slope aspect and soil properties on plant morphology, it is best to compare harvested and unharvested individuals from the same site and preferably within close proximity to minimize such differences.

Using seed sourced from the same parent population is critical to preserving genetic integrity and little is understood about germination characteristics from wild *C. intermedia* populations. The study tested germination rates between green and brown seed and alternative collection methods. When comparisons were done within farms on two occasions collection and seed colour did have a significant influence. Overall, the influence of which farm the seeds were collected from appeared to play the biggest role in determining germination.

## Chapter 1

#### 1.1. Introduction

Honeybush is an endemic South Africa herbal plant used to make tea and, more recently, a variety of other products such as flavour enhancers and natural preservatives (Kokotkiewicz and Luczkiewicz 2009; Joubert *et al.*, 2011). It has a long history of traditional regional use in South Africa, with the earliest official documented record of use dating back to 1705 (SAHTA, 2019). Over the past few decades, the demand within developed countries for herbal teas has grown substantially, and products such as Rooibos (*Aspalathus linearis*) and Honeybush (*Cyclopia* spp.) are increasingly being consumed (Schulze *et al.*, 2015). Honeybush is now exported to more than 25 countries, with the largest consumers being nations such as Denmark, the Netherlands and the UK (Joubert *et al.*, 2011). Coupled with this, rising interest in the unique biochemistry of the plant could mean that future demand may spike unexpectedly if new products are developed (Alexander, 2018). As a result of the increase in consumption, wild populations are heavily relied upon to satisfy the industry's needs (McGregor, 2017). This has resulted in stakeholders raising concerns over the management of wild honeybush and the need for future solutions.

Currently, the material for production is derived from both wild and cultivated crops, although on average, 80% comes from the wild (2012-2016) (McGregor, 2017). *Cyclopia intermedia* is the most prolific and widely distributed of the genus and the overwhelming proportion of the wild crop (85%) still comes from this single species. Considering the above, the sustainability of the industry therefore needs to be secured by ensuring that overexploitation of wild populations does not occur. This can only be achieved if the rate of extraction does not exceed the rate of regeneration (Polak and Snowball, 2017).

The growth of the industry has been met by an effort to increase cultivated varieties (De Villiers and McGregor, 2017). The ecology of the genus *Cyclopia* however, is complex and factors such as slow growth rates and phenolic composition which influence taste, complicate cultivation. A fairly expensive nursery setup is required especially if vegetative propagation is done which yields faster and better results (Joubert *et al.*, 2011).

Some reseeding species grow vigorously and can be harvested annually under a cultivation setting, however sprouting species such as *C. intermedia* grow far slower and require harvest intervals of 4 or 5 years (McGregor, 2017). Considering this, the estimated break-even time for planting this species is 7 years in a cultivation setting on lands (Joubert *et al.*, 2011) which means that only after 11 years will the crop become profitable. As a result, cultivation is not a viable option for many of the current honeybush landowners. Additionally, there are other barriers to cultivation, such as lack of arable land, livelihood dependence, and cultural ties to wild harvesting (McGregor, 2017). Therefore, the practice of wild harvesting will not be easily replaced.

In connection to the above, most honeybush research has focused on the biochemical properties and commercial production of species that are more suitable candidates than *C. intermedia*. Research about the wild propagation potential of *C. intermedia* is almost non-existent, but, anecdotal trials have been done by some landowners, and experienced industry members suggest that trials should start where the plant naturally occurs before trying to commercialize it. Similarly, other aspects of augmentation which are essential to understand before undertaking projects such as economic inputs and outputs, wild habitat characteristics, and seed germination have not been researched. The sensitive ecology of the species and apparent low seedling recruitment rates also add reason for concern if natural populations are being increasingly pressured (G.K. McGregor, *pers. comm.* 2021). Much like other indigenous fynbos crops such as the wildflower industry, altering natural processes through augmentation raises a number of conservation concerns (Treurnicht, 2010). Research is key in overcoming future resource use issues through understanding how to regulate the industry.

The motivation for this research thus comes from multiple sources, namely the sustainability of the industry, the prevalence of wild harvesting, the lack of research, and conservation pressure. This thesis aimed to provide background knowledge to these areas and provides a basis from which longer-term monitoring of the trial sites may be conducted. This research was also motivated to develop preliminary recommendations that will complement other work that has been done, such as the guidelines for harvesting wild populations of *C. subternata* and *C. intermedia* (McGregor, 2017).

#### 1.2. Research Aims and Objectives

The primary focus of this research was to determine if wild populations of C. intermedia can be augmented for the purpose of contributing to sustainable production. In order to achieve this, a number of research objectives were identified. This thesis deals with each objective in different chapters, referred to as chapter aims. Thesis structure lends itself to publication format. Objective 1 (chapter 2) aimed to bring together a review of literature around each topic of importance in order to identify knowledge gaps and what approaches should be taken to address these gaps. Objective 2 (chapter 3) deals with identifying areas of optimal growth and choosing planting sites for the augmentation trials using GIS methods. Additionally wild C. intermedia habitat conditions were characterized through the collection of biophysical data (soil and vegetation surveys). Objective 3 (chapter 4) deals with the augmentation field trials and a cost benefit analysis of the main inputs and outputs involved with the process. Objective 4 (chapter 5) investigated the population characteristics of wild C. intermedia populations on a farm in the Kouga. Additionally, chapter 5 aimed to assess the effect that seed collection method (handpicked and netted) and seed colour (green and brown) has on germination. Seeds were collected from different populations spread across the study area. Objective 5 (chapter 6) is the synthesis of the previous chapters and preliminary recommendations for augmentation.

#### 1.3. Study Area Background

This study is focused on four farms spread across two broad areas namely, the Langkloof and the Kouga Mountains situated within the Eastern Cape province of South Africa. Both areas fall within the larger Cape Floristic Region (CRF) which is a Mediterranean-type biome recognized as a global hotspot for floral endemism and diversity (Manning and Goldblatt 2012). The Langkloof itself is a valley region created by a series of Cape fold mountain belts, the area begins in the south western reaches of the Eastern Cape near the town of Kareedouw and extends just beyond the border of the Western Cape to the town of Haarlem and is bounded by the Tsitikamma mountain range to the south and the Kammanassie and Kouga mountains to the North (Geldenhuys, 1997). The area is further divided by a series of hills running parallel within the kloof, creating northern and southern sections. The southern section is known as the Klein-Langkloof and is considered the hub of South Africa's apple production (De Kock, 2015). Other agricultural activities include intensive monocropping and dairy farming at lower altitudes in the valley bottoms and stock farming in the higher mountainous areas (De Kock, 2015).

Honeybush is also an important resource for many farmers and community members in the Langkloof and Kouga, as it not only accounts for a substantial portion of their income but is also seen as part of their identity and livelihood (McGregor, 2017). The study sites considered to be in the Langkloof (farms 2, 3 and 4) are located on the interior side of the Tsitsikamma mountains which receive 500-750 mm of rainfall annually and have an average maximum and minimum temperature of 24 and 5.5 degrees Celsius (Rebelo *et al.*, 2006). In contrast, the Kouga (farm 1) has a much drier climate with greater extremes, despite being less than 60 km away. The average rainfall ranges between 350- 450 mm annually with a mean maximum and minimum temperature of 28.3 and 2.3 degrees Celsius (Rebelo *et al.*, 2006). Although a sheer climate gradient exists towards the interior, both the Tsitsikamma and Kouga mountains share general habitat conditions indicative of mountain fynbos. Shallow and nutrient poor soils derived from quarzitic Table Mountain and Witteberg geologies as well as periodic fires which are necessary for reproduction are two of the main defining factors (Esler *et al.*, 2014).



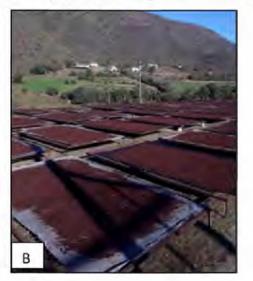


Figure 1-1: Image A) *C. intermedia* seedling 11 months post planting for the augmentation trial. Image B) Harvested *C. intermedia* material being processed for tea at Nooitgedacht (photo by T. Makhuza).

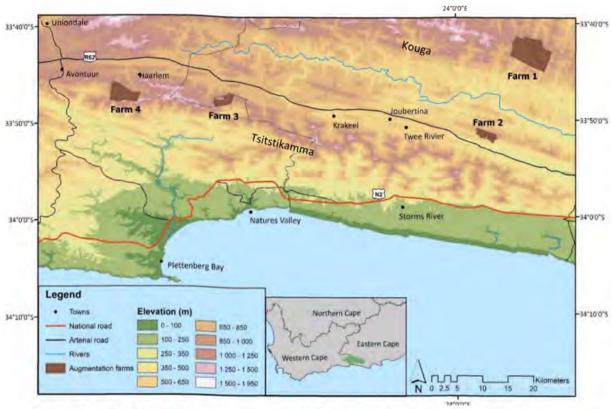


Figure 1-2: The four farms chosen as study sites spread across a the Tsitstikamma and Kouga mountain ranges

- 1.4. Assumptions and Limitations
- Table 1-1: Summary of the main assumptions and limitations of the study and which chapter they are related to

Chapter no.	Assumption	Limitation
3	Habitat conditions differ between areas in close proximity where honeybush is present vs absent	Biophysical habitat data collected from one area only (Kouga mountains)
3		Limited biophysical variables used for habitat characterization (soil and vegetation)

4	GIS site selection process is representative of optimal growth conditions	Logistic and pandemic-imposed restrictions to monitoring other sites adequately
4	Cost Benefit Analysis (CBA) inputs: discounting rates, project lifespan, labour and construction requirements	Economic information availability concerning augmentation
5	Survey site data representative of entire population structure	Wild <i>C. intermedia</i> population surveys from one area only
5	Seed collection technique and colour will influence germination	Visual bias when sorting green and brown seed and scale of germination trial

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#### Chapter 2: Literature review

#### 2.1. Ecology of fynbos and the genus Cyclopia

Fynbos is one of only five recognized Mediterranean regions globally, which are most clearly defined by their characteristic climate of mild wet winters and hot dry summers accompanied by a fire season (Vogiatzakis *et al.*, 2006). Although these characteristics mark the boundary of these regions, a large degree of climatic and landscape variability exists both between and within Mediterranean ecosystems, this internal variability produces biodiverse hotspots and drives specialized adaptations (Cowling *et al.*, 1996; Di Castri and Mooney, 2012). This is evidenced by the fact that vegetation diversity and endemism in these ecosystems rivals that of tropical rainforests. Considering that these areas only cover 2% of earth's surface and contain approximately 20% of all vascular plant species, their biodiversity turnover is disproportionate when compared to other biomes (Cowling *et al.*, 1996; Vogiatzakis *et al.*, 2006). A mixture of grass or trees with dense woody shrubland with small sclerophyllous leaves is characteristic of Mediterranean regions (Rebelo *et al.*, 2006).

An important aspect of these regions are natural disturbance regimes which refer to periodic events, such as fire and drought, that affect and determine resource availability (Le Maitre and Midgley, 1992; Pausus and Keeley, 2014). These disturbances have to a large extent shaped the ecological evolution of these systems, for example fire has become necessary for the completion of the reproductive cycle for most plants within these regions (Pausus and Keeley 2014). Long periods where fire is absent results in species diversity loss and community alteration (Privett *et al.*, 2001; Pausus and Keeley, 2014). Despite requiring disturbances, Mediterranean ecosystems are also highly sensitive to changes in the frequency and extent of these pressures (Myers *et al.*, 2000; Underwood *et al.*, 2009). This makes them particularly vulnerable to activities which change or remove disturbances such as repeated burning to increase grass cover for grazing and clearing land for crop production. In most Mediterranean areas the landscape has been changed to such a degree that vegetation patterns cannot be understood apart from historically induced human impacts (Pausas and Vallejo, 1999).

An example is the increased frequency and intensity of fires in Spain due to old abandoned farmland vegetation having more trees and increasing the amount of combustible material (Pausas and Vallejo, 1999). In South Africa the fynbos biome faces similar threats from farmland and urban expansion. Approximately 40% of the Agulhas plain, an area recognized globally for its fynbos diversity, has already been transformed (Privett *et al.*, 2002).

Fynbos, originally called "fijnbosch" received its name from Dutch settlers who arrived in the Cape and saw that the timber of this dominant shrubland was too slender for any harvesting (Esler *et al.*, 2014). Later in 1916 the term 'fynbos' was used in botanical literature for the first time, and ever since has become the accepted term in scientific literature. The name has also been assimilated into the tourism and wildflower picking sector to promote its uniqueness (Esler *et al.*, 2014). The fynbos biome is part of the larger Cape Floristic Region, which is approximately 90 000 km<sup>2</sup> (Manning and Goldblatt, 2012) stretching from Grahamstown in the Eastern Cape, westward towards Cape Town and then north to Clanwilliam.

The CFR is defined by a mosaic of biomes including succulent karoo, fynbos, thicket and forest biomes (Schutte, 1995). Most of this area which is considered to be fynbos is distinguished by the presence of three plant families, Proteaceae, Ericaceae, and the Restionaceae (Esler *et al.*, 2014). These are key in separating "true fynbos" from similar vegetation types such as renosterveld and strandveld which are also encompassed in the wider management definition of the fynbos biome (Esler *et al.*, 2014). It is within the true fynbos that the various *Cyclopia* species occur. One of the remarkable features of fynbos is the great diversity of species between geographically similar habitats. An example of this is the comparison between the Cape Peninsula near Cape town and the Kogelberg, which are only separated by a few kilometers (False Bay) yet share less than 50% of the same species (Esler *et al.*, 2014).

The above example illustrates the complexity of trying to understand exactly what determines plant assemblages and distributions within this biome, though proportion of rainfall, extent of summer droughts and soil properties are the main factors that exert an influence (Manning, 2007). Rainfall patterns and quantity vary greatly across the fynbos biome. Predominantly winter rainfall occurs in the extreme west, to the east and north east the rainfall regime is more accurately described as bimodal, although west of Knysna the summer months are driest and further east towards Port Elizabeth the winter months become the driest (McGregor, 2017a). Seasonality of rainfall is thus better described as a subtle gradient whereby the percentage of rainfall that falls in winter drops from west to east (Bradshaw and Cowling, 2014).

Underlying geology plays a pivotal role in determining where different fynbos vegetation types grow (Richards *et al.*, 1997). Most of the high lying areas are comprised of the Table Mountain and Witteberg Groups which are predominantly quartzite and sandstone, making the soils nutrient poor and acidic (Esler *et al.*, 2014). Approximately 81% of all vegetation units occur on these harsh sandy substrates (Rebelo *et al.*, 2006) and thus most fynbos species are considered edaphic specialists. In lower lying valleys and vleis, the soil is more fertile with higher amounts of nutrients such as nitrogen and phosphorus derived from the underlying softer shales and accumulated weathered rock (Esler *et al.*, 2014). In some coastal low-lying areas, the soil is more alkaline as it is derived from ancient marine deposits and limestone. Shrubby fynbos communities indicative of "true" fynbos are generally more adapted to the nutrient poor soils derived from quarzitic geologies at higher altitudes whilst lower lying areas give way to more grassy fynbos communities with less abundance of ericoids and restiods (Esler *et al.*, 2014).

All honeybush species belong to the legume family, Fabaceae, bright yellow flowers and stout shrubby stems with trifoliate leaves are distinctive features of the genus *Cyclopia* (Manning and Goldblatt, 2012). The genus received its name from the Greek work "cyclops" which refers to the circular depression in the base of the calyx where the pedicel attaches itself to the flower (Kies, 1951; Schutte, 1997). All species occur exclusively within the winter rainfall or bimodal fynbos biome, but do not extend as far as the biome itself does. Their distribution is spread unevenly along the Cape fold mountains and coastal plains from Clanwilliam in the west to outside Port Elizabeth in the east (Manning and Goldblatt, 2012). Their range is further bounded by mountains in the north such as the Kou Bokkeveld, Klein Swartberg and Kouga Mountains and their concentration rapidly declines north of the Caledon district (Schutte, 1997). Some species are far more prolific than others. Of the 23 species, some such as *C. aurescens* are endangered and confined to high altitudes (1800 m) whilst others are far more widespread and occur across a range of altitudes such as *C. genistoides* (60- 1170 m)

(Joubert *et al.*, 2011). Local names for some species give an indication of their habitat. *C. intermedia* for example is known as "bergtee" because it is only found in mountainous areas, whilst *C. subternata* ("vleitee") and *C. genistoides* ("kustee") occur at lower altitudes in the valleys and on the coastal plains (Esler *et al.*, 2014). The species of interest in this thesis will from here on be referred to as *C. intermedia*. *Cyclopia intermedia* grows preferentially along the cooler and wetter south facing slopes in greater densities at altitudes between 500 and 1700 m on sandy to loamy well drained quartzitic soils which are nutrient poor (Joubert *et al.*, 2011).

Although south facing slopes are the ideal habitat, *C. intermedia* has also been observed growing on drier north or east facing slopes. Slope aspect significantly influences microclimatic and edaphic conditions (Ackerly *et al.*, 2002) which suggests that *C. intermedia* can tolerate a range of environments. Despite this, data shows that harvestable populations are located in areas where conditions are ideal (McGregor, 2017a). Therefore, south facing slopes are more suitable from an augmentation and management perspective.

The sweet-smelling flowers are thought to be exclusively pollinated by carpenter bees (*xylocopid*) and that other pollinators such as honeybees do not actually assist in transporting pollen between flowers though they may get nectar (Schutte, 1997). Timing of life cycle phases may differ slightly due to environmental factors such as rainfall or temperature, though generally flowing occurs between August to October, and seed-set happens from November to the end of December (Joubert *et al.*, 2011; Barnardo, 2013). Because *Cyclopia* spp. belong to the pea-family (*Fabaceae*) and are thus leguminous it produces pods which contain seeds that are eventually dispersed when the pod cracks or springs open (Schutte, 1997). Once the seed lands on the ground, a fleshy elaiosome serves as an attractant for ants which carry the seed underground (Schutte, 1997).

Similar to other Mediterranean ecosystems, fire is a crucial element in fynbos vegetation functioning (Cowling *et al.*, 1994). Seasonal dry conditions coupled with hot temperatures drive periodic fires producing a major evolutionary pressure that is intimately linked to the reproductive and regeneration strategies of these plant types (Pausas and Vallejo, 1999). Two

dominant population survival strategies have been identified, namely resprouting and reseeding.

Both strategies have tradeoffs and the occurrence of either trait appears to be highly mixed within communities (Pausus and Keeley, 2014). Added to this, many species exhibit a combination of reseeding and resprouting to different degrees, which means that regeneration must to be understood as a continuum (Pausus and Keeley, 2014).

On the one end of the spectrum are obligate seeders, which rely solely on post-fire recruitment of underground seedlings as they are killed off by fires as adults (Marais *et al.*, 2014). This strategy requires that the plant produce thousands of seeds during its lifespan to ensure sufficient replenishment of the fire-cued seedbank (Schutte and Van Wyk, 1995; Marais *et al.*, 2014). Species which exhibit this trait usually are shorter lived and grow more vigorously in dense stands, allocating more resources to the production of seeds (Keeley, 1997).

In contrast obligate resprouters, as the name suggests, rely on an underground lignotuber (a storage organ) that allows the plant to resprout after fire. Most do not have seeds which are fire resistant and require inter-fire periods long enough to allow for seedling recruitment (Cowling *et al.*, 1994; Schutte and Van Wyk, 1995). The plant partitions a large portion of its resources into growing and storing carbohydrates in the lignotuber and is usually slower growing, longer lived and able to survive multiple fire seasons (Cowling *et al.*, 1994). Plants which combine resprouting from a lignotuber and rely on seeds which germinate post fire are known as facultative seeders (Marais *et al.*, 2014). Although resprouters persist through multiple fires they are still reliant on seedling recruitment for new individuals and ultimately population survival.

*Cyclopia* spp. have adopted both obligate reseeding as seen in species such as *C. subternata* and facultative seeding strategies as seen in *C. intermedia* (Marais *et al.*, 2014). Although *C. intermedia* meets the facultative seeding category it is colloquially known as a resprouter. Understanding regeneration characteristics is vital for proper management since human pressures such as harvesting affect individual plant reproductive and survival potential. For resprouting species, harvesting removes the leaves which are responsible for the production

of photosynthates. This energy is subsequently stored in the lignotuber and used when required during a resource expensive event such as recovering from a disturbance or flowering (Le Maitre and Midgley, 1992). Removing the leaves (by harvesting or fire) essentially inhibits the plants immediate ability to replenish the tuber, resulting in a decrease in resprouting vigor (McGregor, 2017a). This may be a threat as research has shown that resprouting vigor is a significant factor in predicting post-fire survival of resprouters (Marais *et al.*, 2014) and more so if a fire occurs within a short interval of harvesting as the combined stress may severely compromise the plants' ability to survive.

In terms of seedling recruitment, for both resprouting and reseeding species, harvesting the plants before they have the chance to set seed, or harvesting too frequently will reduce the number of seedlings available for recruitment over a period of time (Kruger and Bigalke, 1984; McGregor, 2017a; Slabbert *et al.*, 2019). Therefore, it is important to recognize that both resprouters and reseeders are susceptible to population declines post-fire if potential seedling recruitment is reduced in the long term (McGregor, 2017a).

#### 2.2. Industry Overview

The commercial sale of branded honeybush tea began in the 1960s, however, the industry remained small and attracted limited interest up until the early 1990s (Joubert *et al.*, 2011). During the 90's a surge in research by the Agricultural Research Council (ARC) and the South African National Biodiversity Institute (SANBI) combined with marketing and the establishment of key role players such as SAHTA (South African Honeybush Tea Association) helped to catalyze the growth of the industry (Bester, 2013; Polak and Snowball, 2017). Important events during this time included for example, the first exportation of honeybush tea and the development of guidelines which producers implemented to optimize quality (Joubert *et al.*, 2011). Demand for honeybush has steadily grown owing to product diversification and international health-conscious shifts in consumer purchases. An average of 390 tons is exported each year (2010-2016) worth R23 million and the livelihoods of many are currently supported by the industry including approximately 150 wild harvesters

(McGregor, 2017a). Therefore, much is at stake to ensure that the supply of honeybush is sustainable and to ensure the continuation of the industry.

The potential value of the industry has also meant that honeybush has been recognized as a vehicle for local economic development (LED), which is suggested as one of the foremost means to uplift economically marginalized communities (Polak and Snowball, 2017). However, much like the industry itself, LED based on natural resources is at risk of failure if it is unsustainable. Given the increase in demand and the fact that most of the raw material extraction and production is situated in rural contexts such as the Langkloof, there are concerns over issues of illegal, incorrect or over-harvesting (Schutte, 1997; Potts, 2017).

Indeed, it has been acknowledged amongst industry members that poaching of wild honeybush is a serious problem which threatens the sustainability of the industry (HCOP, 2020). In light of both legal and illegal pressure on wild populations, efforts towards cultivation may help to meet market demand and thus reduce the need to source from the wild. What needs to be considered is the feasibility of cultivation within the immediate socioeconomic and landscape context. Commercial cultivation occurs far more widely in the Western Province whilst wild harvesting is more prevalent in the Eastern Cape. Approximately 131 of the 147 ha under cultivation as of 2016 were in the Western Cape (McGregor, 2017a).

Cultivation has both benefits and pitfalls that are evident-there is a fair amount of knowledge about commercial cultivation in the form of observations and research (Joubert *et al.*, 2011), although further insight is needed in terms of potential problems such as genetic bottlenecking. Other major limitations include the choice of reseeding species for cultivation which produce rapid large yields but are shorter lived, input costs for production infrastructure and the potential impact on taste and quality of the tea. For example, Joubert *et al.*, (2011) reported that cultivated crops of *C. subternata* fetch a lower market price compared to *C. intermedia* due to the latter tea being known for its exceptional quality. Changes in tea quality and properties may also negatively affect market demand as international standards are high and consumers may be put off by lower grade product (as reported at the HCOP, March 2020). This would, in turn, negatively affect everyone involved in the supply chain, especially those who are highly reliant on the industry for a living such as local families who live on the farms. Despite the challenges, cultivation has relieved pressure on wild populations, creates jobs and promotes the use and rehabilitation of old agricultural fields (Potts, 2017). In contrast to cultivation, there is far less information available about the wild honeybush resource, including its exact quantity and the long-term potential impacts of harvesting (McGregor, 2017a). Wild harvesting is currently regulated through NEMBA (National Environmental Management: Biodiversity Act) and provincial ordinances. Under NEMBA a bioprospecting permit must be issued by the national Minister of Environmental Affairs only after the applicable material transfer and benefits sharing agreements are approved (McGregor, 2017a). In the interest of this research only the ordinance details relevant to the Eastern Cape will be mentioned.

Cyclopia intermedia falls under a protected flora status in the Eastern Cape which requires permits for sellers, who are the landowners of the property on which wild honeybush grows, and growers who are landowners cultivating honeybush on their properties (De Villiers and McGregor, 2017). In terms of further regulation, a biodiversity management plan (BMP) is being developed for C. subternata and C. intermedia with the aim of setting up systems for monitoring and managing commercial extraction to ensure the long-term survival of the species whilst respecting the livelihoods of stakeholders (HCOP, March 2020). The BMP has many stated objectives and activities, and there are a few of particular importance that may be supported by this research project. The first relates to managing wild honeybush in such a manner as to avoid degrading biodiversity and ecosystem function (HCOP, March 2020). This can be assisted by understanding what ecological components are associated with wild honeybush occurrence especially if there are companion species that may be threatened by harvesting activity. The second is to inform management practices via a protocol for ecologically appropriate cultivation (and augmentation) which includes avoiding genetic mixing and sourcing seed from the appropriate populations (HCOP, March 2020). Because augmentation involves expanding populations, understanding wild seed viability rates and survival of seeds and seedlings sourced from parent populations may provide valuable insight.

Contributing to the support of these legal frameworks are guidelines for sustainable harvesting practices which are being incorporated into management of wild honeybush. These stipulate harvest intervals and methods based on expert knowledge and the ecology of the plant in order to reconcile industry needs and sustainable utilization (McGregor, 2017b). An essential component in the wild harvesting system is the harvesters themselves. Traditionally, males from families who live on the farm or come from nearby communities are the ones who work as harvesters and will work as a team (McGregor, 2017a). The nature of the terrain and work makes it extremely physically demanding and the amount of honeybush harvested may vary greatly amongst members of a team. Wages may vary from R1.50 per kg to R3.50 per kg (2017 rates) depending on the difficulty of access with some experienced harvesters earning between R1200-R1500 a week (McGregor, 2017a). Dependence on this source of income is very high for many of these harvesters as there are often no other employment opportunities available. Secondly, wages are used to support other family members and get cycled back into the local economy through purchasing of goods and services from informal traders (Machethe, 2004). These considerations add to the need for wild augmentation to be explored as a means to sustain the practice and possibly even expand harvesting opportunities, leading to more employment in the industry.

### 2.3. Wild Augmentation

Augmentation is a term that falls under the broader field of conservation ecology and has been defined as "the addition of individuals of a species within the geographic boundaries of an existing local population or metapopulation" (McMurray and Roe 2017: 2). Ideally, this is done from the same genetic pool (George *et al.*, 2009; Laikre *et al.*, 2010). The goal of augmentation is ultimately to increase or restore the desired resource whilst minimizing ecological change or damage (George *et al.*, 2009). Augmentation can be necessary for a number of reasons. Two of the main ones are direct over-exploitation leading to a collapse of wild stocks and habitat loss (Bottin *et al.*, 2007; Godefroid *et al.*, 2011). Large scale breeding programs are often required to successfully restore and maintain populations. Standard techniques for vegetation augmentation include artificially cultivating and reintroducing those individuals or relocating and supplementing (propping up) existing populations using propagules from parent material (Joubert *et al.,* 2009; Godefroid *et al.,* 2011).

Augmentation has been experimented with the world over, for example wild populations of *Arenaria grandiflora* (Mountain sandwort) a rare species which grows in similar conditions to fynbos has been successfully reintroduced, and population numbers reinforced through supplementing with seed in mountainous regions of France (Bottin *et al.*, 2007). Although augmentation is a valuable conservation tool, success rates reported by Godefroid *et al.*, (2011) specifically related to plant reintroductions are generally quite low. Importantly success is defined in the context of the paper as plants not only surviving but also reproducing and contributing to population growth. The study investigated 114 projects and found that survival rates were approximately 53%, whilst flowering (19%) and fruiting (16%) rates of those surviving were generally low (Godefroid *et al.*, 2011).

Some of the main success and failure factors identified were location of project, site preparation, understanding of the biological requirements and monitoring (Godefroid *et al.,* 2011). The same study found a link between projects occurring inside protected areas, and within the correct habitat parameters being more successful (Godefroid *et al.,* 2011).

In view of these points, the primary considerations for this study are choosing sites based on an understanding of the plant's ecology and site preparation. There is a large research gap with regard to planting honeybush directly into the wild. A variety of studies have focused on seed germination and growth characteristics for honeybush species under cultivated conditions but little is understood about germination and seedling survival rates in the wild. The only study to date (Beyers, 2016) used field trials to attempt to establish seed germination and survival rates on fynbos, old fields and ploughed fields for *C. intermedia* and *C. subternanta*. Additionally, seedling transplant survival rates were also tested, but only for the species *C. subternata*. Two treatments were applied, one where vegetation was cleared at ground level and removed, leaving the roots intact and another where no clearing was done. This was applied to the fynbos and old field plots, whereas the ploughed plot was already considered cleared. The study obtained mixed results and therefor it is difficult to ascribe possible explanations. Of particular interest were the germination percentages for *C.*  *intermedia*, which was relatively low across all treatments but the highest percentage (16%) was obtained in uncleared fynbos followed by the old fields (15%) whilst cleared fynbos had the lowest (1%). Although cleared fynbos had the lowest number of germinated seeds, this may have been due to predation. In terms of seed survival rate, the findings from lowest to highest survival were as follows: fynbos cleared, old field uncleared, fynbos uncleared, old field cleared, and ploughed. The same findings were produced for growth rates as well as *C. subternata* seedling survival, with the ploughed land and old field cleared showing significantly better results. The study provided a number of explanations for these results, for example the ploughed field and old lands had much higher phosphorus content than the fynbos soils which may have had a counter effect on the succession rates and overall survival of other fynbos genera (Hawkins *et al.,* 2008) as they are adapted to growing in nutrient deficient soils, but may have been beneficial for honeybush growth (Power *et al.,* 2010).

## 2.4. Wild Augmentation Risks and Benefits

There is currently no published research concerning the impact of augmentation on wild honeybush populations. However, wild populations of other animals and plants have been subject to changes such as genetic alteration through augmentation projects for decades around the world (Laikre *et al.*, 2010; Godefroid *et al.*, 2011). The point must be made here that the purpose of augmentation in the context of this research is to boost production in an ecologically sound way and thus differs from purely conservation orientated augmentation which is aimed at restoring endangered populations. Nonetheless the approaches and precautions learned from other cases of augmentation are valuable and provide the basis for considerations during this study.

The possibility of either positive or negative changes to the genetic integrity of a species is difficult to determine. This is primarily because effects can occur in two forms which interact and fluctuate between generations. The first type are those which change the genetic structure among populations of a species, for example by reducing or increasing the natural rate of genetic differentiation (Laikre *et al.*, 2010). The second type are those which influence the composition of genes of individuals within a population, this can occur for example in the

form of altered or replaced alleles. These changes ultimately have cascading consequences, potentially leading to an overall loss of adaptive capacity or reproductive fitness (Laikre *et al.*, 2010).

A documented case is that of the depression of natural reproductivity of Atlantic salmon (*Salmo Salar*) due to large populations of captive bred salmon being released into the wild over a short period of time (McGinnity *et al.*, 2009). Similar concerns are raised by Potts (2017) in the context of honeybush farming and its potential impacts on genetic diversity. As fynbos species exhibit clear evolutionary divergence at small spatial scales, populations will differ in their response and adaptations to local conditions (Potts, 2017). This divergence and adaptation will translate into a strong association between the geography and those populations' genetic lineages, resulting in phylogeographic structuring. (Potts, 2017). Although not all species exhibit this, there are factors which make it likely that *Cyclopia* are subject to phylogeographic structuring, such as seed dispersal proximity patterns from ants which carry *Cyclopia* seed underground constraining and localizing populations (Potts, 2017).

Because of the possibility of advantageous local adaptations, it is vital to preserve and avoid contamination of these natural genetic lineages. To ensure this preservation, any seed used for augmentation in an area must be sourced from the same wild parent populations. Since adaptive traits are conferred via alleles, by using genetic material which contains only the original parent information one can assure that local traits are not substituted for ones which did not originate locally (Potts, 2017).

Currently there is limited understanding of *Cyclopia* genetics, although research into the extent of genetic phylogeographic structuring of the genus has recently been done. Results from Galuszynski and Potts, (2020) show that across the natural range of *C. intermedia* 23 distinct haplotypes were identified suggesting genetic uniqueness. Although four of these are shared, the occurrence of this is non-uniform and instead seems to be related to historical climate shifts, topographic complexity and connectivity between populations, with 62.8% of variation caused by mountain ranges (Galuszynski and Potts, 2020). In the western extent, no shared haplotypes were identified, despite very close proximity of populations from the Anysberg, and Klein Swartberg (Besemfontein and Swartberg pass) whilst towards the east,

seemingly distinct populations shared certain haplotypes across the Langkloof, Groot Swartberg, Cockscomb and Baviaans mountain ranges (Galuszynski and Potts, 2020). In light of this, the study recommends treating individual populations as genetically distinct units for management since a significant level of haplotype richness and uniqueness has been observed (Galuszynski and Potts, 2020).

Another aspect of augmentation which is important to consider are the techniques used in conjunction with planting and sowing in order to increase the chances of success. Augmentation research from the Cape wildflower industry may hold insights for honeybush augmentation projects since the fynbos context is much the same. There are similarities between the mechanisms used by honeybush and other fynbos species in response to disturbances such as ploughing or burning (Schutte *et al.*, 1995). These techniques are often used to prepare the veld for receiving seed or seedlings for the purpose of increasing the number of individuals of a species that would otherwise naturally be present (Treurnicht, 2010). A study done by Joubert *et al.*, (2009) compared the impact of different ploughing methods on fynbos vegetation diversity and community for the purpose of augmenting populations of commercial wildflower species.

Ploughing is generally conducted with a toothed implement which can be adjusted for depth. In this study shallow ploughing was done at ~ 7 cm and is considered to be a low disturbance activity as it only loosens topsoil, leaving most underground root structures intact (Joubert *et al.*, 2009). Deep ploughing in contrast, is not restricted to the topsoil layer because the soil is loosened to a depth of >10 cm and is thus regarded as a high disturbance activity as many root structures can be sufficiently damaged so as to kill the plants (Joubert *et al.*, 2009).

Two virgin fynbos sites were ploughed using the deep and shallow plough method, thereafter they were seeded with commercial wildflower species such as *P. compacta and L. platyspermum*. The study also compared ploughed augmented (seeded) sites post fire with sites that burnt but were not augmented. Results showed that augmented sites which were subject to shallow ploughing had the highest species richness, possibly due to shallow ploughing being a low disturbance activity. With regards to resprouting species, they were far less abundant in the deep ploughed sites and reseeders had the highest cover percentage in the deep ploughed burnt sites. Overall, it was concluded that minimal disturbance augmentation techniques can be used to increase commercially important species without jeopardizing fynbos conservation. Management practices such as repeated burning and deep ploughing should be avoided due to the higher impact it has on natural ecosystem dynamics such as competitive exclusion (Joubert *et al.*, 2009).

A separate study conducted by Treurnicht (2010) examined the impact of these commonly used techniques (ploughing and burning) on species richness, abundance and structure over a longer period of time (8-20 years post disturbance). In contrast, Treurnicht (2010) found that growth form abundance which refers to the structural category such as shrubs or climbers and composition as well as overall species richness was significantly reduced in some treatment sites when compared to the control. This corresponds with a large body of research demonstrating that species composition changes over time after a disturbance (Connell and Slatyer, 1977; Kruger and Bigalke, 1984; Privett *et al.*, 2001). Although a number of interconnected variables influence plant community dynamics it is known for example, that large overstorey serotinous species (such as *P. compacta* and *L. platyspermum*) play an important role in stabilizing community establishment (Heydenrych, 1999; Privett *et al.*, 2001). But if these large overstorey species which are commercially valuable are sown in high numbers it can lead to an overtopping effect created by an unnaturally dense overstorey, impacting understory diversity and function (Vlok and Yeaton, 2000).

The findings from Treurnicht (2010) also highlight the need to acknowledge the complexity within the fynbos biome particularly with regards to its response to disturbances over longer temporal scales. Despite the unknowns, management practices should approximate natural disturbance regimes as best it can, for example it is known that fynbos types have different average fire intervals as well as fire seasonality (Heelemann *et al.*, 2008), therefore controlled burning should be done in those respective seasons. In the context of honeybush augmentation, it is difficult to extrapolate how both preparation techniques (burning or ploughing) and method of production (sowing or planting seedlings) may impact ecosystem functionality over time.

With reference to the preceding discussion, several points are pertinent to this study and honeybush augmentation in general. Firstly, it is vital to source seed from the same genetic pool as the population/ area that is being subjected to sowing or planting; secondly certain veld preparation techniques may simultaneously increase production but alter community composition more drastically than others and thus a balance needs to be found. Lastly, depending on the specific objectives of the project, augmentation planting/ sowing densities should approximate those of the natural population densities so as to avoid a potential overtopping effect as those seen within the wildflower industry with certain large overstorey Proteaceae. With specific reference to C. intermedia: because it is a slow growing resprouter and tends to occur in less dense stands with lower recruitment rates (Schutte et al., 1995) it will not have the same impact as fast growing dominant overstorey species such as those mentioned for commercial wildflower use. Given the need to balance commerce and conservation, research is needed with regards to the impact wild honeybush augmentation has on the environment with consideration of its economic potential. Long term postaugmentation monitoring of the associated vegetation community is needed to gain an indepth understanding of the impact of such a practice.

## 2.5. Wild *C. intermedia* Mapping and Optimal Site Selection

Determining wild resource distribution and habitat characterization are vital steps in managing and conserving ecological resources effectively. Such information often serves as the basis for creating biodiversity management plans and helps inform resource use practices (Matthews and Whittaker2015). Wild resource distribution can be mapped in a number of ways, such as using expert mapping, species distribution modeling (SDM) or a combination of methods (Randin *et al.*, 2006). Because living resources have defined geographical locations, it is possible to use variables which are known to determine or correlate with occurrence, to predictively map or model where the resource would occur (Loiselle *et al.*, 2003).

Predictor variables are for example, climatic conditions, biome type and elevation (Randin *et al.,* 2006) and species or communities which exhibit stronger spatial restrictions to specialized habitat conditions such as those shown by fynbos, can be more confidently modelled as their habitat range is less variable (Midgley *et al.,* 2003). In conjunction with this, well documented habitat types and locality records mean that models can be cross validated and checked for increased accuracy. In the case of wild honeybush populations, there has been limited research into precise biophysical attributes associated with each preferred habitat, hence the need to collect this data during the study (chapter 3). However, distributions and general environmental conditions such as preference for mesic sites, slope, aspect and elevation, are known for individual species (Schutte, 1997; Joubert *et al.,* 2011; Beyers, 2016).

General habitat data in combination with extensive locality records and expert knowledge have been used to generate SDM and distribution maps of *C. intermedia*. Barnardo (2013) used a combination of four types of models and known locality records to determine *C. intermedia* distribution under current and future climate scenarios using statistical approaches in R (version 3.3.3). Similarly, McGregor (unpublished PhD data 2020) used GIS methods and a mixture of data sources including expert mapping to model *C. intermedia* across its known habitat range. The resulting distribution patterns showed a tight restriction to the fynbos biome within the Cape Fold mountains, correlating with other research and local knowledge (Manning and Goldblatt, 2012; Joubert *et al.*, 2011; Schutte, 1997).

Similar techniques can be used at finer spatial scales to locate areas where ideal environmental characteristics overlap to choose optimal sites for planting. This process is known as multi-criteria decision making and is commonly used in agricultural development in the form of crop-land suitability analysis (Samanta *et al.*, 2011). The process uses multiple sources of input data to locate areas where all desired criteria are met. Large amounts of highly specified input data can be used to increase accuracy of the selection. For example, Samanta *et al.*, (2011) used high resolution physical and chemical soil classifications coupled with climatic data layers to develop a land suitability rating for rice crop production in Papua New Guinea. In the context of identifying optimal sites for honeybush augmentation, this will prove valuable because sourcing seed or growing seedlings is a time consuming and expensive process, therefor choosing sites which will maximize survival is important.

#### **2.6.** Habitat Characterization; Landscape Features and Fynbos Soils

At a landscape scale the influence of topography, elevation and geological types on plant diversity and distribution is well documented and evident in the fynbos biome (Rebelo *et al.,* 2006). Although clear community boundaries are observed, the exact reasons for this are complex as many interconnected factors affect structure and composition. Fynbos montane regions which are topographically complex give rise to a mosaic of environmental conditions and subsequent habitats which drive endemism and diversity (McDonald *et al.,* 1995; Esler *et al.,* 2014).

The Kouga mountains where farm 1 is located are no different. The geological makeup of this area is comprised mainly of acidic lithosol soils derived from sandstones of the Table Mountain Group as well as quartzitic sandstones of the Witteberg Group (Rebelo et al., 2006). These large-scale characteristics are important because they influence local soil composition. For example, Campbell (1983) found that Table Mountain groups contained on average lower total nitrogen content than Witteberg groups, though within each group there were significant differences found depending on the location of sampling. Richards et al., (1997) demonstrated in Soetanys fynbos that vegetation communities are correlated with changes in soil profile, though, similar to Campbell (1983) a large degree of variation was observed within sites. Between the five communities, pH, Nitrogen (N), Phosphorus (P), Calcium (Ca), Magnesium (Mg), Potassium (K) and Carbon (C) were measured. Important findings included the strong gradient of decreasing pH, C and N contents of soil going downslope from one community to another and the significance of the difference in nutrient contents between community boundaries (Richards et al., 1997). Nutrient content changes between communities found near the bottom of the slope on colluvial sand were significantly less than communities on the slope with limestone and sandstone. There were also drastic seasonal fluctuations particularly for P, and N, with some sites peaking during Autumn and others during Summer (Richards et al., 1997). All sites showed the lowest available P during the October sampling thereafter overall levels increased with the highest amount measured in December for some sites (Richards et al., 1997). For nitrates there was a less distinctive pattern but overall levels were lower in summer months with a clear peak in June and July for sites situated on the slope with limestone or sandstone as an underlying substrate.

A number of causes were attributed to the results. Overall high N contents on the limestone substate were attributed to high pH which does not inhibit nitrification compared to low pH of non-limestone soils (Richards *et al.*, 1997). Fluctuating levels of nutrients from the limestone samples indicates a level of sensitivity to temperature and moisture changes driven by seasons, whilst other fluctuations were probably due to microclimate differences caused by rocks and vegetation (Richards *et al.*, 1997). Other research has also shown that nutrient gradients and fluctuations are explained by steepness and aspect of slopes, depth of soils, erosion and runoff rates (Kutiel, 1992; Bergh *et al.*, 2014,). These factors subsequently further impact the soil profile, changing properties such as permanent wilting point which refers to the point at which the water content in the soil is so low that its molecular force with which it binds to the soil exceeds that of the plants ability to absorb water through its roots (Brady, and Weil, 2008).

As fire is a feature of fynbos systems, the impact of this on soil profile must be taken into account. It is known that fire both changes soil properties and mobilizes nutrients which in turn increase or decrease their availability in the ecosystem (Caon *et al.*, 2014). The impact of a fire on soil nutrients is mainly determined by the nature of the fire itself (intensity, duration and recurrence) and but also by factors such as topography, vegetation cover and soil type (Caon *et al.*, 2014).

Nutrient dynamics are complex and currently little is understood about the combined effect of cycling mechanisms such as fire or aerosol depositions (Bergh and Compton, 2015). Annual pulses in nutrients within fynbos biomes are significantly influenced by larger scale factors such proximity to the coast, prevailing winds and fie regime. An analysis of nutrient sources and fluctuations in mountainous fynbos post-fire (Swartboshkloof) in the Western Cape showed that marine aerosols were the main source of Cl, Na and SO<sub>4</sub> during summer, and macro- nutrients such as nitrogen and phosphorus in fynbos fluctuate post fire, possibly impacting initial plant regrowth and diversity (Bergh and Compton, 2015). Nutrient abundance shows a return to pre-fire levels quickly in streams and more steadily in soil, approximately 8 months after, although more realistically 1-3 years would be required for it return to the same levels completely (Bergh and Compton, 2015). Other research on Mediterranean ecosystems has suggested that the macro nutrients N, P, K increase in some instances post fire but and in others decrease, depending on a number of factors including vegetation type and fire intensity. On lowland sand fynbos it was shown that mean pH of soil increased in the first month post-fire but returned to pre-fire levels after 7 months, similarly organic matter % increased by around 2-2.5% post fire but returned after a few months, followed by a seasonal dip (Winter) eight months post fire (Brown and Mitchell, 1986). Fire also influenced total phosphorus and resin-extractable phosphorus which refers to the bio-available amount for plants to use. Total phosphorus 12 months post-fire was shown to be significantly different, however immediate post fire levels were not. Depth also had a significant influence over total P. Available P (resin-extracted phosphorus) levels were significantly higher both immediately and 12-month post fire and differences were more pronounced between summer months than winter (Brown and Mitchell 1986). Furthermore, there was a depression in total phosphorus from June-August between all depth ranges.

Soil composition associated with wild *C. intermedia* has been documented though more studies are needed. Barnardo (2013) examined physical soil characteristics including soil textural profile and field capacity which relates to water content and availability to plants once saturation has occurred (Assouline and Or, 2014). When comparing sites where *C. intermedia* was found to sites where it was absent, soil moisture levels were significantly higher where it was present, confirming previous knowledge of preference for cooler wetter slopes (Schutte, 1997; Joubert *et al.*, 2011).

In the same study, soil composition results showed that at all three sites, sand comprised the majority of the soil (above 80%), then silt (10-15%) and lastly clay (3-5%), confirming that *C. intermedia* grows on predominantly sandy to loamy well drained soils. No studies have specifically looked at the nutrient concentrations for sites where honeybush is present compared to adjacent sites where it is absent. This project aimed to supplement this knowledge gap through conducting detailed nutrient and physical soil analyses at presence and absence sites on different post-burn age veld.

## 2.7. Habitat Characterization; Vegetation Community

Fynbos vegetation has been observed to grow in diverse yet distinct communities with unique compositional and functional traits (Cowling and Gxaba, 1990; Privett *et al.*, 2001). This diversity has led to more than 119 different fynbos types being identified throughout the biome itself (Rebelo *et al.*, 2006), yet fynbos has increasingly come under threat from both commercial agriculture and urban expansion (Allsop *et al.*, 2014.). In the competing worlds of commerce and conservation it is unrealistic to expect landowners to sacrifice their livelihoods for the sake of complete ecosystem protection (Camboni and Napier, 1993). Instead, options for maintaining a level of protection and originality need to be encouraged. This is particularly relevant in the case of wild augmentation as the practice itself introduces changes within a natural system as opposed to completely altering that system by establishing monocrop plantations.

An important step in managing ecosystems and their resources is understanding what the biological components are that make up the system (Magurran *et al.*, 2010). In this instance an inventory of the species associated with wild *C. intermedia* will enable us to monitor levels of change to floristic patterns caused by augmentation. Collecting this data will require vegetation surveys of the communities found with honeybush and as a comparison those found adjacent to these sites that are without honeybush. Determining why some communities contain honeybush is complex as many studies have shown that an assortment of both external and internal factors govern species arrangement and occurrence in fynbos (Cowling and Gxaba, 1990; Rebelo *et al.*, 2006).

Internal factors include competitive interactions and life history traits (Cowling and Gxaba, 1990; Vlok and Yeaton, 2000) whilst external factors are disturbance regimes and abiotic resources (Kruger and Bigalke, 1984; Richards *et al.*, 1997). These factors interact with one another to create an ecologically dynamic system which changes over time (Kruger and Bigalke, 1984; Privett *et al.*, 2001). Therefore, the main focus of this research section is to determine species richness and dominance of plant communities associated with honeybush, rather than describe the reasons behind the makeup of these communities. The purpose of this approach is primarily to discover whether or not there are distinct types of communities that can be used to identify and typify honeybush habitats. The study also aims to identify

indicator species which are species that can be used to reliably predict the presence or absence of another species or type of habitat (Bal *et al.*, 2018). In conjunction with biodiversity management, this information will help identify areas which meet the ecological requirements for *C. intermedia* increasing the chances for successful augmentation. Although there is a diversity of literature about fynbos vegetation communities, studies specifically related to honeybush associated communities are lacking.

Barnardo (2013) conducted a phytosociological study for three locations where C. intermedia was absent and present and found that at least one of the sites (Swartberg) the species composition where C. intermedia was found was significantly different to where it was absent. However, at the other two sites (Kouga and Barrydale) species variation between absence and presence sites were not great enough to classify them into distinct communities. Commonly occurring species found with C. intermedia across the three sites were Leucospermum cuneiforme, Elegia filacea, Leucadendron salignum, Protea neriifolia, Protea repens, Rhodocoma fruticosa, Grubbia sp., Erica passerinae, Helichrysum teretifolium, and Seriphium plumosum (Barnardo, 2013). Despite the general similarities in vegetation community between both absence and presence sites, overall species diversity was always higher, and dominance indices were lower, indicating a more varied community where honeybush occurred (Barnardo, 2013). In light of these findings, and the need to gather more information in order to manage wild honeybush effectively, this study sampled vegetation communities where wild honeybush is both absent and present at farm 1 in the Kouga. There are different approaches to vegetation surveys and selecting an appropriate sampling method can be challenging in species rich sites such as those found within fynbos. This study used an approach commonly applied for fynbos vegetation research (Van Wilgen, 1981; Privett et al., 2001) involving the use of sampling quadrats as portions of the community to represent overall community composition.

#### 2.8. Cost Benefit Analysis

Modern agricultural sciences, like other economic fields makes use of financial evaluative tools to assess the feasibility of projects. This evaluation is an essential step in forecasting the outcome of investments and structuring business decisions to increase success (Hanley *et al.,* 2009). Amoung the various techniques used, is a cost benefit analysis (CBA), described as a systematic technique that determines options which provide the best approach in terms of benefits in labour, time and costing (David *et al.,* 2013). Cost benefit analyses are extremely flexible, have a diversity of applications and are used at varying scales within agricultural economics. For example, they have been used to assess and quantify benefits from erosion preventative conservation practices (Zhou *et al.,* 2009) at a watershed level to water re-use schemes at a national level (Haruvy, 1997).

Costs and benefits can also be captured and summarized in enterprise budgets which, similarly, are used to make decisions and compare production practices (Tranel, 2018). In this context, the cost and revenue of dedicating land to wild honeybush production would be compared to alternative land uses such as grazing cattle or sheep. This requires that information be collected through interviews about each activity in the production process of different land uses which are specific to that farm. Given that wild honeybush augmentation is still in its infancy and that the overarching goal of augmentation is currently more focused on conservation than as a means of propelling the development of the industry, a full enterprise budget and comparison of competing land uses was not undertaken. It is also worth considering that unique costs and benefits are involved in wild honeybush augmentation and there is currently no baseline information. Therefore, doing a comparison against other land uses which have well defined inputs and outputs may be misleading.

The process of conducting a CBA involves 8 practical phases (Mullins, 2014), summarily stated they are; (1) specifying the purposes and scope of the project, (2) identifying, defining and categorizing the various costs/impacts and benefits, (3) quantifying the costs and benefits, (4) recording and dealing with impacts that are difficult to quantify, (5) discounting project costs and benefits into present values, (6) calculation of net present value, (7) sensitivity analysis and (8) interpreting and reporting results. Regarding the first step of the CBA (specifying the purposes and scope of the project), there are usually two categories of valuation, private and social, which refer to the parties whom are accruing the respective cost or benefit (Hanley et al., 2009), this study will focus only on the private (landowners) valuation. Stated more specifically, the main goal is to determine the private economic value of an augmentation project specific to a farm setting. This tailors the analysis in terms of certain assumptions that need to be made. The assumptions made during this study are briefly provided below and will be discussed further in chapter 4. In terms of phase 2 and 3, costs and benefits were identified and quantified from actual augmentation projects, field data, market prices and current labour rates. Factors such as grow time in the nursery will drastically influence the cost of production due to labour and water, however longer times in the nursery will lead to stronger seedlings which may result in better survival. A grow time of 3 months was chosen for this study, but nursery periods of up to 8 months may be recommended. An expanded study should be done to test the impact of such changes. Due the variability in costs such as fuel and labour which will depend on distance to the population of plants, the study excluded the cost of fuel and assumed a constant labour rate. Similarly for the benefits, the study opted to simplify the analysis by assuming the average yield of the plants and price paid for the raw material remained constant. A raw material price of R15 per kg for C. intermedia in 2021 was decided upon by consulting industry members (G.K. McGregor, pers. comm. 2021).

Phase 4 of the CBA involves identifying and quantifying complex impacts such as societal benefits from biodiversity or livelihood changes from industry expansion. As stated above the purpose of this analysis was limited to the private benefits only and therefore including broader intangible aspects will complicate the analysis as it may involve ascribing monetary values to cultural or other non-physical entities (Beria *et. al.,* 2012).

In terms of phase 5, when conducting a multi-year analysis two main adjustments in the form of inflation and discounting need to be considered. Inflation which is the general rise in prices over time will affect the value of the inputs and outputs over the lifespan of the project. To deal with this change the value of the costs and benefits in year zero/start of the project (which was 2021) can be inflated by the inflation rates as managed by the South African Reserve Bank (3-6%) over the period of the project (DEAT, 2004). This approach may be risky as prices can be affected by unpredictable events such market recessions. For example, the price paid per kg for raw honeybush increased steadily from R8 in 2014 to R20 in 2020 (G.K. McGregor and E. Smith, SAHTA workshop, Feb 20<sup>th</sup>, 2020). However, in 2021 the price decreased considerably to R15. Another way of dealing with inflation is to use the "real" price as chosen in year zero and for all the other years (DEAT, 2004), as mentioned this project opted for the latter method and chose to use the raw honeybush price paid for in 2021 for the entire lifespan of the project.

The second adjustment which needs to be made is applying a discounting rate which brings the future value of costs and benefits into present terms (Mullins, 2014). This is to ensure that future benefits are not overly estimated and accounts for the riskiness of the project. It is also to ensure that projects which have longer lifespans are not favoured over shorter ones. This project opted to use the official 8% rate as reported (Mullins, 2014), though a lower rate of 5% which is more conservative will also be used.

Phase 6 of the CBA is calculating the Net Present Value of the project. This involves discounting costs and benefits to the present year using a discounting rate and time factor. The NPV was calculated as the summation from the beginning of the project (time =0) to the end of the project (t =n). The standard NPV equation used was used:  $[(B_t - C_t) / (1 + r)^t]$  whereby B = Benefits; C = Costs; r = discount rate; t = time period. In this research a time frame of 4 harvest cycles or 16 years was used and thus the equation would be;  $[(B_0 - C_0)] + [(B_1 - C_1) / (1 + r)] + .... [(B_{16} - C_{16}) / (1 + r)^{16}].$ 

Sensitivity analysis in step 7 usually involves calculating how the adjustment of input parameters will affect the investment. In this study limited parameters will be used, focusing mainly on lower and higher discounting rates and survival rates of seeds and seedlings. For example, the study will run a scenario where 25% survival is compared with 35% to assess to what degree this change affects the outcome. The final step in a CBA is interpreting the results. Since the outcome of the analysis is expressed as a ratio of the net benefits to costs, any ratio above 1 is viewed as a positive investment.

## 2.9. Cyclopia Seed Ecology and Germination

Seed dormancy is a common evolutionary adaptation that has been documented in a large number of plant families (Baskin and Baskin, 2004) including the Fabaceae to which the various honeybush species belong. The trait itself is complex, and multiple classes of dormancy have also been identified. The reasons for dormancy are determined by interacting environmental and physiological factors, and generally the trait is exhibited as a survival mechanism when conditions for post germination survival are least conducive (Baskin and Baskin, 2004).

Most shrubby fynbos species have resilient seeds which are evolved to survive long periods of dry conditions through strategies such as serotiny for example, exhibited in many proteacea species or myrmecochory, (carried underground by ants) (Le Maitre and Midgley 1992; Kruger and Bigalke, 1984). It is known that *Cyclopia* display a form of physical or combinational dormancy (Koen *et al.*, 2017), which means that once fully matured seed is dispersed it needs to undergo a process of scarification in order to germinate, even if the combined environmental conditions are ideal for seed germination (Baskin and Baskin, 2004).

Studies related to growth and germination characteristics for *Cyclopia* have primarily focused on increasing the commercial potential for species that are suited to cultivation. Far less is known about the natural levels of germination for species such as *C. intermedia* and what factors may affect this. Current literature indicates that a variety of factors influence germination, including seed age, seed colour, type of species (reseeder or resprouter) and treatment (Motsa *et al.*, 2017). What complicates seed studies further are natural levels of variability caused by a host of other influences such as genetics, climate and region (Motsa *et al.*, 2017).

Wild germination levels compared to those of clonal cultivars for *C. subternata* and *C. genistoides* varied considerably when tested. Motsa *et al.*, (2017) showed that wild *C. subternata* had a germination percentage of approximately 53% whilst clonal seeds were approximately 65%. The following year however the difference was much less, (only 1%) and the wild seeds had the higher germination rate. For the resprouter (*C. genistoides*) the wild

germination rate was approximately 67% whilst the clonal cultivar was less at 52%. The scarification method used was soaking the seeds in 98% sulfuric acid for 60 mins.

Koen *et al.*, (2017) tested the germination characteristics for dimorphic (green and brown) seeds paired with different treatments. Dry heat scarification results (*C. subternata*) indicate that green and brown seed have germination success rates at different temperatures, brown germinated most successfully at 2 min/100 °C (70.6%) whilst green was most successful at 4 min/100 °C (74.6%). Both green and brown seed geminated more successfully following wet heat treatment. These results support the findings of Mbangcolo (2013) who similarly showed that short treatment periods of 30 seconds in 100 °C water increased germination rates significantly in young seed (1 year old) but longer exposure times became detrimental.

Age was also shown to exert a significant influence, the seeds were neither scarified nor stratified so as to avoid introducing variables. Brown *C. subternata* seeds germinated significantly better with age: 3-year seed germination was 8.4% compared to 1 year which was 1.7%, the increases for green seeds of different ages were insignificant (Koen *et al.*, 2017). Overall green seed showed greater overall germination rates when treated but brown seed was more successful untreated. Koen *et al.*, (2017) concluded that green seeds were generally observed to be less permeable than brown seed and thus required higher scarification exposures to achieve optimum results. Brown seed mortality rates following treatment, were also higher which indicated that brown seed is more vulnerable to damage which is most likely due to micro-cracks within the seed coat increasing permeability and exposure.

From these studies it is evident that seed colour and age play a role in germination success and this has implications for augmentation projects which aim to collect and use seed from the wild. In view of this, the effect of seed colour and collection technique on germination rates were tested in the study by collecting seeds from 4 farms spread across the Langkloof and Kouga areas.

#### 2.10. Population Characteristics

Globally, wild plant populations have been altered through human induced pressures and many species have either gone extinct or are declining (McCune and Vellend, 2013). One such pressure which is the focus of this research is wild harvesting. "The impacts of harvesting can be understood in terms of changes in the ability of the species to replace itself and maintain a given level of stock" (Levels, 1998). What is known about the sensitive ecology of *C. intermedia* implies that it may be at a heightened risk of such impacts. Considering that other species of plants have been affected through selective harvesting in the form of reduced seedling recruitment (Coltman, 2008) and altered growth rates (Tickten, 2004), it seems necessary to monitor and characterize honeybush populations in order to avoid long term negative trends. Population characteristics can refer to a number of different measurements. For this research, demographic ratios and density, recruitment rates and growth features (allometric measurements) were focused on. Allometric measurements refer to quantifying anatomical features of an organism and their proportions (Damuth, 2001). These measurements may be particularly insightful for determining average stem or canopy sizes for different aged plants.

In terms of assessing the demographic ratios for these populations, there are numerous methods for surveying and the approach taken depends largely on the intended outcome, nature of the species and environment. In Mediterranean type biomes which are reproductively fire dependent, collecting demographic data from various sites at different successional stages may yield more insightful results. This is due to a combination of factors such as community competition, reproductive maturity and recruitment rates associated with changing post-disturbance conditions and age (Roy and Sonie, 1992).

For example, germination and age structure of populations of the shrubs *C. monspeliensis* and *C. albidus* two obligate seeders found throughout the Mediterranean and North African coastal areas showed that recruitment occurred almost entirely within five years following fire, reproductive maturity begins within the first-year post fire, but peaks during year 4, and natural population size declines from years 12-15 post fire (Roy and Sonie, 1992).

In fynbos similar factors influence demographic structure, Privett *et al.*, (2001) examined the change in vegetation composition over 30 years post fire and found that species richness was highest immediately after fire and thereafter successional processes caused the disappearance of shorter-lived species, particularly non-spouting serotinous Proteacea such as *L. coniferum* and *L. laureolum*. Currently there is a lack of research about wild honeybush demographic structuring and little is known about the long-term changes within and between populations caused by harvesting or burning.

An early study done by Du Toit and Campbell (1999) investigated seedling recruitment and survival rates of two honeybush species (C. longifolia and C. pubescens) in natural veld where existing parent populations were found. Although the study did not alter natural seed bank numbers, different treatments were applied to the C. longifolia population as opposed to a natural burn which swept over the C. pubescens sample area which may have influenced germination rate and success. Results confirmed previous knowledge of seeding species having significantly higher recruitment rates than resprouting, despite both life strategies depending on seed banks for new generations. C. pubescens displayed an average recruitment ratio of 62 seedlings per adult after 5 months and 227 after 17 months (Du Toit and Campbell, 1999). In comparison the resprouter had a recruitment rate of 2.8 seedlings initially and after 17 months only increased to 3.8 per adult (Du Toit and Campbell, 1999). Some important factors to consider from these results are the post treatment conditions of either site, particularly the C. longifolia site which experienced heavy flooding and some erosion two months after, as well as being artificially burnt in September (February has been suggested as the ideal season). Compounding this- the state of the accompanying vegetation was older than the site where C. pubescens was found, meaning that fire intensity and duration may have resulted in unfavorable scarification (Du Toit and Campbell, 1999).

Nonetheless, recruitment rates were low across treatments for the resprouter and it is expected that the reseeder will have far higher recruitment as it apportions more resources into seed production and subsequent recruitment to ensure population persistence (Du Toit and Campbell, 1999). This research thesis will look at the demographic structure of 5 wild *C. intermedia* populations at the research site in the Kouga. Information about seedling (and young plant) to parent ratios may be an important motivator for augmenting wild populations

since natural recruitment rates may be lower than what is necessary to maintain or increase populations that are subject to harvest pressure.

## 2.11. Theoretical Framework: Agroecological systems

Farming in the modern era has seen major advancements with regards to improved efficacy and yield due to higher levels of control over agricultural processes, however this has often been at the expense of natural system health and functionality (Swinton *et al.,* 2007). As system components are altered, biodiversity and ecosystem services originally supported diminish or disappear (Swinton *et al.,* 2007). Therefor farming wild landscapes in such a manner so as to preserve their natural state as far as possible will simultaneously preserve their functionality whilst allowing for production (Gliessman *et al.,* 1998). Unfortunately, this is difficult as natural systems are complex and sensitive to human influence, (Cilliers *et al.,* 2013) hence the need for a conservation agriculture framework to understand to the best of our ability how to balance human and natural components.

Conservation agriculture is an umbrella term for a range of agricultural management methods that prioritizes both the conservation of ecosystem processes and subsequently biodiversity alongside production (Hobbs, 2007). From this, agroecological systems can be understood more specifically as an agricultural environment which intentionally recognizes and manages the impacts agricultural/anthropogenic processes have over ecological integrity, in addition it aims to maintain productivity and even try improve it though maintaining or restoring a natural state (Bockstaller *et al.*, 1997, Kremen *et al.*, 2012).

The rationale for this approach in systems where productivity is depended upon the integrity of natural processes is clear. In a fynbos farming context, wild honeybush populations are reliant on a functional ecosystem, and therefore to maintain this whilst increasing wild production is of utmost importance and benefits both people and nature. Successfully managing agroecological systems according to Van Velthuizen (2007) requires an understanding of the biophysical attributes which determine resource occurrence and abundance. This information is used to monitor the state of the resource and environment in order to ensure sustainability. This aspect will be addressed by collecting data to characterize the natural conditions associated with *C. intermedia* and may provide valuable information for future augmentation projects which seek to use data about indicator species or soil profile to choose sites for planting. Since this framework also places emphasis on recognizing the system as a whole, it ensures that we acknowledge that the process of wild augmentation involves introducing anthropogenic disturbances into a natural ecological system such as altering the genetic pool sizes. As this disturbance must be kept to a minimum it becomes clear that principles such as sourcing seed from local genetic populations and avoiding high disturbance activities such as ploughing or repeated burning should be adhered to.

#### 2.12. Conclusions

Wild honeybush populations are precious natural and social resources that are facing increased pressure from a growing industry. The sustainability of livelihoods and the industry itself is heavily reliant on these populations and therefore solutions for maintaining them are necessary. Augmentation has been suggested by members of the industry for years as a possible solution to increase wild production (Q. Nortjé, pers. comm. 2020). Until cultivation can satisfy demand enough to offset overharvesting, options such as these must be explored. This literature review has demonstrated that there is an abundance of information about fynbos ecology and augmentation in general. Important points from the literature include: understanding a species ecological requirement prior to augmentation improves success; fynbos ecological complexity requires a cautious approach and disturbance magnitude should be minimized; genetic integrity must be preserved; understanding natural demographic and seed germination characteristics will lead to better management and utilization. Gaps identified in the literature which need to be addressed are: economics of augmentation; detailed habitat information for C. intermedia; augmentation trials for C. intermedia; and seed germination studies. This study aims to contribute to filling these gaps to further our understanding of the potential to use augmentation and to ultimately add to the body of knowledge around the sustainable use of the wild honeybush resource.

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# **Chapter 3: Habitat Characterization**

#### 3.1. Aim

The aim of this chapter is to characterize the environmental conditions (of areas) where *C. intermedia* is present and absent. Understanding what distinguishes presence or absence areas may be important for choosing augmentation sites that maximize success. In order to this a GIS approach in combination with biophysical data collection was used. The first objective was to map broad-scale distribution of *C. intermedia* using a multiple criteria selection (MCS) process. In addition, optimal growing areas and study sites were identified on a local scale using a similar method. The second objective was to assess sites in terms of soil profile and plant diversity.

#### 3.2. Introduction

Currently the honeybush industry is facing uncertainties around the regulation and management of wild honeybush resources (HCOP, 2020). As the majority of the product is derived from these sources, specifically C. intermedia, it is critical that wild stocks are assessed and maintained. Managing wild resources equates to understanding the environment within which that resource occurs. Both biotic and abiotic components exert an influence over the state and occurrence of resources and therefore need to be documented (Van Velthuizen, 2007). This principle applies equally in the context of wild fynbos as ecological knowledge underpins responsible resource use (Allsop et al., 2014). Fynbos industries have demonstrated that monitoring associated ecological components and assessing impacts to guide management have resulted in increased environmental protection and industry sustainability. For example, after the expansion of the rooibos and wine industries a number of species were listed as threatened and there was a general reduction of diversity in areas transformed for production by the early 2000s (Raimondo et al., 2009; Allsop et al., 2014). In response to these issues, new resource use policies and organizations such as the Flower Valley Conservation Trust have been developed to include biodiversity principles in the management of these industries (Allsop et al., 2014). Alongside protecting the environment, an understanding of ecological conditions is valuable as wild production in fynbos may be highly dependent on maintaining ecosystem originality.

Because fynbos is highly diverse, competitive exclusion is thought to be less of a factor in determining species occurrence than resource availability. In addition, species complementarity is high (Richards *et al.*, 1997; Fridley, 2001).

This high level of complementarity means that many species derive benefits from each other such as shade and soil stability (Holmes and Richardson 1999). Much like the target species within the wildflower industry, honeybush grows alongside other species which share the same environmental preferences, some of which may be threatened or protected. Ideally, an augmentation management plan needs to consider this full range of species and acknowledge that wild production may benefit from maintaining areas with high species diversity. In addition to these preceding points, strong associations between plant distributions and soil properties have been observed for fynbos genera such as Proteaceae and Restionaceae (Richards *et al.*, 1997) and this kind of association seems to apply for *Cyclopia* spp. as they are distributed across a vast range of habitats with each species exhibiting clear preferences (Joubert *et al.*, 2011). The general habitat characteristics for *C. intermedia* are known but there is limited local scale data on wild populations in the Kouga mountains. This chapter deals with collecting information about soil characteristics and vegetation community diversity where *C. intermedia* is absent or present. At a broad scale, a GIS-based approach using MCS based on known preferences was used to map the distribution across the Eastern and Western Cape. This was compared to local scale expert mapping of populations to determine if 'modelling' of potential honeybush distribution correlated with actual populations and might be a suitable tool for optimal site selection.

## 3.3. Methods

# 3.3.1. Study site

Farm 1 is located in the Kouga mountain range and is situated near the farming settlement of Nooitgedacht approximately 30 km north of the town of Kareedouw in the Eastern Cape. The Kouga mountains fall within the fynbos biome of the Cape Floristic Region and are extremely species rich (Rebelo *et al.*, 2006). The geology and topography of the farm is characterized by resistant quarzitic rock of the Cape fold mountain belt and extreme montane heterogeneity exists in the form of parallel scarps and deep gorges which were created by ancient tectonic events of folding and subduction (Rebelo *et al.*, 2006). This landscape complexity has resulted in unique and varied vegetation distributions. In particular the eastern montane region of the CFR has large patches of vegetation types distinct from fynbos such as forest and thicket found in valley bottoms which can be seen on the farm.

The larger vegetation unit of the region is classified as Kouga sandstone fynbos and is recognized by its high grass component (Rebelo *et al.*, 2006, Esler *et al.*, 2014). The average annual rainfall for the area is approximately 350-450 mm, it rains throughout the year with peaks in winter and spring. Temperatures vary dramatically between seasons: the average

maximum temperature in summer (January) is approximately 29 degrees Celsius and in winter, (July) 13 degrees Celsius (Rebelo *et al.,* 2006). Wild populations of honeybush (*C. intermedia*) are scattered throughout the area and predictably occur on moist south east to south west facing slopes. Some populations are found in less typical sites on shallower soil near large rocky outcrops that appear to be much drier and less suitable.

The landowners (Nortjé's) have produced quality honeybush tea derived from wild *C. intermedia* for decades. Raw material is processed in-house at processing facility located in Nooitgedacht. The wild populations on the farm have been sustainably managed with local expert knowledge of harvest and fire recovery for three generations. Study sites indicated on figure 3-1 below on farm 1 were chosen through a multiple criteria selection (MCS) process

(Section 3.4.2.) Each site has two plots where habitat data was collected and augmentation trials were done.

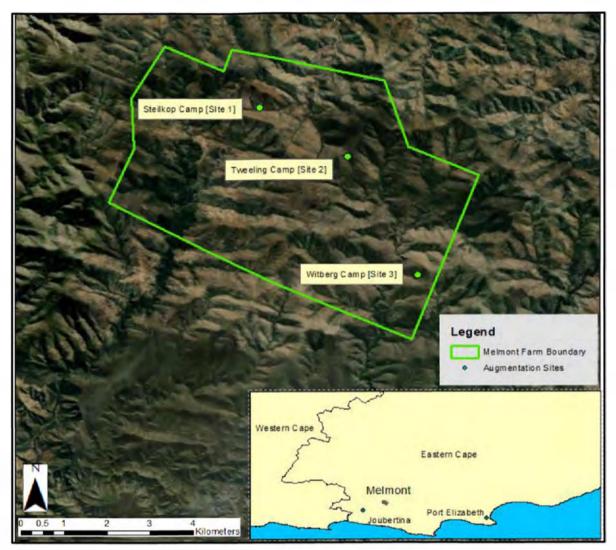


Figure 3-1: farm 1 selected study sites for augmentation and habitat data collection based on multiple criteria selection process of ideal habitat conditions

#### 3.3.2. Optimal Growth Areas and Study Site Selection

In support of the broader aims to characterize wild C. intermedia habitat and conduct field augmentation trials, the study used known ecological preferences (referred to as environmental parameters) to map optimal growth areas and choose sites that would be most representative of natural conditions on farm 1. In addition, the study also mapped a regional scale potential distribution using the mapping software ArcMap 10.2 using a similar MCS process with various data layers (appendix 1). Information concerning the environmental parameters were obtained from McGregor (unpublished PhD data, 2020). For the distribution mapping, an expert map polygon layer which defines a broad distribution boundary of C. intermedia (McGregor unpublished PhD data, 2020) was used as the outline within which all analysis occurred. This procedure involved first selecting out areas which were considered "untransformed" from the National landcover data. Thereafter all areas which met the following three criteria: elevation 500-1800m, >350mm mean annual rainfall, and fynbos vegetation (not renosterveld) were selected and made into a new layer using an "intersection" function. Following this, a slope and aspect model layer was generated, and the preferred habitat parameters selected out and made into separate layers (slope 5°- 35°, aspect NW, SE, S, SW, W). The final step was to create an output by intersecting all the preferred environmental parameters. The process is represented by the cartographic model 1 (figure 3-2). Areas where all layers defining the typical environmental characteristics of C. intermedia habitat intersected were considered the most likely to host wild populations (figure 3-5).

A similar MCS procedure was applied to determine optimal growing areas and study sites at farm 1 using the following environmental parameters: slope (5°- 35°), aspect (NW, SE, S, SW, W), and elevation (500-1800m). Once these environmental parameters had been intersected the output was a suitable conditions layer. Once this had been done, the suitable conditions layer was then intersected with known locations of patches of harvested wild honeybush which have been expertly mapped by the landowner (G.K. McGregor, *pers. comm.* 2020). The output of this intersection was named optimal growth areas. It was assumed that areas indicated on the expert map are areas which have the most favourable growing conditions since they already contain patches of wild honeybush. The process of identifying optimal

growth areas is shown in model 2 (figure 3-3) and the final output map is shown in figure 3-6. After optimal growth areas had been identified experimental sites were chosen within those areas. It was important to include fire as a factor when choosing sites, since *C*. *intermedia's* ecology is intimately linked to it. This involved determining the fire history of the farm over the past 5 years using MODIS fire imagery, represented by model 3 (figure 3-3).

Once the fire history had been mapped, areas where fires overlapped with optimal growth patches were identified by intersecting the output of model 2 and model 3 (figure 3-3). This allowed for the selection of sites which had different burn histories and which fell within optimal growth areas. Site 1 (Steilkop camp) burnt in November/December 2016 and site 3 (Witberg camp) in December 2019. For site 2 (Tweeling camp) a controlled burn within an area of optimal growth was conducted in June of 2020 and the previous natural fire was in September 2014. At each site, plot placement was done by choosing one area where wild honeybush was present and one where it was absent. The absence plots were located adjacent (between 50- 100 m) to the presence plots but they did not contain honeybush.

Honeybush landowners have commented on the patchiness of honeybush growth, suspecting that over small scales there may be factors in the soil or plant community that constrain honeybush (G.K. McGregor, *pers. comm.* 2020). The rationale for placing plots within close proximity is based on the assumption that general conditions of both plots are suitable for *C. intermedia* growth but that there may be specific differences with regards to soil or vegetation that distinguish the plots. Stated otherwise the study tested whether absence plots which appear to have the correct ecological conditions near wild populations do indeed differ significantly from the presence plots. Additionally, the absence plots were placed close to presence plots because the study aimed to promote augmenting areas in which wild *C. intermedia* would grow naturally for the sake of minimizing the impact on biodiversity and increasing the chances of success.

Topographic conditions were very similar for both plots at each site, limiting the impact of other topographic influences such as slope aspect and rise. Thus, the main plot treatments were time after fire (referred to as site age and measured in months) and absence or presence of wild *C. intermedia*. Considering part of objective 1 was to use GIS methods to identify

optimal growth areas, the effectiveness of this was assessed by looking at what percentage of the total ideal topographic conditions layer occurred within the expert map. The comparison was done under the assumption that if the process is able to identify areas which overlap with the expert map, then it is possible to use environmental parameters to identify potential augmentation sites which represent ideal conditions, and should give an indication as to how much land is ideally available/ suitable for augmentation. In reality this process requires ground truthing and further validation to decide if such sites are suitable, additionally there are other factors such as rock cover which are not included in the selection process which will influence results.

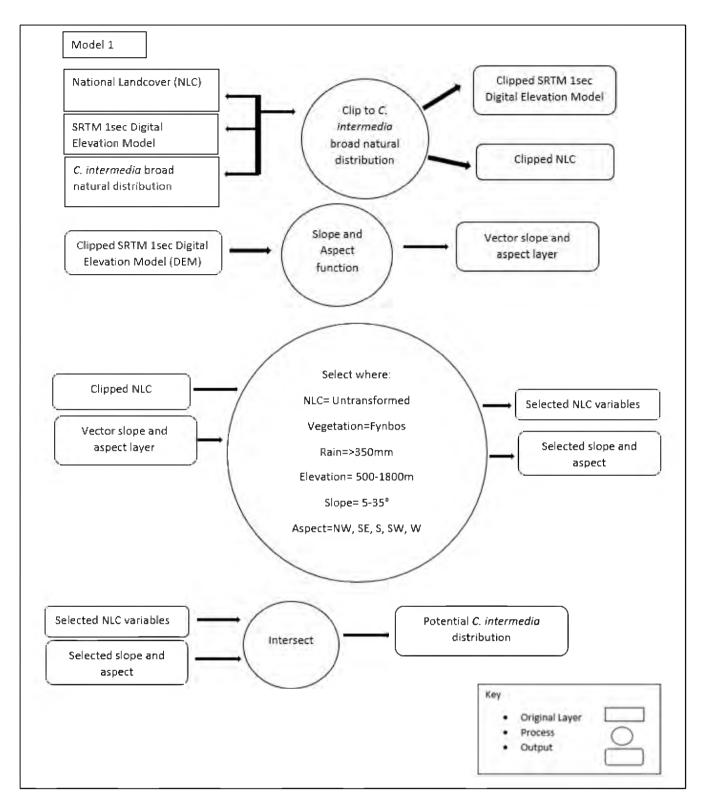


Figure 3-2: Cartographic model 1 showing the MCS process for predicted distribution. Areas of untransformed land that are found within the fynbos biome were intersected with environmental parameters (elevation 500-1800m, >350mm mean annual rainfall, slope 5°-35°, and aspect NW, SE, S, SW, W)

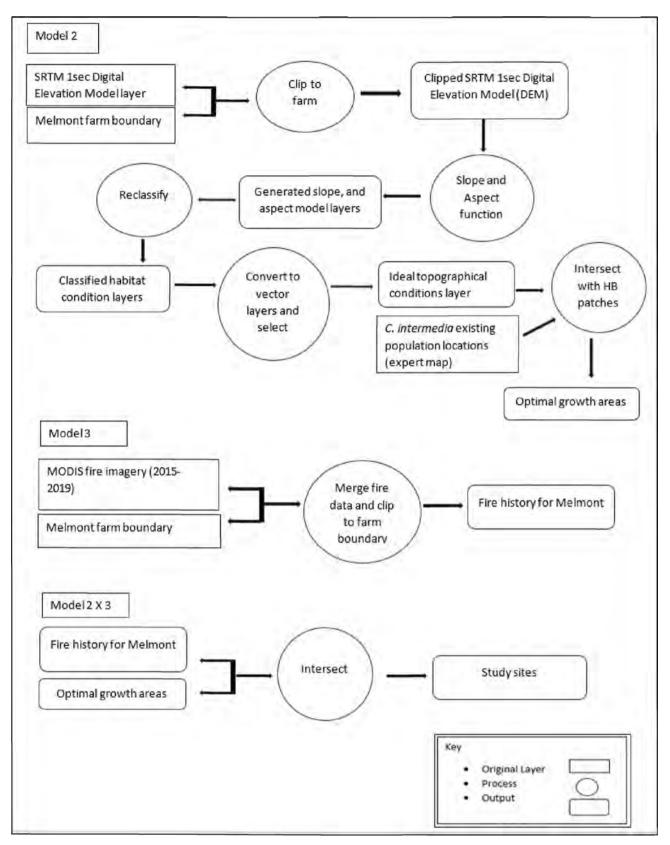


Figure 3-3: Cartographic models showing optimal growth areas identification process (model 2), areas which had burnt in the last 5 years; fire history (model 3) and the intersection of models 2 and 3 to choose sites for the study

#### **3.3.3. Vegetation Sampling**

Presence and absence plots at each of the three sites on the farm were sampled using a 50 m long transect with a sampling design of 3 quadrats placed along the beginning, mid-point and end of the transect (figure 3-4). Each quadrat was 4 m<sup>2</sup> and within each quadrat all plants were identified down to the lowest taxonomic rank possible with the help of taxonomist Dr. W. Vos (private consultant) and any unidentified specimens were pressed for later identification. Two heterogeneity indices Simpsons (D') and Shannon (H') were used to determine relative species richness and evenness (Stirling and Wilsey, 2001). The compliment form of Simpsons (D') calculated as (1-D') was used meaning that a higher value equals greater evenness or lower dominance of one species over the others. Even though sampling was done in Spring many species were not inflorescence making them especially challenging to identify. With the result that some specimens were only identified at the genus/ family level. A complete species list can be found in appendix (2). Sorenson's Coefficient (CC) was used to measure community similarity, whereby  $CC = \frac{2C}{S_1+S_2}$ . The total number of shared species between the two communities (C) is multiplied by 2 and divided by the total number of species found in each community. Similarity values range between 0 and 1, a value of 1 represents perfect community overlap. Total number of species associated with each plot was obtained by summing all the species found within each quadrat along the transect. It is important to note that vegetation sampling at site 2 was done outside of the controlled burn area, thus the age of the vegetation community was approximately 6 years. At site 1 and 3, the age of the vegetation was 4 years and 11 months respectively when sampled.

## 3.3.4. Soil Sampling and Landscape Features

Soil sampling was done by collecting 3 bulk soil samples (made from 5 sub samples) at both the absence and presence plots for each of the three sites at farm 1. Each sub sample was acquired by digging a hole with a small spade and consisted of bagging all the soil within the hole. The dimensions of each sampling hole were approximately 10 cm x 10 cm x 10 cm. The subsamples were spread out approximately 10-15 cm from each other. Sampling dates and frequency are shown in table 3-1. Soil samples were collected once in winter (June 2020) for sites 1 and 3. For site 2 samples were taken at the beginning of June 2020 before the controlled burn and then after the controlled burn later in June 2020. Thereafter a sample was taken five months later in November of 2020 (Spring). At each of these times a control sample located nearby the sampling plots was taken but was not exposed to the fire. The sampling layout is shown in figure 3-4. In table 3-4 the soil samples reported for site 2 are the first samples taken post fire (June 2020). This was done because pre-fire the veld age at site 2 was the same as that of site 1. Since the study aimed to test the different nutrient levels associated with different post fire age veld, the sample taken directly after the fire at site 2 was chosen as the appropriate sample to represent 0-month-old veld. All sites are located on slopes that were south to south west facing but elevation, aspect and other factors such as rockiness and depth will all influence soil properties (Ackerly *et al.*, 2002). Data loggers in the form of soil temperature and moisture probes were placed near each plot to determine what sort of changes are experienced between these sites. Soil analysis was done by BemLab and included mean phosphorus (P), Carbon (C), pH, trace metals and silt/sand/clay content. The

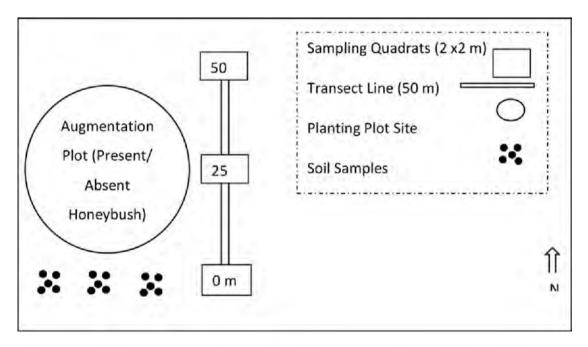


Figure 3-4: Vegetation and soil sampling design. Vegetation sampling transects were placed adjacent to augmentation plots and each quadrat was spaced evenly along the beginning, mid-point and end of the transect. Soil sampling clusters were spaced 2 meters from each other and consisted of 5 samples with dimensions 10 cm x 10 cm x 10 cm

study also recorded general landscape features of site elevation, aspect, slope, rockiness and GPS location.

Table 3-1: Sampling dates and frequency. The number of months post fire is counted from the time of the previous fire for each site. Vegetation sampling was done on veld which was last burnt in September 2014 for site 2 and not in the controlled burn area

	Site 1: Steilkop	Site 2: Tweeling	Site 3: Witberg		
	(Burnt December 2016)	(Natural burn September 2014) (Controlled burn June 2020)	(Burnt December 2019)		
Soil Sampling	1 x June 2020 (43 months)	1 x June 2020 (pre-fire) 1 x June 2020 (0 months) 1 x November 2020 (5 months)	1 x June 2020 (7 months)		
Vegetation Sampling	1 x November 2020 (47 months)	1 x November 2020 (74 months)	1 x November 2020 (11 months)		

# **3.3.5.** Statistical Analysis

Data were recorded and managed using Microsoft Excel and analysed using the statistical programming language R version 4.1.0 (R Core Team, 2016). Soil property data were tested for normality using Shapiro-Wilk tests, and all significance calculations were based on a 95% confidence limit. Two-way ANOVAs were used to test if factors (A) 'site age' (43 months, 0 months, 7 months) and (B) 'honeybush' (presence or absence of honeybush) influenced soil properties, P-values significance level is p< 0.05. Site 1 represents a 43-month post-fire site, site 2 represents 0-month post-fire and site 3 the 7-month post-fire site. Post hoc multiple comparisons were done using Tukeys Honest Significant Difference (HSD) with P-values significant at p< 0.05. Samples with shared superscripts are not significantly different and degrees of freedom (*df*) are given below each factor. Labeling convention for the camps and plots at each site was as follows; Site 1: Steilkop present (1 P), absent (1 A), Site 2: Tweeling present (2 P) absent (2 A), Site 3: Witberg present (3 P) absent (3 A).

#### 3.4. Results

#### 3.4.1. Species Map, Optimal Growth Areas and Study Sites

The map of *C. intermedia* distribution (figure 3-5) was produced from a multiple criteria selection of suitable habitat conditions. The species range spans approximately 500 km from east to west and 100 km north to south. The areas covered by the intersection of suitable conditions (indicated green on the map) cover approximately 4900 km<sup>2</sup>. The plant is continuously distributed in both large and small isolated patches along the Cape fold mountain belt. This pattern is well recognized with this species from previous research (Kies, 1951; Schutte, 1997; Joubert *et al.*, 2011; McGregor, 2017).

Optimal growth areas identified on farm 1 in the Kouga mountains are shown in (figure 3-6). The total amount of farm area shown on the map is approximately 3700 ha in size and of that 660 ha is honeybush bearing land according to the expert map. The MCS analysis used to identify optimal growing areas shows that 1067 ha of the total farm area of 3700 ha is suitable land for augmentation. Of the 1067 ha approximately 40% (425 ha) intersected with the expert map, indicating that a fair proportion of the areas identified by the model correlated with the expert map. The majority of the rest of the land identified which fell outside of the expertly mapped patches is likely to be unsuitable for augmentation. This is due mainly to limited input variables, which don't account for finer scale landscape variability such as rock cover, soil type and vegetation community composition, thereby overestimating the actual amount of land that is suitable for augmentation.

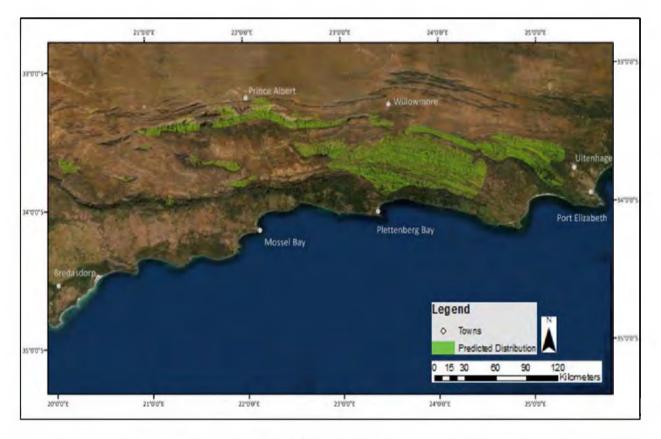


Figure 3-5: Model 1 output: Potential distribution of wild C. intermedia based on the overlap of known suitable environmental parameters. The total potential distribution covers an area (indicated green on the map) of approximately 4900 km<sup>2</sup>. It extends along the Cape fold mountains east of Uitenage to the Witteberg in the Western Cape.

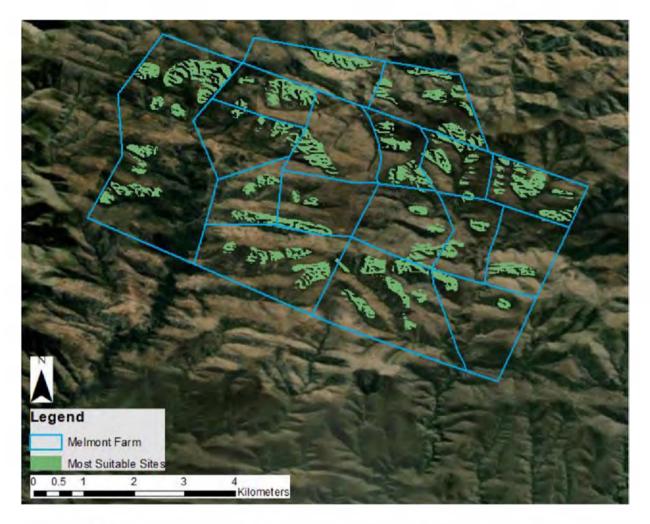


Figure 3-6: Model 2 output; areas shown in green meet the optimal growing criteria and were identified by intersecting the environmental parameters (elevation 500-1800m, slope 5°- 35°, and aspect NW, SE, S, SW, W), and expert map patches

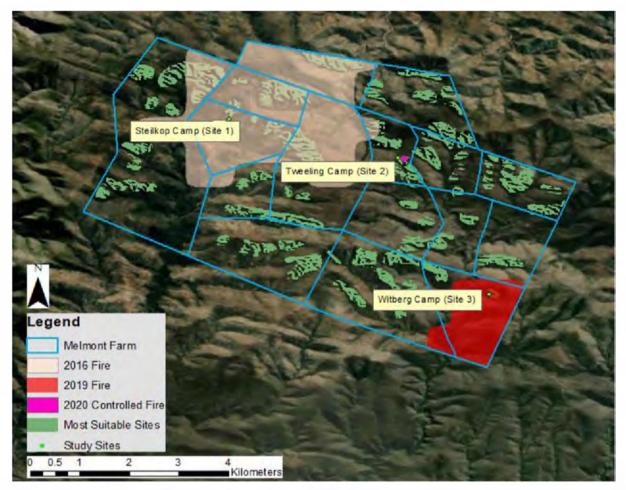


Figure 3-7: Study sites chosen within areas identified by the intersection of the environmental parameters, expert map and recent fires. Sites 1 and 3 were placed within veld burnt by natural fires whilst a controlled burn was conducted within an area identified by the intersection of environmental parameters for site 2

## 3.4.2. Species Composition

In total 83 species were identified from all the sites on farm 1, indicating diverse floral communities. Of those identified, 11 belong to the Proteaceae and Ericaceae families and are protected, no threatened or endangered species were identified from the survey in terms of the SANBI red list of species. Only *C. intermedia* was identified as having a declining status. Overall, the average heterogeneity indices (H') and ( $\lambda$ ) were slightly higher at plots where honeybush was present (2.62) (0.89) compared to where it was absent (2.45), (0.87). This indicates that species abundance and richness was elevated in presence sites. The proportion of shared species varied notably. At site 1 community similarity was lowest with a coefficient value of 0.21, followed by site 2 (0.42) and site 3 had the most similar community between the absent and present plots (0.55). All sites were dominated by a mixture of montane shrub and grass. It was unexpected that site 2 which had the oldest vegetation, was less species rich and shared a higher percentage of the same species despite being located on a more sheltered, wetter slope than site 1 and 3. For site 1 the dominant species shared by both plots were Ficinia gracilis, Diherteropogon filfolias and Brunia noduliflora, for site 2 it was only Erica demissa and Cliffortia linearifolia. At site 3 there was also three shared dominant species, namely, Themeda triandra, Tristachya lucethrix and Erigrostis superba. At site 2 the vegetation was dominated by taller and more dense shrubs, whilst site 1 and site 3 had much shorter vegetation that was spread more sparsely.

Table 3-2: Vegetation community comparison for sites at farm 1. The average community diversity index H' was higher at the presence plots with 2.62 compared to 2.45 for absence plots. Average dominance (λ) was very similar between present and absent plots with values of 0.89 and 0.87. Sites 1 and 3 were dominated by the grass species *Diherteropogon filfolias, Eragrostis superba, Themeda triandra* and *Tristachya lucethrix*. Site 2 was dominated more by the shrub species *Cliffortia linearifolia* and *Erica demissa* 

Site ID	Total no.	Shared	СС	Species	Dominance	Dominant Species
	of Species	Species		Richness H'	λ	
1 (P)	27	5	0.21	2.68	0.89	Ficinia gracilis, Restio triticeus, Diherteropogon filfolias, Brunia noduliflora
2 (P)	20	8	0.42	2.56	0.89	Diheteropogon filifolias, Wildenowia glomerata, Cliffortia linearifolia, Erica demissa
3 (P)	24	13	0.55	2.61	0.88	Themeda triandra, Eragrostis superba, Rhodocoma fruiticosa, Tristachya lucethrix
Mean	24			2.62	0.89	
1 (A)	20	5	0.21	2.31	0.84	Ficinia gracilis, Brunia noduliflora, Agathosma mundtii, Diheteropogon filifolius
2 (A)	18	8	0.42	2.4	0.88	Babartia orientalis, Erica demissa, Cliffortia linearifolia, Leucadendron salignum
3 (A)	23	13	0.55	2.63	0.91	Themeda triandra, Pentaschistis pallida, Tristachya lucethrix, Erigrostis superba
Mean	20			2.45	0.87	

# 3.4.3. Landscape Features

Table 3-3: General plot characteristics for the sites at farm 1. Site 1 was situated on a gentle slope with low rock cover and shallow ground compared with site 2 which had a steep incline and deep soil with low rockiness. Site 3 had a mixture of gentle and moderate slopes with very shallow soil and high rock cover

Site name	Elevation (m)	Aspect	Slope	Co-ordinates	Description
				33° 42′ 03″ S,	Rock cover 15-18%
Site 1 P	969	s	Gentle 5-8°	24° 08′ 51″ E	Fairly shallow soil
				33° 42′ 01″ S,	Rock cover 10-13%
Site 1 A	974	S	Gentle 5-8°	24° 08′ 49″ E	Fairly shallow soil
				33° 42′ 39″ S,	Rock cover 2-5%
Site 2 P	978	S	Steep 25-30°	24° 10′ 21″ E	Deep soil
				33° 42′ 39″ S,	Rock cover 5-8%
Site 2 A	969	sw	Steep 25-30°	24° 10′ 23″ E	Deep soil
				33° 44′ 10″ S,	Rock cover 35-40%
Site 3 P	918	sw	Gentle 5-8°	24° 11′ 33″ E	Shallow soil
				33° 44′ 10″ S,	Rock Cover 30-35%
Site 3 A	907	sw	Moderate 10-15°	24° 11′ 31″ E	Shallow soil

## 3.4.4. Rainfall and Soil Characteristics

Rainfall data is shown in figure (3-8). Peak rainfall occurred in August (48 mm), October (78 mm) and December (42 mm) of 2020. The average summer ( $24 \pm 12.9$  mm) and winter rainfall (17 ± 4.3 mm) was similar to previous data ( $22 \pm 12.9$  mm), ( $19 \pm 15.5$  mm) for the same farm (Barnardo, 2013). Autumn rainfall was however lower ( $24 \pm 4.4$  mm) when compared to the same data ( $34 \pm 6.5$  mm) (Barnardo, 2013). Overall, a total of 362 mm of rain occurred over the study period, approximately 90 mm less than what had been previously recorded (Barnardo, 2013). Soil temperature and moisture for site 3 is shown in figure 3-9 and figure 3-10. Data for sites 1 and 2 was unable to be retrieved. The soil temperature for the present plot at site 3 was marginally higher than for the absent plot for most of the year. Soil temperature fluctuated according to a seasonal pattern for both plots and peaked during the summer months of January (present- 22.5 degrees Celsius), (absent- 21.5 degrees Celsius) and February 2021 (present- 21.3 degrees Celsius), (absent- 20.8 degrees Celsius) and dipped lowest during winter in August 2020 (present- 11 degrees Celsius), (absent- 10.2 degrees Celsius) of 2021.

Soil moisture was more varied between the two plots, with the present plot having higher moisture readings from June (2020) until February (2021). Soil moisture was almost identical for March (2021), thereafter the absent plot had higher soil moisture readings until the end of the study (July 2021). Co-occurrence of soil moisture peaks with rainfall is most evident during October (17% present), (15.8% absent) and December (17% present) (15.2% absent). Lowest soil moisture was measured during the months of June 2021 (4% present) and July 2020 (6% absent).

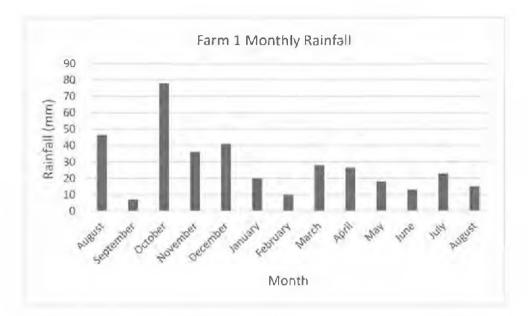


Figure 3-8: Farm 1 monthly rainfall over the study period from August 2020 until August 2021. Peak rainfall occurred in October 2021 with 78 mm and lowest rainfall occurred in previous month of September 2020 with 7 mm

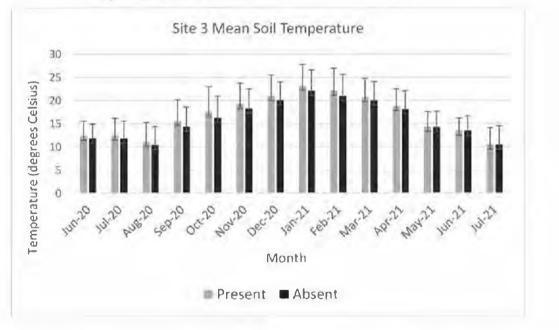


Figure 3-9: Site 3 mean soil temperature for present and absent plots over the period from June 2020 until July 2021. The highest average soil temperatures were measured during the summer months of January (present- 22.5 degrees Celsius) (absent- 21.5 degrees Celsius) and February (present-21.3 degrees Celsius), (absent- 20.8 degrees Celsius) and lowest in August 2020 (present-11 degrees Celsius), (absent- 10.2 degrees Celsius) and July 2021 (present-10.1 degrees Celsius) (absent- 10.2 degrees Celsius)

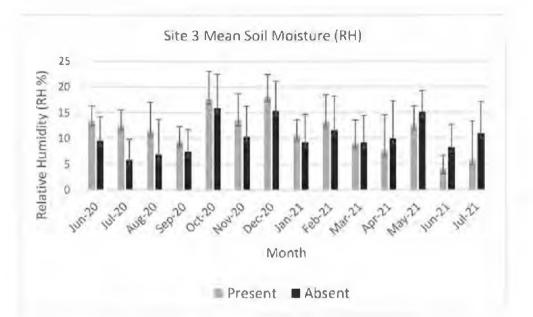


Figure 3-10: Site 3 mean soil moisture measured as relative humidity (RH). Soil moisture showed correlation with rainfall. Peak soil moisture occurred for both plots in October (17% present), (15.8% absent) and December (17% present) (15.2% absent). Lowest soil moisture was measured in June 2021 (4% present) and July 2020 (6% absent)

Table3-4 shows the soil analysis for each plot at the sites on farm 1. Average soil acidity (pH) was similar throughout with the exception of plots 2 (P) and 3 (P), which had a significant difference (p< 0.05). Nutrient levels of phosphorus (mg·kg<sup>-1</sup>) at the plot level were only significantly different between 2 (P) and (A) (p< 0.05). When comparing across sites, site 3 had significantly higher levels than site 1 (p< 0.05). The factors site age and absence or presence of honeybush as well as the interaction had significant p-values (p<0.05). Potassium levels (mg·kg<sup>-1</sup>) were significantly higher at site 2 (A) than site 3 (A), and site 1 (P) and site 1 (A) (p< 0.05). Both treatments had a significant effect over potassium (p< 0.05). Carbon (%) was very even between most sites, only 1 (A) and 2 (P) showed a significant difference (p< 0.05), in terms of treatment effects only site age had a significant influence (p< 0.05).

Magnesium base saturation (%) difference at the plot level was significantly different between plot 2 (P) and (A) (p< 0.05) and insignificantly different between site 1 (P) and (A)

(p> 0.05) the same insignificant result was shown for site 3. Magnesium overall was highest at site 1 and the factor site age was significant (p< 0.05). Potassium (%) was insignificantly different between plots 1 (P) and (A), the same result was shown for plots at site 3 (p>0.05). There was a significant difference between plots 2 (P) and (A) (p< 0.05), the factor honeybush proved to be significant (p< 0.05). Looking at calcium (%) the same result at the plot level as potassium (%) was shown, only plots 2 (P) and (A) being significantly different (p< 0.05). Site 3 had significantly higher levels of calcium than both sites 1 and 2 (p< 0.05) and the factor site age was highly significant (p< 0.005) whilst the presence or absence of honeybush was significant as well as the interaction between the two factors. (p < 0.05). Sodium (%) was significantly lower at site 3 than at sites 1 and 2 (p< 0.05), at the plot level only plots 2 (P) and (A) had a significant difference (p< 0.05). In terms of soil physical properties, clay content (%) was significantly higher at site 1 (A) than 1 (P) (p< 0.05) and sites 2 and 3 did not differ significantly (p> 0.05). Silt content (%) was significantly higher at plot 3 (P) compared to 3 (A), no significant difference was found at the plot or site level between sites 1 and 2 (p> 0.05). Sand (%) did not differ significantly at the plot level for all sites (p> 0.05) but site 2 and 3 did have significantly higher content than plot 1 (A).

	Site 1 (P)	Site 1 (A)	Site 2 (P)	Site 2 (A)	Site 3 (P)	Site 3 (A)	Interaction ( <i>df=2</i> )	Site Age ( <i>df =2)</i>	Honeybush ( <i>df =1)</i>
Acidity									
рН	$4.3 \pm 0^{ab}$	$4.5 \pm 0.1^{ab}$	3.9 ± 0.5ª	$4.4 \pm 0.3^{ab}$	4.8 ± 0.1 <sup>b</sup>	4.5 ± 0.2 <sup>ab</sup>	p >0.05	p <0.05*	p>0.05
Nutrients									
Phosphorus (mg∙kg <sup>-1</sup> )	3.2 ± 0.1 ª	2.1 ± 0.1ª	2.6 ± 0.1 ª	6.9 ±0.3°	4.4 ±0.4 <sup>b</sup>	5.4 ±0.5 <sup>b</sup>	p<0.05*	p<0.05*	p <0.05*
Potassium (mg·kg <sup>-1</sup> )	171 ± 3.8 <sup>ab</sup>	155 ±11ª	160 ± 1.5 <sup>ab</sup>	240 ± 10.8 <sup>c</sup>	219 ± 9 <sup>cb</sup>	187 ± 7.1 <sup>b</sup>	p <0.05*	p <0.05*	p <0.05*
Carbon (%)	3.7 ± 0.4 <sup>ab</sup>	3.4 ±0.4 <sup>a</sup>	4.4 ± 0.7 <sup>b</sup>	$4.1 \pm 0.1^{ab}$	4.2 $\pm 0.1^{ab}$	3.9 ± 0 <sup>ab</sup>	p>0.05	p <0.05*	p>0.05
Base saturation									
Magnesium (%)	24 ± 0.18ª	23.9 ±0.57 <sup>a</sup>	22.4 ± 1.9 <sup>ab</sup>	19 ± 0.36 <sup>b</sup>	19.7 ± 0.55 <sup>ab</sup>	19.1 ± 0.33 <sup>b</sup>	p >0.05	p <0.05*	p >0.05
Potassium (%)	7 ± 0.28 <sup>ab</sup>	7.7 ±0.7 <sup>b</sup>	5.7 ± 0.2ª	7.1 ± 0.17 <sup>b</sup>	7.7 ± 0.4 <sup>b</sup>	$6.7 \pm 0.1^{ab}$	p >0.05	p >0.05	p>0.05*
Calcium (%)	35.6 ± 1.8ª	37.4 ±1.6ª	20.5 ± 0.3 <sup>b</sup>	31 ± 2.6ª	51.3 ± 1.5°	44.8 ± 0.6 <sup>c</sup>	p <0.05**	p<0.05**	p <0.05*
Sodium (%)	3.3 ± 0.3 <sup>c</sup>	$3.4 \pm 0.14^{bc}$	3.6 ± 0.16 <sup>c</sup>	2.9 ± 0.15 <sup>b</sup>	1.8 ±0.2ª	1.8 ±0.15ª	p>0.05	p <0.05**	p>0.05
Particle Composition									
Clay (%)	15 ± 2 <sup>b</sup>	20 ± 1.2 <sup>c</sup>	13 ± 2 <sup>ab</sup>	11 ± 1.1ª	12 ± 1 <sup>ab</sup>	10 ± 1.15ª	p >0.05	p <0.05*	p <0.05*
Silt (%)	17 ± 2.3 <sup>ab</sup>	$16 \pm 6.1^{ab}$	15 ± 1.5 <sup>ab</sup>	15 ± 1.15ªb	18 ± 2 <sup>b</sup>	12 ± 2ª	p >0.05	p >0.05	p <0.05*
Sand (%)	$68 \pm 1.1^{ab}$	64 ± 5ª	72 ± 1.15 <sup>b</sup>	74 ± 1.15 <sup>b</sup>	$71 \pm 2^{bc}$	78 ± 3.1 <sup>c</sup>	p <0.05*	p <0.05*	p >0.05

Table 3-4: Selected soil properties for the six plots at farm 1, showing mean ± and standard deviation. P-value significance level is p<0.05\* and very significant at p<0.005\*\*. Values with shared superscript letters are insignificantly different

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Table 3-5 displays the effects of fire on soil properties for site 2 at farm 1. Acidity changed significantly between winter post fire and spring post fire samples for both the present and absent plots (p< 0.05). Treatment type honeybush was a significant factor (p< 0.05). Winter pre-fire pH was not significantly different to winter post fire in either absent or present plots (p> 0.05). Phosphorus (mg·kg<sup>-1</sup>) showed a steady rise from winter pre-fire levels to spring post fire for both plots; the absent plot (A) spring post fire sample was significantly higher than all other samples (p< 0.05) and winter post fire samples were significantly higher than the pre-fire winter samples for both absent and present plots (p< 0.05). Both treatment types and the interaction between them proved significant (p< 0.05). Potassium (mg·kg<sup>-1</sup>) showed a similar increasing trend after the fire, the average levels were significantly higher post fire for both plots when compared to pre fire levels (p< 0.05).

Looking at the results for carbon (%) there was an initial reduction from winter pre-fire levels to winter post-fire though the result was insignificant for both plots (p< 0.05). After winter post- fire there was a significant increase to spring post-fire (p< 0.05) for both plots. The treatment site age proved to have a significant influence (p< 0.05). Base saturation results were mixed with some cases displaying a decreasing trend and in others an increase with time after fire is observed. Sodium (%) decreased significantly from winter pre-fire to spring post fire in both plots (p< 0.05). Site age was a highly significant factor (p< 0.005). Potassium (%) had the opposite result and increased significantly from winter pre-fire to spring post-fire in both plots (p< 0.05), the factor site age proved to be highly significant (p< 0.005). Calcium (%) decreased between the winter pre-fire and post-fire samples for both plots, but only the absent plot showed a significant decrease (p< 0.05). After this initial decrease a significant increase from winter post-fire to spring post-fire is observed for both plots (p< 0.05), the absent plot had significantly more calcium in the spring post-fire sample than the present plot (p< 0.05). The treatment site age had a highly significant influence (p< 0.005) whilst honeybush was significant (p< 0.05). Magnesium (%) did not differ significantly between any of the samples.

 Table 3-5: Fire effects on soil properties for site 2 at farm 1 showing mean ± and standard deviation. P-values are significant at p<0.05\* and very significant at p<0.005\*\*. Values with shared superscript letters are insignificantly different</th>

	W. Pre-Fire (P)	W. Post-Fire (P)	S. Post-Fire (P)	W. Pre-Fire (A)	W. Post-fire (A)	S. Post-fire (A)	Interaction ( <i>df=5</i> )	Site Age ( <i>df=5</i> )	Honeybush ( <i>df=1</i> )
Acidity	w.rie-ine (i)	(')	(')	(^)	(^)	5. 1 03t-life (A)	(uj-3)	(uj=5)	(uj-1)
рН	4.13 ± 0.15 <sup>ab</sup>	3.86 ± 0.06ª	4.5 ± 0.26 <sup>b</sup>	4.3 ± 0.45 <sup>ab</sup>	4.4 ± 0.34 <sup>b</sup>	5.16 ± 0.15 <sup>c</sup>	p>0.05	p>0.05	p <0.05*
Nutrients Phosphorus				2 22 4 0 02 <sup>b</sup>				0 0 5 * *	0.05*
(mg·kg⁻¹)	$2.67 \pm 0.3^{ab}$	$2.2 \pm 0.1^{ab}$	8.5 ± 2.9 <sup>d</sup>	3.23 ± 0.93 <sup>b</sup>	6.9 ± 0.26 <sup>c</sup>	16.56 ± 5.3 <sup>e</sup>	p <0.05*	p <0.05**	p <0.05*
Potassium (mg·kg <sup>-1</sup> )	142.5 ± 56 <sup>b</sup>	160 ± 1.53 <sup>b</sup>	343 ± 17.35 <sup>d</sup>	99.7 ± 75.1ª	239.6 ± 10.7°	450.3 ± 40.9 <sup>e</sup>	p<0.05*	p <0.05**	p <0.05*
Carbon (%) Base Saturation	_5.9 ± 1.8 <sup>bc</sup>	4.39 ± 0.7 <sup>ab</sup>	6.18 ± 0.77 <sup>c</sup>	4.75 ± 0.87 <sup>ab</sup>	4.1 ± 0.07ª	7.27 ± 1.3 <sup>d</sup>	p>0.05	p <0.05*	p>0.05
Sodium (%)	4.16 ± 0.71 <sup>c</sup>	3.6 ± 0.15 <sup>cb</sup>	2.77 ± 0.64 <sup>ab</sup>	3.57 ± 0.75 <sup>b</sup>	2.91 ± 0.15 <sup>ab</sup>	2.07 ± 0.07ª	p>0.05	p <0.05**	p >0.05
Potassium (%)	4.15 ± 1.6 <sup>ab</sup>	5.7 ± 0.2 <sup>b</sup>	9.60 ± 0.74 <sup>d</sup>	3.28 ± 2.58ª	7.09 ± 0.17 <sup>c</sup>	8.89 ± 1.6 <sup>d</sup>	p>0.05	p <0.05**	p>0.05
Calcium (%)	23.08 ± 4.6 <sup>a</sup>	20.54 ± 0.3ª	48.03 ± 3.7 <sup>d</sup>	40.8 ± 16.1 <sup>c</sup>	31.37 ± 2.6 <sup>b</sup>	58.12 ± 1.81 <sup>e</sup>	p <0.05*	p <0.05**	p <0.05*
Magnesium (%)	20.3 ± 6.6ª	22.38 ± 1.9ª	18.9 ± 1.07ª	19.63 ± 2.6ª	19 ± 0.36ª	21.1 ± 1.5ª	p>0.05	p>0.05	p>0.05

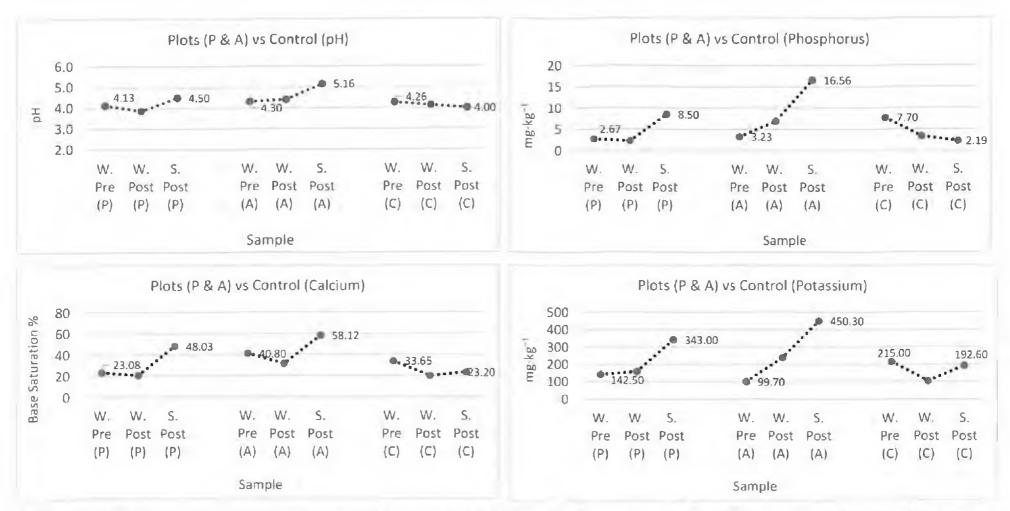


Figure 3-11: Selected soil properties showing some of the control vs plot (absent and present) results for site 2 on Farm 1. The abbreviations are as follows: P-Present, A-Absent and C-Control

#### 3.5. Discussion

Documenting aspects of the of the wild C. intermedia resource is critical as usage rises. In order to do this an understanding of the species ecology is vital. Similarly, using augmentation to boost wild production of C. intermedia requires that we know what ecological characteristics will optimize augmentation success. Broad physical descriptions are available in the literature regarding favoured honeybush habitats. On a local scale however, farmers often comment on the patchiness of C. intermedia growth as it is sometimes absent in areas which share the same conditions. By investigating soil and vegetation communities in absent and present sites this chapter aimed to improve our knowledge of what characterizes honeybush habitats. Additionally, an important aspect of the species biodiversity management plan is to ascertain resource distribution and inform sound cultivation practices which include augmentation. This chapter aimed to contribute towards this by using GIS methods to map regional distribution using a MCS process. In conjunction the method of using environmental parameters to locate ideal areas for augmentation was tested by comparing the outcome with known locations of honeybush. This method could be recommended as a way for landowners to identify areas which are suitable, thus contributing towards the goal of sound cultivation.

The distribution map showed an expected restriction to southerly facing slopes of the Cape Fold mountainous areas across the Eastern and Western provinces, concurring with previous research (Schutte, 1997; Joubert *et al.*, 2011). Intermittent patches of occurrence with large spaces between some populations is due to biome boundaries which change abruptly in response to climatic and geographic conditions. Areas closest to the coast where no *C. intermedia* is predicted are dominated by mixture of lowland vegetation types such as renosterveld and strandveld in the West and thicket and grassy fynbos to the East (Rebelo *et al.*, 2006). Towards the interior, areas which receive less rainfall transition into succulent Karoo and dry thicket vegetation types which are not suitable for *C. intermedia*.

One area where this is evident is the little Karoo basin which is situated between mountain ranges such as the Groot Swartberg and Outeniqua both of which contain wild populations (Rebelo *et al.,* 2006; Galuszynski and Potts, 2020). Whilst these results may be helpful in prioritizing areas or assessing to what extent the species might occur on private versus

protected land, more detailed ecological data associated with honeybush occurrence needs to be incorporated for better results. This is because predictive mapping can result in either an over or under estimation of the resource as variables which influence distribution are not included in the model and outputs require validation such as ground truthing (Ray and Burgman, 2006; Draugelis-Dale, 2008). The results therefore, are by no means conclusive. However, at a local scale, optimal growth area mapping using the same topographic input criteria and data produced satisfactory results as most of the area known to contain wild *C. intermedia* overlapped with the areas identified in the model. This indicates that MCS can potentially be used as a time and cost-effective method for choosing augmentation sites, especially if the process is refined by more input data.

In terms of the soil and vegetation data, it appears that there was no consistent difference between sites where *C. intermedia* is present compared to adjacent sites where it is absent. Rather, what was observed is that variations exist within and between sites. This result is unsurprising, since fynbos is known for its complexity and heterogeneity. The results are also similar to previous research. In comparing sites with and without *C. intermedia*, Barnardo (2013) showed significant differences for vegetation community at some sites and none at others at a regional scale. The scope of this research result is very limited since data collection was conducted on one farm in the Kouga mountains. Soil conditions or vegetation communities may be consistently similar or different in other areas. Secondly there are other biophysical variables such as soil micro-organism communities which are known to relate to community composition that could be unique to areas where *C. intermedia* is found (Slabbert *et al., 2010*).

Large scale data collection is necessary for a more comprehensive picture of honeybush habitats. One possible method for collecting data across vast areas is incentivizing resource users (Miller-Rushing *et al.*, 2012), in this context harvesters could take plant or soil samples when harvesting wild populations of honeybush. Knowing what characterizes honeybush habitats will allow for the detection of any changes to the ecological status of these systems. This is vital since fynbos management must be tailored according to each area and type. (Holmes and Richardson, 1999). Assessing rare or endangered species co-existence with *C. intermedia* could also prevent the industry from running into similar conservation concerns

that other wild fynbos crops have encountered. In the case of the rooibos industry, continued expansion of plantations on favourable soils has contributed to 168 plant species being "red-listed" in those areas according to SANBI (Esler *et al.*, 2014).

Populations of *C. intermedia* were found to occur in species rich areas on farm 1. Diversity and evenness were slightly higher in presence plots and community similarity changed unevenly between plots at each site. In site 1, community similarity between the plots was only half of that at site 2. The plots at site 3 shared the highest number of species, over half of the community was the same. This suggests that communities within which wild *C. intermedia* populations grow in the Kouga mountains can be both distinct and similar to adjacent areas where it is absent even though veld age and topography are broadly the same.

Grass species were common across all sites which is to be expected since Kouga sandstone fynbos which is the prevailing type found on the farm is known to have a high grass component (Rebelo *et al.*, 2006). Other common species such *Brunia noduliflora* which is seen as an indicator for *C. intermedia* (according to local knowledge) was found to be dominant for site 1 only. Slightly higher diversity levels associated with the presence plots are difficult to explain since both plots at each site were placed on the same slope and aspect and within close proximity effectively removing the influence of topography as a factor which is known to affect diversity (Cowling and Lombard, 2002). Additionally, the effect of fire on diversity at the plot level was ruled out by placing plots close together such that they were subjected to the same burn regime. Approximately every 10 years the veld containing wild honeybush patches is burnt to control vegetation height and encourage healthy *C. intermedia* growth (Q. Nortjé, *pers. comm.* 2020).

Fynbos communities have extreme alpha diversity and certain patches within that community are more diverse than others (Campbell and Van der Meulen, 1980). It may be that, sampling plots for the presence sites were placed within these areas of higher diversity. As for differences across sites in community diversity and composition, the age of the vegetation, fire history and actual location on the farm most likely had the biggest impact. Site 1 had postburn vegetation of 4 years and was situated near a valley bottom, site 2 had 6-year-old vegetation but was situated near a valley head on the side of a seep and site 3 had 11-monthold veld and was near the top of a mountain with no valley. Sites 1 and 3 receive less shade and are overall more harsh sites than site 2. The combined influence of these factors would have determined the plant community at these sites.

The results for soil properties between sites were also mixed with some trends being displayed. Much like the vegetation results, spatial variation of nutrient levels in soils is driven by a number of complex processes (Rice *et al.*, 1993; Caon *et al.*, 2014). Whilst this study aimed to assess whether site age (post-burn) and the presence or absence of honeybush was a significant factor influencing various soil properties, it did not investigate the particular causes for this and only general observations can be made. Low soil pH values overall were expected as fynbos soils are generally acidic (Esler *et al.*, 2014).

Looking at P, K, C content it was expected that site 1 which had deeper loamy soil would have higher nutrient contents compared to site 3 which had shallow rocky soil and was situated on a steeper gradient near the top of a hill. This is because topography influences local microclimates by changing soil temperature and moisture (Richards *et al.*, 1997). These changes in turn influence soil processes, more moist soils generally have better decomposition and nutrient cycling rates, which results in higher nutrient levels (Sierra *et al.*, 2015). What was observed however was that the oldest veld (site 1) had the lowest levels of these nutrients whilst site 3 had slightly higher levels. The increase of these nutrients was most likely due to the fact that site 3 was burnt approximately 7 months before sampling and the soil had not returned to the low nutrient pre-fire levels indicative of most fynbos soils (Bergh and Compton 2015).

The impact of fire on nutrients and pH was most clearly seen from site 2. Pre-fire soil profile from the control samples were similar to those measured at the other sites, and therefore any changes observed are at least in part linked to the impact of fire. The increase in soil pH from pre- to post burn is supported by other studies (Caon *et al.*, 2014). Generally, research has shown that there is a loss of macro-nutrients within the A level soil horizon (<20 cm subsurface) post fire due to volatilization and convectional transport caused by high temperatures but that low intensity fires are able to release nutrients bound up as plant matter without causing as much loss (Rice *et al.*, 1993; Caon *et al.*, 2014). Due to this fact,

sample depth must also be considered, samples for this study were taken within the A level range, shallow enough to be affected by the fire. The results for total potassium and phosphorus as (mg·kg<sup>-1</sup>) showed a significant increase from pre-to post fire indicating that the fire may have been of low intensity causing a release of nutrients in the form of ash which was collected as part of the sample.

Overall, there was an increase for nutrients whilst some minerals such as sodium and magnesium showed decreases. Another important observation was the magnitude of these nutrient changes between the two plots at site 2. The absent plot for an unknown reason showed on average greater differences between its samples. These differences could have been compounded by seasonal affects, as the control samples seem to show decreases and increases correlated to when the sample was taken. Whilst it is known that nutrient and mineral fluctuations occur in Mediterranean soils due to seasonal changes, the exact correlation between a particular season and a change in soil property has not been well established (Bergh and Compton, 2015). However nutrient levels such as phosphorus have been shown in some instances to decrease during Spring (Richards *et al.*, 1997) which was also shown by the control samples in this research. In general, for site 2 when compared to sites 1 and 3, there was a substantial pulse in macro-nutrients for the newly burnt site but this pulse only occurred a few months after burning and differed in magnitude between samples taken at close proximities for an unknown reason.

As a final point, it is interesting to compare the levels of nutrients associated with *C. intermedia* to those found with wild *C. subternata* which were analysed in roughly the same Kouga and Langkloof areas. For example, levels of K found at the farm 1 sites were double or triple the amount compared with the 54-57 (mg·kg<sup>-1</sup>) found for *C. subternata* (Riksen, 2019) and similarly Mg levels were also higher. The pH levels as well as P and Na appears to much higher where wild *C. subternata* grows (Riksen, 2019) whereas Ca is higher where *C. intermedia* is found. These results may be due to the type of underlying geology since habitat preferences between the two species are different, further studies are needed to establish more comprehensive soil profiles for all honeybush species and especially for those of commercial interest.

#### 3.6. Conclusions

From an agroecological perspective managing wild *C. intermedia* resources by increasing or sustaining populations through augmentation equates to managing the entire fynbos system in a way that aims to preserve its integrity. Since honeybush is an indigenous plant, it has evolved to occur within the same habitat conditions as many other species and is likely to benefit from some of these in ways which are not well understood yet. Maintaining biodiversity whilst undertaking augmentation will simultaneously ensure that the conservation conscious image of the industry is promoted and increase wild production. In order for this to be achieved the biophysical attributes of wild honeybush habitats need to be well understood so that potential change to ecosystem components can be monitored and so that production is maximized by choosing areas where the physiological needs of the plants are best met. The results in this chapter showed that at a farm scale *C. intermedia* occurs across a range of conditions and that occasionally the variation within these sites over small distances is greater than the difference between sites located further away. This kind of small-scale heterogeneity is inevitable in fynbos systems and discovering features which distinguish some areas from others is challenging.

Nonetheless some of the notable results with regards to vegetation was the higher species diversity associated with *C. intermedia* presence and a local indicator species only being found as a dominant for one site. The soil analysis similarly was variable, important macro-nutrients phosphorus and potassium (mg) did not differ significantly between plots at sites that were not burnt recently but for the site that was freshly burnt an increase was observed. Coupled with this increase was a decrease however in other elements such as sodium and magnesium post-fire for the freshly burn site. To what extent these fluctuations in soil properties influence *C. intermedia* survival and growth is unknown but the results from the augmentation experiment in chapter 4 seem to show that *C. intermedia* does benefit. In summary, *C. intermedia* by virtue of its tolerance for different conditions as reflected in the wide distribution and the nature of fynbos ecosystems being so heterogenous means that

finer scale habitat conditions will vary from place to place. Future studies need to be done to determine if there are possibly other characteristics such as soil organism communities that may be used to distinguish areas of occurrence.

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# Chapter 4: Augmentation Trials and Cost Benefit Analysis for Wild C. intermedia

## 4.1. Aim

The aim of this chapter is to evaluate the ecological and economic viability of augmenting wild populations of *C. intermedia*. This will be accomplished through two main objectives. The first objective is to conduct a cost benefit analysis of the augmentation process using different scenarios to simulate potential real-world outcomes. The second objective is to plant *C. intermedia* seeds and seedlings directly into plots where wild populations occur and also into adjacent plots where it is absent.

# 4.2. Introduction

Honeybush tea and related products derived from various *Cyclopia Spp.* comprise a relatively small but rapidly growing industry in South Africa (Joubert *et al.*, 2011). The potential for growth due to increased international health awareness and research into the unique biochemistry of the plant has meant that production is increasing but currently is unable to keep up with demand (Du Toit *et al.*, 1998; Potts, 2017). Of particular concern is the amount of material derived from wild harvested populations which are scattered throughout the Western and Eastern Cape provinces within the sensitive fynbos habitat (McGregor, 2017). Wild populations face a number of other threats including invasive alien trees, habitat loss and altered fire intervals (Rouget *et al.*, 2003). In conjunction, wild harvesting has been done for generations amoung local communities and farmers and is not only a primary livelihood but also has special cultural significance (McGregor, 2017).

The practice itself therefore has ecological, economic and cultural complexities which require acknowledgement if future solutions are implemented which ensure the sustainability of the industry. In light of this, options for expanding production through commercial planting are taking place but are limited in ways previously mentioned. One of the serious concerns with cultivation is proximity to wild honeybush leading to genetic contamination (Potts, 2017). The farming landscape within which honeybush occurs naturally, particularly wild *C. intermedia* makes it vulnerable to this concern as some populations which have distinct ecotypes may be located close to areas that are ideal for cultivation using either a different species or ecotype. Even if the same ecotypes are used, selective breeding pressures that are present in the wild are altered under cultivation and this may result in negative genetic changes which may be conferred if pollination between wild and cultivated tea occurs (Potts, 2017; Milla *et al.*, 2015).

As is the focus of this research, one plausible solution may be augmenting already existing wild populations by planting seeds and seedlings directly into the veld by using locally sourced seeds. If this method can be ecologically and economically justified then it is poised to be one of the solutions for honeybush farmers who rely on wild stocks and who are unable to use other means of increasing production. A cost benefit analysis (CBA) was used to quantify the economic prospects of different augmentation scenarios using higher and lower survival rates of seeds and seedlings as well as discounting rates. For the field trials, seeds and seedlings were planted into plots where wild populations of honeybush were present and in plots adjacent where it was absent. Plots were then also placed on different post fire aged veld.

#### 4.3. Materials and Methods

#### 4.3.1. Study Site

Four farms within the Langkloof and Kouga region of the Eastern Cape were chosen as augmentation sites. Three of the farms involved (De Hoek-farm 4, Somersets Gift-farm 3, and Walletjies- farm 2) were considered as separate case studies to Melmont (farm 1). This was due to constraining factors such as travel-imposed restrictions caused by the Covid-19 pandemic during the monitoring periods which created irregular monitoring intervals. Distance between the farms and cost were also practical limitations. For some of the farms only a small number of seed collected from parent populations was available for the trials which is why some farms have fewer study sites. The results from farms 2, 3, and 4 were not compared statistically to farm 1. Wild populations of *C. intermedia* are distributed across the region shown on the map (figure 4-1) but are confined to high mountain ranges. These ranges subsequently create climatic gradients which means that conditions may change abruptly over small distances. The general climate for the area is classified as Cfb according to the Köppen and Geiger (2011) system which is a temperate oceanic climate with precipitation spread throughout the year with an average of 650-710 mm annually and an average temperature of 16 degrees Celsius. This classification applies more accurately to farms 3 and 4 which are situated against the interior side of the Tsitsikamma mountains, known for having higher rainfall and more moderate temperatures. Climatic conditions for farms 1 and 2 are more arid, though differ between each other as well. Farm 1 is located further into the Kouga mountain range and has an average rainfall of approximately 450 mm. Farm 2 is located directly in the Langkloof region near Kareedouw which has an average annual rainfall of about 525-580 mm (Van der Waal *et al.*, 2012) and receives more rainfall than farm 1 but less than farms 3 and 4.

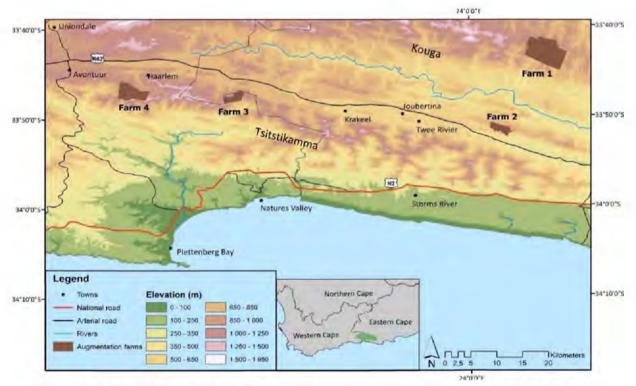


Figure 4-1: Map of the four farms where the augmentation trials were conducted. Farm 1 located in the Kouga mountains was chosen as the focus of this chapter

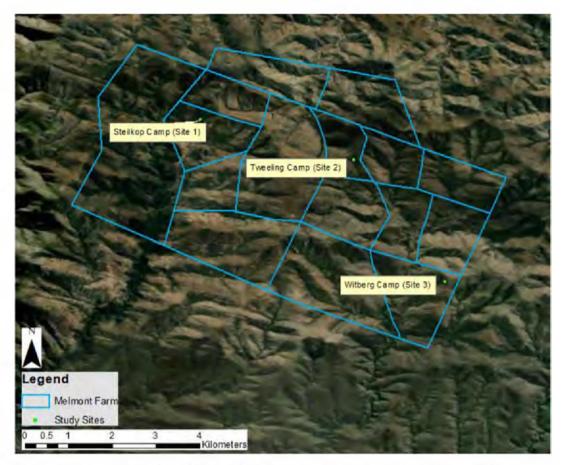


Figure 4-2: Farm 1 augmentation trial sites. Each site consisted of two plots, one inside an area where wild honeybush occurred referred to as a 'present' plot and one outside the population referred to as an 'absent' plot

## 4.3.2. Planting Procedure

Seeds used for the augmentation trials were sourced from each respective parent population found on the study site farms. This study used a scarification method based on local nursery experience which involved immersing the seeds in a small amount of boiling water for 30 seconds. Different scarification methods have been tested on *Cyclopia* species with varying degrees of success. As discussed, some of the standard methods include soaking in sulfuric acid solutions, dry heat treatments and wet heat treatments. Choosing the best method can be difficult as multiple factors such as seed age, species and colour may affect how resistant the coating is. In certain cases, wet heat treatments such as soaking seeds in boiling water for short periods of time (between 30 seconds and a few minutes) are the most effective method. Under lab conditions *C. intermedia* seeds had germination rates of 30 to 44% when treated in

boiling water for 10-60 seconds (Mbangcolo, 2008) whilst under field conditions germination only reached a maximum of 16% (Beyers, 2016) using 80°C water for three minutes.

Seedlings were then grown from these seeds in a nursery for 3 months prior to the trials. Seedlings kept under nursery conditions for longer (recommended 8 months) will ensure stronger plants and increase survival. Therefore, future research using longer trial periods is necessary to test the impact on survival rates. The 3-month nursery period used in this study was due to the seedlings not being grown in a nursery prior to the commencement of the study, and one of the main objectives of the study being to monitor the plants under veld conditions for one year.

The planting procedure involved first digging small holes about 10 cm deep for the seedlings and 2 mm deep for the seeds. The planting pattern was laid out as rings of seeds and seedlings beginning at 1 m distance from the center of the circle and moving outward by 1 m, shown in figure 4-3. One seedling was planted per site on three of the five rings whilst two seeds were used at each site on two rings, overall. Each plot totaled 24 seedlings and 64 seeds.

Each seedling was individually removed from its grow-tray and placed within the hole. Care was taken to ensure the roots were placed facing down and as much native soil from the grow

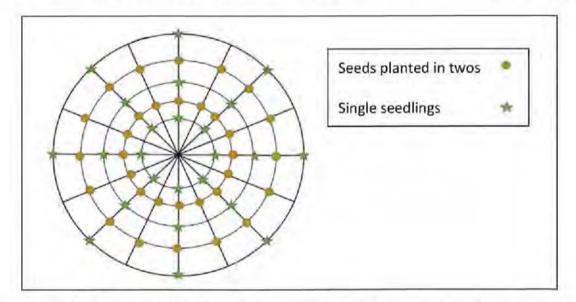


Figure 4-3: Augmentation planting plots were designed as concentric circles spaced 1 m apart from each other. Each plot had 5 rings and was 10 m in diameter with a total area of 78.53 m<sup>2</sup>. A total of 64 seeds and 24 seedlings were planted on each plot

tray was used to cover the roots. Thereafter each seedling was watered with approximately 60 ml. Seeds were given approximately 20 ml of water. On farm 1 the night after planting 13 mm of rain fell which helped to stabilize the seedlings and set the soil around the new holes. Fences were also constructed around each site to minimize the impact of herbivory and other possible disturbances. It is not possible to rule out interference from other animals such as baboons and mice which may have been responsible for seed and seedling losses. The same planting procedure was applied for the other farms, with the exception of fencing around the sites.

## 4.3.3. Plot layout and Treatments

Experimental plots were designed as concentric circles with a radius of 5 m covering a total area of 78.53 m<sup>2</sup>. The circles were constructed directly in the veld by planting a central pole in the middle, thereafter tape that was marked at 1 m intervals for 5 m was placed in eight cardinal directions outward from the center and secured with a pole at the outer ring to ensure it remained in position as shown in figure (4-4). Once this was completed, the circle was further divided up by measuring the distance between each outer pole marker and halving it to give 16 spokes in total. Rocks were then packed around the plot and marked with luminescent spray paint and planting lines on each spoke were further marked with large rocks. Seedling positions were also subsequently measured using differential GPS for long term monitoring. Each site had two plots and with two treatments. The first was 'honeybush'



Figure 4-4: Plot construction using a center pole and tape

indicating the presence or absence of wild honeybush already growing in the area where the plot was placed. The second treatment was post-burn age of the veld referred to as 'site age'. For the main study site (farm 1) site 1 was placed on 4-year-old veld (43 months) post fire, site 2 was burnt three days before planting and was considered 0 months and site 3 was on 7-month-old veld (figure 4-5). At the other farms the details about site layout and veld age are given as: De Hoek (farm 4) - 1 site on 3-year post burn veld, Somersets Gift (farm 3)- 2 sites on 1-year post burn and newly burnt, and Walletjies (farm 2) -1 site on unknown age veld.

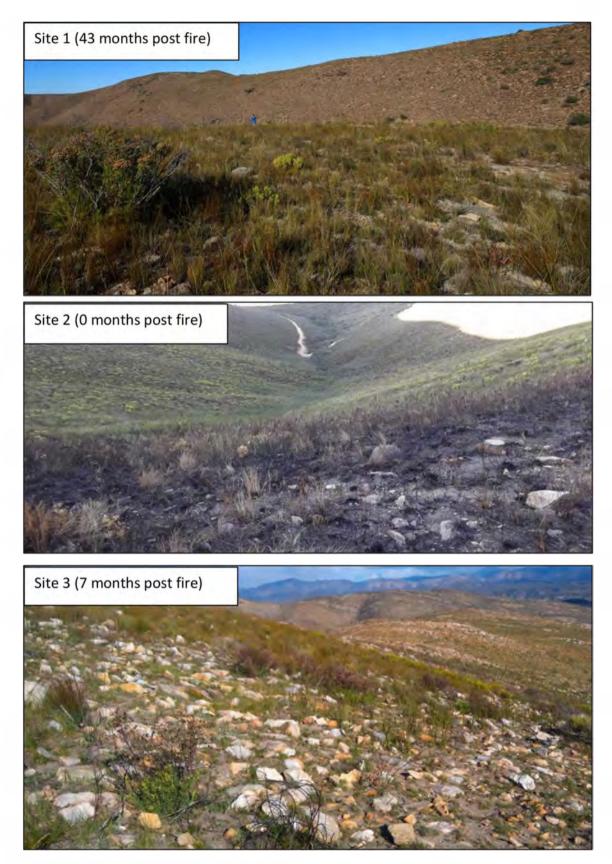


Figure 4-5: Augmentation sites on farm 1.

# 4.3.4. Monitoring

Seeds and seedlings were monitored and measured approximately every two months for one year (June 2020 until June 2021). The time interval between monitoring trips varied between from 8-14 weeks. Time after planting is given in weeks to illustrate more precisely the difference between monitoring intervals in table 4-1 below. Seedlings were individually measured from the base to tip of highest growth point both prior to planting and thereafter, average seedling length and growth rate was obtained for each site from these measurements. Classification of monitoring results were done in the following categories, for seeds: 1 (Germinated- living), 2 (germinated - dead), for seedlings: 1 (living), 2 (dead), 3 (herbivory/ disease). Plants which showed clear signs of herbivory or were destroyed by other means were excluded from growth analysis but included in the survival analysis.

Monitoring Trip	Weeks after planting	Interval
T1	11	
T2	20	9
ТЗ	30	10
T4	44	14
T5	52	8

## 4.3.5. Cost Benefit Analysis Procedure

The scope of the economic analysis in this thesis is to provide a basic overview of the expected profitability of augmentation based on the limited knowledge available. Additionally, the general focus of this research is on the ecological aspects of augmenting *C. intermedia* and therefore an expanded economic study is still required. Due to a lack of literature regarding potential costs and benefits involved in wild augmentation, and because of the specific processes that are unique to honeybush farming, information had to be gathered from different sources (appendix 3). Details about labour and planting densities per hectare were obtained from an upscaled augmentation project that was conducted in June 2021 (M. Sephton, *pers. comm.* 2021). The method applied in this section began with logging each material expense for both seed and seedlings covering the collection, seedling production and harvesting costs.

The study derived the costs of the nursery by pricing a basic setup of shade net and spray/mist irrigation for an area big enough to house 10 000 seedlings in Unigrow trays (appendix 4). The running expenses were calculated as having two part time employees for 3 months at R21.70 per hour (Fasken, 2021) for approximately 4 hours per day. This particular nursery setup is based on industry advisors experience and is representative of the type of setup a honeybush landowner would be willing to use as opposed to setting up a professional nursery. This influences the outcome of the CBA dramatically as both the size and nature of the operation is different. For example, the goal of a professional nursery is to sell the seedlings it produces annually and thus makes a profit within a short period of time as opposed to a honeybush farmer who would only derive benefits from seedlings which have survived 4-5 years after being planted. Additionally, the seedlings would only need to be grown in the nursery for 3 months during the late spring/summer season and thereafter it is not recommended to try produce seedlings. For this reason, an employee would only be necessary for 3 months. Bearing this risk in mind the study also compared the outcome of an augmentation investment using bought seedlings as opposed to seedlings produced by the landowners themselves in the instance that the landowner prefers not to establish a nursery.

The expense of seed used in this study is worked out as the cost of collection from a native/parent population on the farm that is going to be augmented. This is relevant because

buying seed sourced from other areas/populations will lead to genetic contamination. The calculation for this collection cost is based on the amount of time it would take a team of about five pickers to collect 1 kg of seed given a certain density of wild plants (M. Sephton, *pers. comm.* 2021). This calculation can be extremely variable. In this example the density of plants was approximately 1500-2000 per hectare. Next the cost of harvesting was roughly worked out from a residential farm staff team of about eight members and harvest quantity data reported in the wild honeybush harvesting guidelines (McGregor, 2017).

The amount each member can harvest per day is contingent on factors such as experience, weather and location and once more this quantity is extremely variable and will be difficult to extrapolate to other situations. The amount for an 'easy' site ranged between 43 and 166 kg per member (McGregor, 2017). Considering that augmented sites are being harvested the assumption will be made that these sites as well as the spacing of the plants will be easy to access and harvest and therefore an average of 100 kg per person per day was chosen. The approximate time taken during this study to plant seedlings (3 min/seedlings) was extrapolated to 1 ha in order to calculate the labour expense for seedling augmentation. The study also opted to exclude other expenses such as transport costs during harvesting trips and alternative harvesting team options such as hiring a team which then receive a percentage of the crop as opposed to being paid a day wage. The above assumptions and exclusions will influence the final outcome of the CBA and this must be kept in mind.

Next the expected benefits were calculated per hectare using the inputs shown in table 4-3. The recommended harvesting quantity of cropping 80% of plants out of the population every 4 years was used based on the current guidelines (McGregor, 2017). Average yield data from farm 1 ranged between 0.29 and 0.51 kg per plant (McGregor, 2017). Therefore, a median amount of 0.4 kg/ per plant wet material was used. The harvest yield per plant is kept constant for the sake of the calculation but note that yield will change with plant age and harvest regime. Honeybush plants which are harvested do produce more harvestable stems than plants which are not subject to harvesting (Barnardo, 2013), provided they are rested for sufficient intervals between harvests. The wet harvested material price of R 15 may be lower than what is paid for premium wild *C. intermedia* and this price may rise substantially in the future. However, this value was chosen as a conservative input for the CBA. Additionally, the

dry climatic conditions have resulted in reported yields being lower than usual, even for high rainfall areas (G.K. McGregor and M. Sephton, 2021. HBCOP virtual meeting).

The final step was calculating the net present value (NPV) and benefit to cost ratio for multiple scenarios considering different survival rates and discounting rates. The benefit to cost ratio was obtained by first calculating the total benefits (referred to as actual crop value) by subtracting the harvest expense from the crop yield that was gained. The actual crop value was then divided by the initial costs of the project to give a ratio. The NPV was calculated as the summation from the beginning of the project (time =0) to the end of the project (t = n). The standard NPV equation used was used:  $[(B_t - C_t) / (1 + r)^t]$  whereby B = Benefits; C = Costs; r = discount rate; t = time period. In this research a time frame of 4 harvest cycles or 16 years was used and thus the equation would be;  $[(B_0 - C_0)] + [(B_1 - C_1) / (1 + r)] + .... [(B_{16} - C_{16}) / (1 + r)]$ r)<sup>16</sup>]. This longer time frame was chosen because the initial seedling expenses are high and benefits are only derived every 4 years, thus crop cycles are slow. Unlike other crops however, which require continuous inputs such as pest management the only other expense after planting wild honeybush are those associated with harvesting. A second reason for the choice of time frame is that the species is extremely long lived, there are some populations which have been wild harvested for 35 years and other planted populations which have been harvested for their 5<sup>th</sup> time in 20 years (McGregor, unpublished PhD data, 2020). In table 4-4 a breakdown of the steps involved in the NPV calculation is given over a period of 4 harvest cycles. Table 5-5 provides the NPV for likely survival rates based on the results from the augmentation trials for the different seed and seedling options. The relationship between benefit to cost ratio for 5 and 8% discounting rate and increasing survival is explored further in the final analysis in figures 4-6 to 4-9. An example of the spreadsheet used in excel for calculation is provided as an appendix (table 7-5) and a link to the full spreadsheet is in the caption.

# Simplifying Assumptions

- Cost of labour; remains R21.70 per hour as given in year zero (2021)
- Cost of fuel; excluded
- Seed collection costs; based on certain density (1500-2000) and picking team size (5 members)
- Seedling production costs; based on small scale nursery (10000 per annum), limited staff (2 members) and growing period (3 months)
- Seed augmentation costs; based on actual projects, 1 ha using 10000 seeds and 5 staff members (R108.5/ha)
- Seedling augmentation costs; based on actual project and extrapolated to 1 ha. Planting time approx. 3 min/ seedling using 3000 seedlings and 5 staff members (R3375/ha)
- Harvesting costs; based on average harvesting data (100 kg/per person/per day) and full-time farm employees
- Discounting rates; based on official rates suggested (5-8%)
- Harvest yield; remains constant and based on average obtained from actual field data (0.4 kg/per plant)
- Honeybush price; remains constant from year zero (2021) at R15 per kg wet material

# 4.3.6. Statistical Analysis

All data were captured, organized and analysed using Microsoft Excel and the statistical programming language R version 4.1.0 (R Core Team, 2016). Seedling and seed data were tested for normality using Shapiro-Wilk tests, and all significance calculations were based on a 95% confidence limit (p<0.05). Once this analysis was done, data were treated as non-parametric and Kruskal-Wallis ANOVAs were used to test the treatments 'site age' (43 months, 0 months, 7 months) and (B) 'honeybush' (presence or absence of honeybush). Post

hoc multiple comparisons were done using Kruskal -Wallis multiple comparison test. Samples with shared superscripts are not significantly different. Labeling convention for the camps and plots at each site was as follows; Site 1: Steilkop present (1 P), absent (1 A), Site 2: Tweeling present (2 P) absent (2 A), Site 3: Witberg present (3 P) absent (3 A).

Seedling and seed results presented in table 4-6 show overall mean ± standard error. These values were calculated as follows: each monitoring trip produced data from which an average was calculated, after all five trips had been completed the averages from each trip were summed together and divided by five to produce a final average. Results presented in table 4-7 differ in that the means are calculated using only data from the final monitoring trip and don't include any data from previous trips. Therefore, two separate analyses were done. From a research perspective the first analysis was necessary because the study aimed to use a larger data set to determine the impact of the two treatments over the duration of the study period. The second analysis was done because in a practical farming situation the final result is the one which matters most, hence the need to determine if the results may be different when only the last set of data are used.

# 4.4. Results

## 4.4.1. Cost Benefit Analysis

Table 4-2 below provides the preliminary (collection, production, planting) and harvesting costs involved in an augmentation project for 4 alternative scenarios. The price per seedling is roughly halved after initial nursery construction (from R 3.25 to R 1.65) and the price for bought seedlings is more expensive (R 5.15) than privately produced. The inputs for the benefits calculation are the same for all options (table 4-2) and is calculated per hectare. Due to the augmentation trial results indicating that seed survival is much lower than seedling survival, a total of 10 000 seeds was chosen per hectare whilst only 3000 seedlings) in size is calculated for seedling option 2 B using 25% survival in table 4-4 as an example. The outcome of this scenario shows that the value of each harvest minus harvesting expense in present terms is R 10 560, the benefit to cost ratio is positive (1.52) and the net benefits are

R 14 480 after 16 years without applying any discount rate. However, if 5% depreciation per annum is applied then the investment becomes negative decreasing to - R 1206 and the B:C ratio is 0.96. This indicates that even after the nursery has been constructed the high expenses for production and planting of seedlings will outweigh the returns if 25% survival is achieved and the future value of benefits decreases by 5% per annum.

Increased survival will be necessary for such projects to become profitable. For example, the NPV of the same investment (seeding scenario 2 B) assuming a higher survival rate of 35% shows that the investment remains positive after 5% depreciation (R 9414) but this decreases drastically if 8% is applied (R 1280). Due to the low input costs of seeds the NPV and B:C ratios are less affected by the depreciation rates and returns are higher even under low survival scenarios. This is demonstrated in table 4-5 whereby if seeds are used and a survival rate of 7% on 1 ha is assumed then the NPV after 5% depreciation is R 6816 after 16 years, this decreases to R 4366 at 8%. Although returns increase sharply with survival for seeds, based on the augmentation trial results it is unlikely that more than 10% survival (6/64 seeds) can be expected. Figures 4-6 to 4-9 show the required level of survival for augmentation projects to exceed a B:C ratio of 1 or the point at which profitability is reached. For the project 2 A when the initial nursery construction expenses are accounted for an augmentation project will only become positive after 25% survival is achieved and when 5% discounting rate is applied a minimum of approximately 40% is needed. After the nursery has been constructed (scenario 2 B) the running costs and planting out costs requires a lower minimum survival rate to become positive, for example at approximately 28% survival after 5% discounting is applied the B:C ratio begins to exceed 1. Buying seedlings (scenario 3) to augment requires a drastically higher survival rate in order to cover for the high input costs, approximately 65% survival needs to be achieved for the benefit to cost ratio to exceed 1 at 5% discounting and at 8% discounting the ratio only exceeds 1 at 75% survival. The outcome of these scenarios is speculative as they assume for example, a static relationship between the income received for a harvest and the expense of the augmentation project. In reality, some of the variables used such as yield can fluctuate dramatically and will therefore change when the point of profitability is reached.

Table 4-2: Expenses associated with different seed and seedling augmentation scenarios.

Option 1: Seed			
Seed Weight	eed Weight Cost per kg		Sowing by hand (staff x 5) R21.70/hour
1g = 60 seeds (60000/kg approx.)	R3000/kg (hand collected)	0.17kg seed/ ha used (10000 seeds) = R510	R 108.50 (per ha)
			Total R 618.50 (per ha)
Nursery (Based on 10	000 seedlings/anr	num production)	
Expense		Cost	Recurrence
Infrastructure and bui	lding	R 16 000	Once off
Maintenance and run	ning expenses	R 1000	Per annum
Salaries (2 staff part ti	me)	R 15 000 (approx.)	3 months per annum
Seeds		R 510	Per annum
			Total R 32 510 (approx.)
Option 2: Produced S	eedlings		
Cost per seedling		Planting labour (staff x 5)	Total per ha (3000 seedlings)
		R21.70/hour	
(A) Year 1: R 3.25 incl. construction			R 13 125
(B) Thereafter: R 1.65	reafter: R 1.65 R 3375/ha (3 min/seedling)		R 8325
Option 3: Bought See	dlings		
R 5.15 (current price 2021)		R 3375/ha (3 min/seedling)	R 18 825
Harvesting Expenses (	All options)	R1.80 per kg (approx.)	

Table 4-3: Benefits calculation; items and procedure.

Option		Option: Seedlings (3000/ha)							
Benefits Calculation									
Survival	3%	5%	7%	25%	35%	50%			
Gain (Plants)	300	500	700	750	1050	1500			
Harvest Guideline (80% of population harvested every 4 years)	240	400	560	600	840	1200			
Yield (0.4 kg/per plant)	96 kg	160 kg	224 kg	240 kg	336 kg	480 kg			
Total Principal Value (per ha)	R 1440	R 2400	R 3360	R 3600	R 5040	R 7200			

Table 4-4: Example of NPV calculation and benefit to cost ratio for different discounting rates over 4 harvest cycles for scenario 2 (B) assuming 25% survival of 10000 seedlings planted

NPV and B:C									
	Initial Costs	Harvest 1	Harvest 2	Harvest 3	Harvest 4	Cumulative Total	<b>B : C</b> Ratio		
Production + Planting	-27 760								
Harvesting Costs		-1440	-1440	-1440	-1440				
Crop Yield Value		12 000	12 000	12 000	12 000	48 000			

Actual Crop Value (Total Benefits)		10 560	10 560	10 560	10 560	42 240	
Net Present Value (NPV)	(-27 760 + 42 240) = R 14 480			(42 240/ 27 760) = 1.52			
Net Present Value (NPV)	5% discounting rate			8% discounting rate			
Initial Year	-27 760	-27 760			-27 760		
Harvest 1	8688			7761			
Harvest 2	7147			5705			
Harvest 3	5880			4193			
Harvest 4	4838			3082			
NPV =	-1206			-7017			
B:C	0.96	0.96			0.74		

Table 4-5: NPV outcomes after 4 harvest cycles (16 years) for different augmentation options using seeds and seedlings. Values based on 10 000 seeds (1 ha) and 10 000 seedlings (3.33 ha)

Scenario	Survival (%)	Initial Costs	NPV	NPV (5%)	NPV (8%)
Seeds	3	618.5	4450	2568	1517
	5	618.5	7830	4692	2942
	7	618.5	11 209	6816	4366
Seedlings (2 A)	25	4 <b>3</b> 760	-1520	-17 206	-23 017
	35	43 760	15 376	-6586	-14 720
	50	4 <b>3 7</b> 60	40 720	9346	-2274

Seedlings (2 B)	25	27 760	14 480	-1206	-7016
	35	27 760	31 376	-9414	1280
	50	27 760	56 720	25 346	13 726
Seedlings (3)	25	62 750	-20 510	-36 169	-42 007
	35	62 750	-3614	-25 578	-33 710
	50	62750	21 730	-9644	-21 264

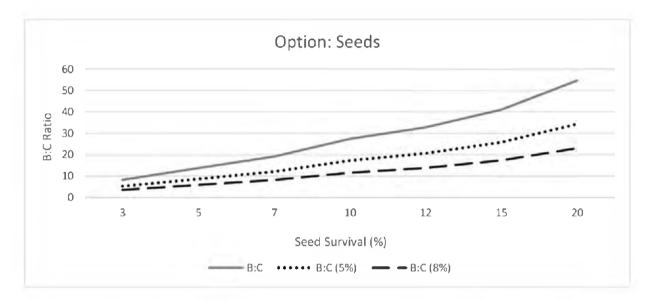


Figure 4-6: Relationship between survival and B:C ratio for 10 000 seeds planted on 1 ha

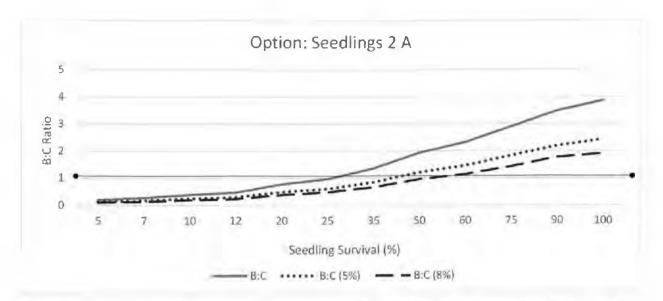


Figure 4-7: Relationship between B:C ratio and seedling survival for seedling scenario 2A based on 10 000 seedlings planted on 3.33 ha. The B:C ratio becomes positive at 25% survival when no discounting rate is applied. For 5% discounting it becomes positive at 40% and 8% discounting at 60%

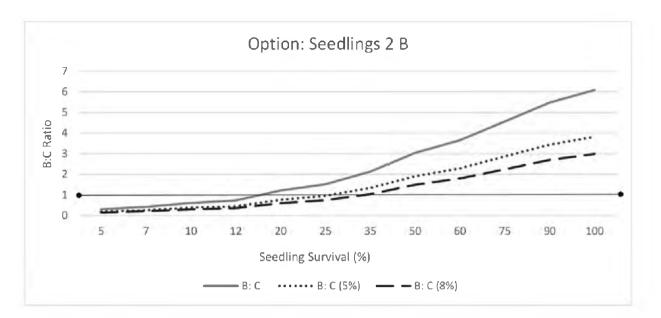


Figure 4-8: Relationship between B:C ratio and seedling survival for seedling scenario 2B based on 10 000 seedlings planted on 3.33 ha. The B:C ratio becomes positive at 15% survival when no discounting rate is applied. For 5% discounting it becomes positive at 25% survival and for 8% discounting it becomes positive at 35%

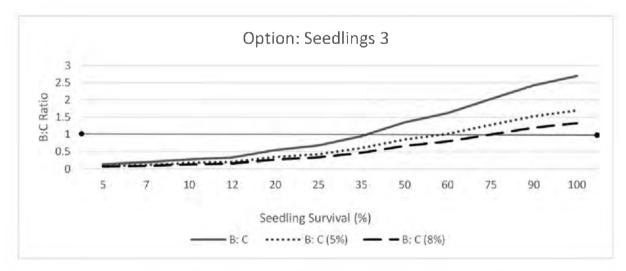


Figure 4-9: Relationship between B:C ratio and seedling survival for seedling scenario 3 based on 10 000 seedlings planted on 3.33 ha. The B:C ratio becomes positive at 35% survival when no discounting rate is applied. For 5% discounting it becomes positive at 60% survival and for 8% discounting it is positive at 70%

## 4.4.2. Augmentation Trials

All plots at each of the three sites at farm 1 experienced herbivory or disease dieback and these plants were therefore excluded from the seedling growth analysis but included in the survival count analysis. Overall, about 17 % of the seedlings were affected in this way. Per site the percentages of affected plants were as follows (mean and standard error calculated from both plots): site 1- 18.8 ± 6.7 %; site 2- 16.6 ± 4 %; site 3- 16.7 ± 5.8 %. Focusing on seedling growth results in table 4-6, the treatment effect of honeybush presence or absence proved significant according to the Kruskal-Wallis ANOVA (Chi-square = 6.3, p value= 0.012), whilst site age was an insignificant factor (Chi-sq = 1.7, p= 0.42). The difference in seedling size (mm) between the present (58.7  $\pm$  21.8) and absent plots (106.3  $\pm$  35.5) at site 1 proved to be significant whilst the other two sites had insignificant differences between their plots. Overall, the average seedling size between the sites was quite similar but site 2 (0 months post burn) showed the best seedling growth (table 4-6). In terms of seedling survival results, only site 1 absent plot had a significantly better survival rate than the present plot. The effect of both treatments was insignificant though only marginally for honeybush presence or absence (Chisq = 3.15, p= 0.076), site 2 had better overall survival with little difference between its plots. The same result was shown for site 3. For the seeds, approximately only 9% of all seeds planted germinated successfully across the sites and only the effect of 'site age' was significant (Chi-sq = 13.3, p= 0.01). Both plots at site 2 had significantly better germination than plots at site 3 and site 1 (presence plot). Germinated seeds which survived were also low, the present plot at site 2 had the best survival (14.3  $\pm$  4.4) followed by the absent plot (13.2 ± 5.7). In terms of treatment effect for survival only 'honeybush' proved significant (Chisq = 5.6, p= 0.04).

Table 4-6: Results for the three experimental sites on farm 1, showing overall mean ± standard error of seedlings and seeds. Significance of factors tested using Kruskal-Wallis ANOVAs. P-values significant at p<0.05 and values with shared superscripts are non-significantly

different. Means were calculated from data collected over 5 monitoring trips during the period of 1 year.

	Site 1 (P)	Site 1 (A)	Site 2 (P)	Site 2 (A)	Site 3 (P)	Site 3 (A)	Site Age	Honeybush
Seedlings								
							Chi-sq = 1.7,	Chi-sq = 6.3,
Height (mm)	58.7 ± 21.8ª	106.3 ± 35.5°	105.8 ± 41.2 <sup>c</sup>	107.8 ± 39.4 <sup>c</sup>	82 ± 31 <sup>ab</sup>	100.9 ± 37.8 <sup>bc</sup>	p= 0.42	p= 0.012*
							Chi-sq = 1.9,	Chi-sq = 3.15,
Survival (%)	38.3 ± 18.4ª	64 ± 23.2 <sup>b</sup>	56.8 ± 24.2 <sup>ab</sup>	62.4 ± 31.4 <sup>b</sup>	48.3 ± 17.5 <sup>ab</sup>	48.2 ± 22.6 <sup>ab</sup>	p= 0.38	p= 0.076
Seeds								
Germination							Chi-sq = 13.3,	Chi-sq = 0.4,
(%)	6.9 ± 1.1 <sup>ab</sup>	12.5 ± 3 <sup>dc</sup>	15.3 ± 5.2 <sup>d</sup>	$10.3 \pm 2.1^{bc}$	3.1 ± 0.85ª	5.6 ± 1.6ª	p= 0.01*	p= 0.57
							Chi-sq = 6.8,	Chi-sq = 0.23,
Survival (%)	9 ± 5.5ª <sup>b</sup>	12 ± 6.3 <sup>b</sup>	14.3 ± 4.4 <sup>b</sup>	13.2 ± 5.7 <sup>b</sup>	6.7 ± 6ª	$11.3 \pm 5.8^{ab}$	p= 0.04*	p= 0.62

Table 4-7 shows the results for data collected from the final monitoring period at farm 1. The effect of both treatments was the same as that of table 4-6 with two exceptions, namely 'honeybush' being a significant factor influencing seedling survival (Chi-sq = 5.15, p= 0.043) and 'site age' being insignificant in influencing germinated seed survival. The differences in seedling sizes are more pronounced in table 4-7. Both plots at site 2 and the absent plot at site 3 had significantly better growth than plots at site 1. Site 2 present plot showed the largest average seedling size (254.2 ± 79.2) followed by site 2 absent (246.7 ± 34) and site 3 absent plot (226.5 ± 97.2). For seedling survival, site 1 (absent plot) had significantly higher levels than all other plots (58 ± 12.3), whilst site 2 and 3 shared the same result with no difference between them. In terms of seeds,

cumulative germination levels were significantly higher in site 2 (present-  $20.3 \pm 2.2$ , absent-  $14 \pm 3$ ) and site 1 (absent-  $15.2 \pm 2.2$ ) compared with site 3 (present-  $4.7 \pm 0.75$ , absent- $6.3 \pm 1.6$ ) and site 1 (present- $7.8 \pm 0.5$ ). Although germinated seed survival rates show that the present plot at site 3 ( $33.3 \pm 3.6$ ) had significantly better survival than all other plots, there were only 4.7% of the seeds that had germinated.

Table 4-7: Results for the three experimental sites on farm 1, showing overall mean ± and standard deviation of seedlings and seeds. Significance of factors tested using Kruskal-Wallis ANOVAs. P-values significant at p<0.05 and values with shared superscripts are non-significantly different. Means were calculated only from data collected during the last monitoring period.

	Site 1 (P)	Site 1 (A)	Site 2 (P)	Site 2 (A)	Site 3 (P)	Site 3 (A)	Site Age	Honeybush
Seedlings								
							Chi-sq =	Chi-sq = 7.1,
Height (mm)	<b>161 ±</b> 69.1ª	198 ± 69.4 <sup>b</sup>	254.2 ± 79.2 <sup>c</sup>	246.7 ± 34°	156.8 ± 75.8ª	226.5 ± 97.2 <sup>bc</sup>	2.2, p= 0.53	p= 0.016*
							Chi-sq = 1.5,	Chi-sq = 5.15,
Survival (%)	12.5 ± 7.6ª	58 ± 12.3°	33.3 ± 8.4 <sup>b</sup>	<b>3</b> 7.5 ± 5.2 <sup>b</sup>	33.3 ± 11 <sup>b</sup>	37.5 ± 4.5 <sup>b</sup>	p= 0.4	p= 0.043*
Seeds								
Germination							Chi-sq = 8.6,	Chi-sq = 1.4,
(%)	7.8 ± 0.5 <sup>ab</sup>	15.7 ± 2.2°	20.3 ± 2.2 <sup>c</sup>	14 ± 3 <sup>c</sup>	$4.7 \pm 0.75^{a}$	$6.3 \pm 1.6^{ab}$	p= 0.005**	p= 0.33
						-	Chi-sq = 4.3,	Chi-sq = 2.85,
Survival (%)	$20 \pm 0.44^{a}$	20 ± 1.1ª	23 ± 2.8ª	22 ± 0.4ª	<b>33</b> .3 ± 3.6 <sup>b</sup>	20 ± 1.2ª	p= 0.06	p= 0.57

## 4.4.3. Seed Germination and Seedling Survival, Growth and Size as per

#### Monitoring Interval

Seed germination per monitoring trip is shown in figure (4-10) below. The highest levels of germination were observed for the present plot at site 2 (20.3%) and the absent plot at site 1 (15.7%). Germination also appeared to occur throughout the study period at these sites, indicating that seeds could germinate a year after initial planting. Germination levels were very similar for all plots after the first monitoring trip, thereafter it remained low for both plots at site 3 (present- 4.7% and absent- 6.3%) and for the present plot at site 1 (7.8%).

The results for seedling survival (figure 4-11) indicate that there was an uneven decline between monitoring intervals. All sites experienced a seedling mortality rate of approximately 15-25% between planting and the first monitoring interval, thereafter the levels between plots began to differ more drastically. For example, site 1 (present) decreased from around 75% to 45% and continued decreasing to 12.5% survival whilst the absent plot at the same site only decreased from 75% to 58% between the third and fourth monitoring interval, with no further seedling mortality being recorded. The present (33%) and absent (37.5%) plots at site 2 experienced a more gradual seedling mortality rate than the plots at site 3 (present-33% and absent- 37.5%) but declined enough during the last monitoring interval to produce exactly the same results. Figure 4-12 displays the results for average seedling sizes for the present and absent plots over 5-time intervals representing each monitoring trip.

The accompanying figure (4-13) measures the time since planting in weeks reference and shows the percentage growth obtained during each monitoring trip. The seedlings grew at a much slower rate during the first 11 weeks after planting (in winter), increasing in size by less than 20% with the exception of site 3 absent plot growing by approximately 25%. From 11 weeks to 20 weeks post planting (spring) the seedlings at site 3 and site 1 (absent plot) showed their highest growth increase approximately 100% and 112% respectively. Whilst site 2 plots increased by approximately 150% and 130% and site 1 present plot by 105% between the 20-week and 30-week period (summer). Growth rate then slowed drastically again between the 44-week and 52-week period.

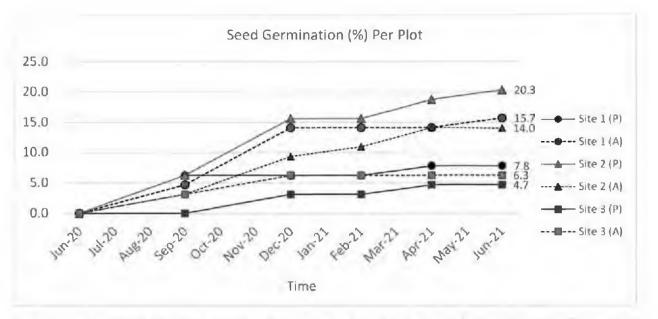


Figure 4-10: Cumulative seed germination for plots at farm 1. Site 2(P), 1(A) and 2(A) had the highest germination percentages of 20.3%, 15.7% and 14% out of 64 seeds planted. Germination at the other 3 plots was considerably lower with site 1(P) having 7.8%, site 3(A) 6.3% and site 3(P) 4.7%

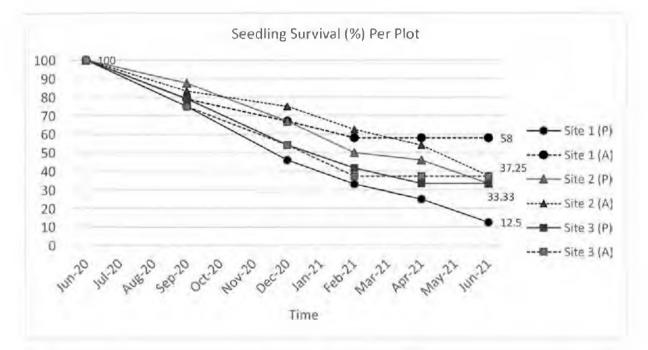


Figure 4-11: Seedling survival per plot for farm 1. Highest survival occurred at sites 1(A) (58%), 2 (A) and 3 (A) (37.5%) out of 24 seedlings planted. Plots 2(P) and 3(P) also shared the same survival rate of 33.3% and 1(P) had the lowest survival of 12.5%

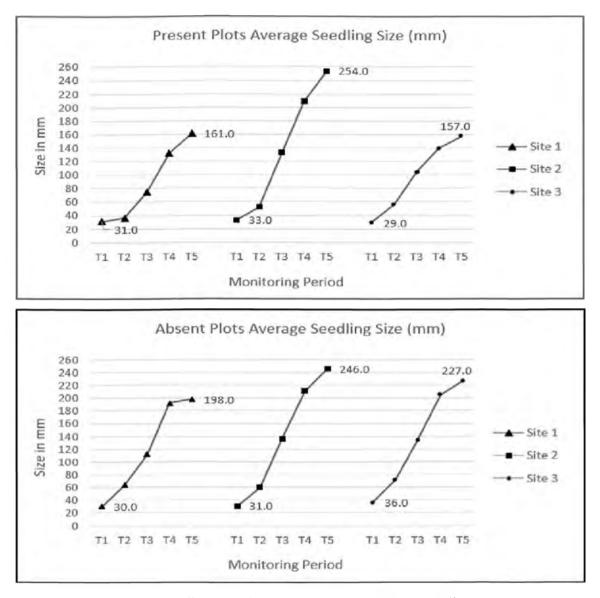


Figure 4-12: Average seedling size (mm) per monitoring trip. Seedlings grew most at sites 2(P) (254 mm) and 2(A) (246 mm) over the duration of the study. Seedling sizes at site 3(P) (157 mm) and 3(A) (227 mm) differed more drastically. Site 1 similarly showed more of a difference with 1(P) (161 mm) and 1(A) (198 mm)

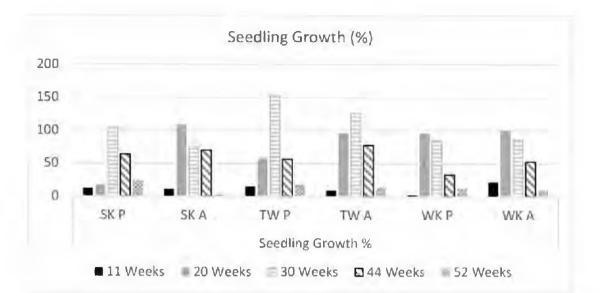


Figure 4-13: Seedling growth (%) post planting. Seedling growth was calculated relative to the average seedling height prior to planting. The slowest growth rates were observed for the first 11 weeks post planting. Thereafter the periods of 20- and 30-weeks post planting showed the most growth

# 4.4.4. Seed and Seedling Results for Farms 2, 3 and 4

The results presented in table 4-8 below show the survival rates as recorded 1 year post planting for the other farm study sites. The average seedling survival was extremely variable both between and within sites for each farm. Farm 2 had reasonably good survival for both the present (29 %) and absent (42 %) plots on 4-year-old veld. Farm 3 had very low survival for both plots at the site situated on an East facing slope (13 %), whilst the results at the West site were much higher for the present plot (71 %) but lower for the absent (21 %). Farm 4 had the lowest survival with only 4 % at the present plot and no survival at the absent. Seed survival rates were low to none throughout with the exception of the present (27 %) and absent plot (13 %) at the West site on farm 3.

Table 4-8: Augmentation trial results for farms 2, 3 and 4. Each plot received 24 seedlings and 64 seeds which totaled 48 seedlings and 128 seeds for each site. Farm 3(P) west site had the best seedling and seed survival of 71% and 27%. The equivalent opposite site at farm 3(A) had far lower survival with 21% and 13%. Farm 2 had the second-best results with seedling survival values of 29% and 42% for the present and absent plots respectively. The east site on farm 3 had the same seedling survival rate of 13% for both the absent and present plots

Study Site	Site Age	Honeybush	Seedling	Seed Survival
	(Years)	(Present/ Absent)	Survival (n=24)	(n=64)
Farm 2	4	Р	29%	0%
Farm 2	4	А	42%	6%
			-	
Farm 3 (East)	1	Р	13%	3%
Farm 3 (East)	1	A	13%	0%
Farm 3 (West)	2	Р	71%	27%
Farm 3 (West)	2	А	21%	13%
Farm 4	3	Р	4%	0%
Farm 4	3	A	0%	0%

# 4.5. Discussion

The economic and ecological aspects of augmenting wild populations of *C. intermedia* were explored in this chapter using a CBA method and field trials. The results are limited to the scale and nature of this research but are nonetheless insightful. As market demand continues to increase so does the incentive to harvest wild honeybush since the resource is already available. The industry urgently needs to shift towards a sustainable way of meeting this demand without pressuring wild populations. The possibility of increasing production can come from different approaches, including cultivation on old fields and ploughed lands (Beyers, 2016). These options however require lands which may not be available for every honeybush farmer. Increasing production by developing new lands is not a sound option as it will lead to further habitat transformation (Allsopp *et al.*, 2014).

In view of the evidence that many fynbos ecosystems are threatened or lost through agricultural development (Privett *et al.*, 2002) this thesis promotes an agro-ecological approach through augmenting wild populations of *C. intermedia* that prioritizes both commerce and conservation. However, in order for landowners to be incentivized to adopt such an approach it needs to make economic sense. The results from the CBA suggest that planting seeds (sourced from parent populations) may be the best option as risk is low because of low input costs and survival does not need to be unrealistically high for worthwhile returns. The benefit to cost ratio also increases rapidly with survival when compared with seedlings but the chances of survival are lower.

Looking at the augmentation trials, seed germination levels for C. intermedia in wild fynbos seem to be very low in most cases. This appears to reflect a natural aspect of the system, as low seedling recruitment rates for this species have been observed anecdotally by landowners (G.K. McGregor, pers. comm. 2020). The presence plot at site 2 produced a reasonable germination rate of 20%, and of those only 23% survived at the time of the final monitoring period (3/64 seeds). If these results were to be extrapolated to 1 ha of augmented veld using 10 000 seeds only 460 plants would be gained. Thus, a low survival rate of 5-7% per hectare for seeds is likely to be an accurate estimate to work with. It is important however to mention here that survival also appears to vary considerably. For example, the present plot for the West site on farm 3 had double the survival as the absent plot at the same site and most of the other farm's sites had either no survival or extremely low levels. The results from farm 1 similarly indicate a degree of variability, albeit lower. As for the seedling CBA results, the initial input expenses carry a heavier risk if the required levels of survival are not achieved. In order to break even on the costs of constructing a nursery and planting 10 000 seedlings approximately 40% survival is required after 16 years or 4 harvest cycles at a 5% depreciation rate, and 75% survival for a benefit to cost ratio of 1.8. It is important to mention here that this calculation is assuming that the costs and benefits are derived from that single augmentation project. In a practical farming sense, the investment risk of building a nursery will be distributed across multiple years of producing seedlings.

After nursery construction each hectare is approximately R 5000 cheaper to augment but the high cost of planting out the seedlings into the veld still translates into high risk. Option 2 B shows that approximately 55% survival is required to get a benefit to cost return that is worthwhile under 5% depreciation. The value of the seedlings option decreases substantially after a discounting rate is applied. This is due to the initial planting and producing expense (which is high) remaining the same, but the actual value of the crop (crop yield- harvesting expense) decreasing due to the depreciation rate. Alternatively, the low initial expense for seeds translates into less of an impact on the harvested crop value when the discounting rate it applied.

Considering the results from the augmentation trial, the reality is that only 1 plot on farm 1 had a seedling survival rate above 55% whist the others were on average about 20% lower. Due to the long project lifespan, low benefit to cost ratios achieved with 30-40% survival for seedlings may not be an attractive option for private investment. On the basis of these trials, even if seedlings can be produced cheaply, seeds are the better option. There are several factors to consider at this point which may impact survival and growth significantly.

## Weather Conditions

It is well established that plants are affected by the prevailing weather conditions in the immediate sense and by climate over long periods of time (Hoffman *et al.*, 2009). Unlike animals, plants cannot actively move in search of resources and therefore need to adapt to survive. The biome transition from fynbos to succulent Karoo is an example of how plants change in response to climate (Rebelo *et al.*, 2006).

Whilst the latter shapes plant physiology in the long term, exposure to extreme weather conditions over shorter periods can negatively affect growth and cause mortality (Hoffman *et al.*, 2009). For most fynbos species periods of drought and high temperatures are a common occurrence, this is also true for *C. intermedia*. When plants are young and have not yet developed deep root systems which contributes towards their capacity to survive these conditions they are at higher risk of mortality. For example, the drought survival ability of

seedlings for two fynbos species, *Passerina vulgaris* and *Leudadendron pubescens* was tested. After 77 days of no water, none of the seedlings had survived and average time to mortality was around 23-26 days (Lechmere-Oertel and Cowling, 2001). Considering the variability in the results, the general climatic conditions must be considered. During the study period, there was moderate to severe drought across the Langkloof region. Landowners at each of the study sites commented on the severity of the dry and hot conditions, particularly towards the end of summer in February 2021. When revisiting farm 1 later in the year (September 2021), landowner and honeybush farmer Quinton Nortjé remarked that it was the possibly the driest year on his farm in over 40 years of farming. The rainfall data in chapter 3 showed that the total rainfall over the study period was 362 mm which was approximately 90 mm lower than the amount measured in a previous study for the same farm (Barnardo, 2013). Although annual rainfall can fluctuate between 350-450 mm for the Kouga, the amount measured during this study period approaches the minimum end of that spectrum. Therefore, the results from the trials are a preliminary indication as to the expected survival under these kinds of rainfall conditions. Survival and growth may be far higher in years with better rain.

#### Site Location

Given the unusually dry and hot conditions, any factors compounding the loss of soil moisture such as slope aspect are important to consider. South facing slopes receive less sunlight and are therefore generally cooler and wetter. Although the difference in survival between the wettest (site 2) and driest site (site 3) was insignificant for farm 1, the impact of slope on survival was most evident on farm 3. The west facing site (present plot) had exceptionally high levels of survival and germination even 1 year post planting, whilst the absent plot at the same site had reasonably good survival. It is noteworthy that both the absent and present plots for the west site were placed on a steep slope which is shaded for most of the day, additionally the plots were placed near a seep and soil moisture would have been consistently high. When compared to the east site on farm 3, survival was very low and seed germination and survival was almost nonexistent.

#### Presence or Absence

The impact of planting at sites which share the same general conditions as those favoured by wild C. *intermedia* but do not have wild populations already growing in them is significant in some instances but not in others for seedling growth and survival. This complicates the interpretation of the results as no clear pattern seems to be present. The results from farm 1 show that overall absent plots did have higher survival percentages but that this was only significant when comparing plots for site 1. Higher seedling survival also seemed to correlate with better growth. The absent plots for site 1 and 3 had significantly larger seedlings than the present plots, but this result was not observed for site 2 (possibly due to the site being freshly burnt). For farm 2 the same result was shown in that the absent plot had higher survival than the present plot but the opposite was observed for farm 3.

## Post Fire Veld Age

Similar to the impact of honeybush presence or absence, the results for the effect of site age on growth and survival were mixed. On farm 1, the newly burnt site (2) had the largest seedlings overall and showed the greatest growth percentage increase between monitoring trips. This result seems to reflect the seedlings positive response to an immediate post fire environment and the subsequent increases in nutrients presented in chapter 3. Increases in nutrients such as potassium, phosphorus and calcium are known to promote plant growth and health (Wang *et al.*, 2013; Malhotra *et al.*, 2018). It's interesting to mention that site 2 also had the lowest incidence of disease dieback but had a high level of what appeared to be insect herbivory (pers. obs. 2021). This could be indicative of the role fire plays as a natural disease eradicator (Pausas and Keeley, 2019) and may prove highly beneficial at reducing losses for larger scale projects, especially if the honeybush in the area is prone to such problems. Currently there is sparse research about honeybush's response to nutrient changes particularly from natural events such as fires.

There is evidence however that some species such as *C. subternata* benefit substantially in the form of shoot growth from the addition of phosphorus fertilization but no significant result was observed for *C. intermedia* in the same study (Joubert *et al.,* 2007). Being an early

successional resprouter *C. intermedia* has adapted to deal with generally low levels of nutrients in fynbos soils through mechanisms such as mutualistic relationships with nitrogen fixing bacteria (*Rhizobia* spp.) which gives them a competitive advantage in post fire conditions (Schutte, 1995). The rise in some nutrients combined with reduced competition and wetter conditions of the site and the unique adaptations of the species to favour burnt veld likely explains the good growth and germination levels observed at site 2.

Other seedling trials also support better growth and survival on lands with higher phosphorus levels compared with fynbos veld for *C. intermedia* and *C. subternata* (Beyers, 2016). Whilst seedling growth and seed germination were higher in the freshly burnt site on farm 1, overall survival was not comparatively improved by burning. Therefore, it seems that burning veld before augmenting is not necessary for increasing survival, at least in the context of Kouga sandstone grassy fynbos. Burning may prove far more beneficial where fynbos is denser and competition for resources is greater. Other techniques of veld preparation which are vital in the wildflower farming industry such as ploughing were not considered in this study and may improve results. However, their impact on co-existent species such as those found in the surveys will likely prove negative.

## 4.6. Conclusions

This chapter aimed to investigate the economic and ecological outcomes of using augmentation to boost production. An important finding that must be considered when interpreting this research is the degree of variability when working within the fynbos system. Regarding the economics of augmentation, success is hinged upon survival rates and these are subsequently dependent on multiple factors which interact to affect the outcome. Some factors such as weather conditions and are beyond the bounds of human control. Given that during the study there were periods of drought conditions, it is entirely possible for future augmentation projects to achieve much higher germination and survival rates under different conditions.

Other factors which were considered in this research and which can be controlled for were post fire veld age and site placement- on favourable slopes and in honeybush present or absent areas. Judging from the growth analysis, *C. intermedia* benefitted considerably from cooler, more moist conditions and newly burnt veld although not enough is known about its natural response to nutrient changes to draw solid conclusions. Survival of seedlings was expected to be highest for the newly burnt site but was instead similar to an older site. Seed germination and survival appeared to be most affected by the fire. Though the results suggest that fire may not be critical for increasing seedling survival in Kouga grassy fynbos, it is important in reducing competition and may be more influential in improving results for areas with dense fynbos vegetation.

There are both positive and negative economic and ecological outcomes of using augmentation based on the results from this study though more experimentation is needed under different circumstances. The most encouraging economic prospects come from using seed as the risk to reward ratio outweighs that of using seedlings. The two biggest drawbacks of using seeds are the quantity needed and low survival rates. The main positive aspects are the ease of use since they do not require specialized growing conditions under a nursery setup and can be planted quickly in the veld and good benefit to cost ratio even under with low survival. Seeds may also be a good way to assess the suitability of a site since conditions which promote seed germination and survival will also do so for seedlings. If seedlings are used then the landowner needs to be aware that the process of planting seedlings out is arduous and expensive and combined with building a nursery a certain level of survival is needed to hedge against these high costs. In support of using seedlings, the study did find that survival is drastically improved compared to seed. Required levels of survival for good benefit to cost ratios for seedlings were not achieved at most sites.

In concluding, honeybush farmers have known the general habitat preferences for *C. intermedia* for a long time. The implication is that they understand what to look for in an area before considering augmentation. In view of this knowledge, there had not yet been attempt to quantify the potential impact of site characteristics on the survival and growth of seeds and seedlings. Most notably, results from this study suggest that 'absent' areas appear to be good candidate sites for augmentation, provided they share the same general characteristics. The scale, timing and location of this study must be kept in mind and larger scale augmentation with follow up monitoring programs are critical if this form of production is to be utilized.

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# Chapter 5: Wild *C. intermedia* population characteristics and seed colour and collection method effects on germination success

## 5.1. Aim

The aim of this chapter was to determine the characteristics of wild *C. intermedia* populations and to test the effect of seed collection method and seed colour on germination success. In order to achieve this aim, three objectives were carried out. The first objective was to survey and characterize wild populations of *C. intermedia* on different post fire aged sites with the purpose of determining demographic ratios and allometric measurements. The second objective was to collect seeds from wild populations at each of the study site farms and categorize the seeds according to how they were collected (hand-picked or netted) and colour (green or brown). The third objective was to run a germination test to determine the influence of collection method and colour.

# 5.2. Introduction

Collecting wild seeds for the purpose of agricultural production has been occurring for millennia and is the foundation of modern crops (Kislev *et al.*, 2004). Since reproduction by seed represents the overwhelming majority of terrestrial plant life cycles, understanding seed ecology is critical for correct management and utilization (Kislev *et al.*, 2004). This understanding is particularly important for species which have specialized seed adaptations that need to be accounted for (Khurana and Singh, 2001; Meyer *et al.*, 2012). In the context of Mediterranean biomes such as fynbos, specialized adaptations put many species into this category including *Cyclopia*. Much of the literature regarding *Cyclopia* seed ecology is focused on the biochemical aspects of germination and far less on wild viability rates. Furthermore, there is no research related to the effect that seed collection technique has on the germination. This information may be critical when deciding how to harvest seed from wild populations since it impacts maturity which in turn influences germination success (Mbangcolo, 2008; Koen *et al.*, 2017). From a nursery perspective, it is vital to know what

natural germination rates are since only a proportion of the total seeds planted will become seedlings. There are different methods for obtaining *Cyclopia spp.* seed from wild populations, one way is to simply collect up seed from pods that were attached to harvested honeybush bundles after they have been dried for processing (Q. Nortjé, *pers. comm.* 2020). The other two methods are to pick seed pods off plants once they are ripe or cover the plants in a fine mesh net and allow the pods to burst open themselves. The latter method has the advantage of catching the seed when they have naturally reached their optimal maturity level. When *Cyclopia* seeds mature they are generally dark brown to light brown but may also be green. This study separated seeds based on colour and collection technique and applied the same scarification method to all to test germination.

In terms of wild C. intermedia population management, there is currently a lack of understanding about natural demographics, including seedling recruitment and the potential differences in growth characteristics between populations. This information may be vital for determining if populations are declining or being altered by pressures such as harvesting. "The most important ingredient required to achieve a truly sustainable form of resource use is information" (Peters, 1994). The fact that C. intermedia is a slow growing resprouter and given the evidence that resprouting species generally have low recruitment rates (Marais, 2012), including Cyclopia spp. (Du Toit and Campbell, 1999) means that it may be particularly sensitive to these pressures. If populations are declining then augmentation may be a necessary management procedure. Establishing whether or not augmentation is necessary will require long term monitoring of both harvested and unharvested populations across the species range. Given the limited scale of this project the goal was to provide information about populations for an area as a preliminary assessment of their growth characteristics. This information may be particularly useful for future projects that combine data from other areas for a more robust analysis. As discussed in chapter 2, recording population characteristics from sites with different post fire ages in Mediterranean biomes is beneficial as fire and successional processes will exert an influence on these data. By recording population demographics and growth characteristics of honeybush at different post fire aged sites this will account for a broader representation.

# 5.3. Materials and Methods

# 5.3.1. Study Site

The study collected seeds from 4 study site farms located in the Langkloof and Kouga mountain range within the eastern extent of the Cape Floristic Region. Only honeybush populations on farm 1 were surveyed. Farms 3, and 4 are characterized by montane Tsitstikamma sandstone fynbos whilst farm 2 is better characterized by Kouga sandstone fynbos and farm 1 by Kouga grassy sandstone fynbos. The CFR is famous for its extremely biodiverse ecosystems and over 9000 species of plants have been found of which are confined to specialized habitats due to landscape heterogeneity (Manning and Goldblatt, 2012). The general conditions for each of the farms varied considerably, farms 3 and 4 receive higher average annual rainfall and are covered by medium to dense vegetation with tall proteoid, ericoid and restiod shrubs dominating on south facing slopes where the honeybush populations are found. Farms 1 and 2 receive less rainfall and the areas where seeds were collected from were dominated by a mixture of grasses and to a lesser extent shrub.

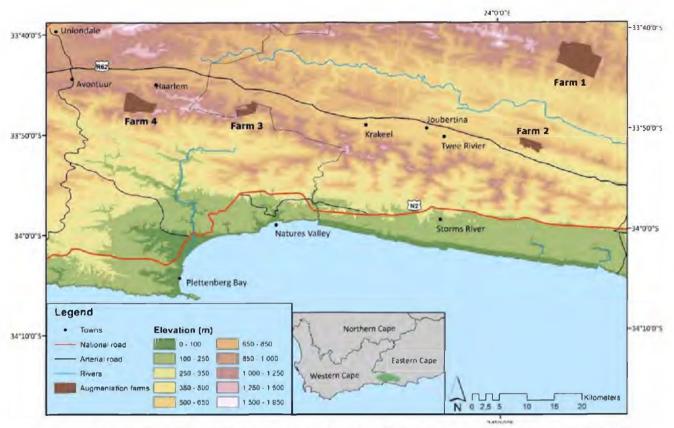


Figure 5-1: Study site farm locations across the Tsitstikamma and Kouga mountains. Seeds for the germination trial were collected from wild populations of *C. intermedia* from all farms whereas only wild populations from farm 1 were surveyed for population characteristics

# 5.3.2. Demographic Survey

Five populations of wild *C. intermedia* were surveyed on farm 1 using 5 x 25 m belt transects. At each site one transect within the population was conducted. The transect positioning was chosen based on visual assessments of the sites to include as many plants as possible. Each site was unique in that the plants were unevenly spaced apart and some areas had dense patches with no plants in between whilst others had individuals spaced more evenly apart. The allometry of each plant was recorded in terms of its height (from ground to highest canopy tip), basal circumference, canopy size and approximate number of stems. Plants were classified based on number of stems; 1- seedling, 2-5 young and mature >5. Using stem counts, basal circumference, or lignotuber size as a surrogate for age are all known methods for studying resprouters (Bowen and Pate, 2004; Lamondt *et al.*, 2011). Although new stems

sprout post disturbance from meristematic tissue located on the rootstock, the number of new stems produced after each disturbance is unknown for *C. intermedia*. Seedlings were easily classified because they had a single stem protruding with no evidence of burning. Bases of young plants were checked for evidence of burning to confirm that the plant was at least as old as the previous disturbance. Most of the mature plants had more than 15 stems and were easily differentiated from the young plants. Canopy size was measured as the length across the longest axis and then again measured perpendicular to that across the widest axis. Height and canopy size were determined using a wooden dowel marked with a tape measure and basal circumference was measured by placing a plastic tape measure around the base close to the ground. These data were collected as a joint effort for two honours research projects at Rhodes University (B. Hofmeyr and T. Makhuza). Three sites are subjected to a similar disturbance history in terms of harvesting, and two sites are unharvested. All harvested sites have not been harvested since their previous fire and are therefore similar in that respect as well. Details for each site are provided in table 5-1 below.

Table 5-1: Disturbance history characteristics for the population survey sites on farm 1. Populations at BK 1, BK 2 and SK all share the same fire history over the last 5 years. The populations at BK 2 and WB are unharvested whereas all other populations have been subject to harvesting

Site Name	Disturbances	Age of population post fire
BK 1	Burnt (2016), harvested site	Approx. 5 years
BK 2	Burnt (2016), unharvested for 20 years	Approx. 5 years
SK	Burnt (2016), harvested site	Approx. 5 years
TW	Burnt (2020), harvested site	Approx. 1 year
WB	Burnt (1996) unharvested	Approx. 25 years

## 5.3.3. Seed Collection and Sorting

Seeds from wild populations were collected from December 2020 until January 2021 using two methods- either collection by hand or by net. For the latter, plants with seed pods were selected from each population and wrapped with a fine mesh to collect seeds once pods had ripened and popped open. This method was based on Baskin and Baskin's (1998) guidelines for seed harvesting that recommends only fully mature seeds should be collected by allowing the pod to naturally mature on the plant and open. Collection by hand involved picking seed pods directly off the plants only if they were dark brown and dry. Both closed and open pods were collected using this method. For farm 3 seeds were only collected by picking and therefore no comparison with the net collection was done. Once the seeds had been collected, they were categorized according, collection method (hand or net) and colour (green or brown). Some populations produced very limited seed pods resulting in a slightly smaller sample size available for to use. The total sample sizes are given in each instance (table 5-5). Sorting was done by inspecting the seeds and separating them into green and brown groups. This method is limited in that a personal bias based on visual perception is introduced.





Figure 5-2: Picture (A) seed collection using a net wrapped around unripe pods and (B) separation of green and brown seeds

#### 5.3.4. Germination

All seeds received the same scarification treatment of soaking in boiling water in a cup until the water cooled. This method was based on local practitioners' experiments with various scarification times (M. Sephton, *pers. comm.* 2021). The seeds were then stratified by storing at fridge temperature (5° C) for approximately 3 weeks until the trial began. Seeds were then planted into trays with a medium of 50% vermiculate and 50% sieved potting soil and placed approximately 1.2 cm under the surface. Germination was counted once the seedling shoot was visible.

#### 5.3.5. Statistical Analysis

Data were recorded and managed using Microsoft Excel and analysed using the statistical programming language R version 4.1.0 (R Core Team, 2016). Allometric data compared between populations was heteroscedastic, therefore a Welch's ANOVA was used to determine if site had a significant influence and between site differences were tested using two-sample Welch T-Tests (Moder, 2007). Means with shared superscripts are not significantly different. Each size class in table 5-4 was analysed separately. Germination results were analysed within farms, using paired T-tests, p-values significance level was set to p < 0.05. To determine the effect of collection method, both green and brown seeds collected using nets were categorized together and compared to green and brown seeds collected using either method were combined and compared against brown seeds collected using either method.

#### 5.4. Results

#### 5.4.1. Population Structure and Characteristics

In table 5-2 below the number of seedlings, young and mature plants found for each population on farm 1 is presented. The average  $\pm$  standard deviation for allometric measurements in terms of number of stems, height, basal circumference and canopy size is provided in table 5-3. Site BK 1 had the highest density of plants (32) and 4 seedlings were

found at this site, all other sites had no seedlings. The proportion (%) of seedlings and young plants to mature plants for this site was (12: 16: 72). The average number of stems for young plants at BK 1 was  $4.2 \pm 0.7$  and  $18 \pm 13.4$  for mature plants. The basal circumference, canopy size and average plant height for mature plants at this site was  $48 \pm 30.5$  cm,  $2433.2 \pm 2095.4$  cm<sup>2</sup> and  $245.6 \pm 151.4$  cm. At site BK 2 there were 2 young plants found and 16 mature plants which means that seedling recruitment is taking place albeit in the years before the time of sampling. The average height and number of stems for mature plants at site BK 2 was  $60.4 \pm 10.7$  cm and  $32.6 \pm 24$  and basal circumference was  $53.3 \pm 26.1$  cm. Site BK 2 has been unharvested for approximately 20 years and has the second largest canopy size for mature plants ( $4199 \pm 2022.7$  cm<sup>2</sup>), the other unharvested population at WB is the oldest and had the largest canopy size ( $12240.4 \pm 4621$  cm<sup>2</sup>). No young plants were found at sites SK and WB. The average number of stems and height for mature plants was SK was  $28.7 \pm 14.8$ ,  $44.4 \pm 10$  cm and for TW it was  $45 \pm 41.5$ ,  $54.4 \pm 14$ . Basal circumference for sites SK and TW were similar ( $61.4 \pm 28$  cm), ( $69.3 \pm 34.4$  cm). Basal circumference and average height for WB was  $116 \pm 24.5$  cm and  $71.3 \pm 14.5$ .

Table 5-2: Age structure of 5 wild *C. intermedia* populations on farm 1. In total 87 plants were surveyed from all the sites. Only 4 seedlings were identified at one site (BK 1). The number of young plants found were as follows; BK 1 (5), BK 2 (2), and TW (1)

Site Name + Last Fire	Seedlings (S) (1 Stem)	Young (Y) (2-5)	Mature (M) (> 5)	Total Plants	Ratio (%) (S: Y: M)
BK 1 (Dec. 2016)	4	5	23	32	12: 16: 72
BK 2 (Dec. 2016)	0	2	16	18	0: 11: 89
SK (Dec. 2016)	0	0	16	16	0: 0: 100
TW (Jun. 2020)	0	1	13	14	0: 7: 93
WB (1996)	0	0	7	7	0: 0: 100

Table 5-3: Average allometric measurements for the 5 wild populations on farm 1. The average number of stems was associated with increasing basal circumference as expected for this species but not in every instance. Growth characteristics of the same demographic groups from different populations varied. The average number of stems for mature plants at BK 1 was  $18 \pm 13.4$  and basal circumference, canopy size and average plant height was  $48 \pm 30.5$  cm,  $2433.2 \pm 2095.4$  cm<sup>2</sup> and  $245.6 \pm 151.4$  cm. Mature plants for BK 2 had an average stem count of  $32.6 \pm 24$  and basal circumference, height and canopy size was  $53.3 \pm 26.1$  cm, 60.4 cm and  $4199 \pm 2022.7$  Site SK mature plants had fewer stems  $28.7 \pm 14.8$  but a larger basal circumference of  $61.4 \pm 28$  compared with BK 2. Mature plants at TW and WB had average stems counts of  $45 \pm 41.5$  and  $80 \pm 27.4$  and basal circumferences of  $69.3 \pm 34.4$  and  $116 \pm 24.5$  respectively

Site name	Stems	Basal circumference (cm)	Height (cm)	Canopy size (cm²)
BK 1				
Seedlings	1±0	5.75 ± 3.9	29.5 ± 4.2	198.5 ± 267.3
Young	4.2 ± 0.7	10.8 ± 4.1	26.7 ± 3.5	245.6 ± 151.4
Mature	18 ± 13.4	48 ± 30.5	47.8 ± 9.8	2433.2 ± 2095.4
BK 2				
Seedlings	0	0	0	0
Young	4.5 ± 0.5	22 ± 3	56 ± 13	975 ± 327
Mature	32.6 ± 24	53.3 ± 26.1	60.4 ± 10.7	4199 ± 2022.7
SK				
Seedlings	0	0	0	0
Young	0	0	0	0

Mature	28.7 ± 14.8	61.4 ± 28	44.4 ± 10	3529.7 ± 3334.5
TW				
Seedlings	0	0	0	0
Young	4 ± 0	10 ± 0	43 ± 0	50 ± 0
Mature	45 ± 41.5	69. <b>3</b> ± 34.4	54.4 ± 14	2914.3 ± 2897
WB				
Seedlings	0	0	0	0
Young	0	0	0	0
Mature	80 <b>± 27</b> .4	116 ± 24.5	71.3 ± 14.5	12240.4 ± 4621

In order to further determine growth characteristics, mature plants (>5 stems) of the unharvested (BK 2) and harvested sites (BK 1) (SK) (TW) were categorized according to their number of stems and, the average canopy size (cm  $^{2}$ ) ± standard deviation per stem group was then calculated (table 5-4). Due to the large discrepancy between post fire age of site WB and the other sites it was excluded from the analysis. Variability in growth characteristics is a feature of natural systems and thus, each site would, by virtue of this variability be different. The analysis was done in order to improve our understanding of this variability. The average canopy size for the 6-20 stem class was largest for the site BK 1 (1695  $\pm$  1767 cm <sup>2</sup>) and smallest at site TW (1082  $\pm$  530 cm<sup>2</sup>). The difference between canopy sizes for this stem class was insignificant (p=0.19). Similar results were shown for the 21-40 stem class as the canopy size was largest for the population BK 2 (5037  $\pm$  1541 cm <sup>2</sup>) and smallest at TW (1607  $\pm$  804 cm <sup>2</sup>). Canopy sizes for sites SK and BK 1 were  $(3394 \pm 1684 \text{ cm}^2)$   $(3770 \pm 2195 \text{ cm}^2)$  for the 21-40 stem class and site TW had a significantly smaller canopy than all the other sites (p=0.01). For the >41 stem class, site SK had the largest canopy size (7716  $\pm$  5232 cm <sup>2</sup>) and BK 1 has the smallest (6406 ± 2877 cm<sup>2</sup>). Differences between sites for this stem class were insignificant (p=0.12).

Table 5-4: Average ± standard deviation for canopy size (cm<sup>2</sup>) per stem class for 3 harvested and 1 unharvested population. Age of regrowth was equivalent for sites BK 1, BK 2 and SK (approx. 5 years). Significance testing was done with a Welch's one- way ANOVA and individual comparisons done with Welch's t-tests. Values with shared superscript are not statistically significant, each stem class was analysed separately

Class (stems)	Site				P-value
	BK 1 (harvested)	BK 2 (unharvested)	SK (harvested)	TW (harvested)	
(6-20)	1476 ± 1295 ª	1695 ± 1767 ª	1595 ± 346 °	1082 ± 530 °	P= 0.19
(21-40)	3770 ± 2195 °	5037 ± 1541 ª	<b>33</b> 94 ± 1684 <sup>a</sup>	1607 ± 804 <sup>b</sup>	P= 0.01*
(>41)	4988 ± 1776 ª	5628 ± 787 °	7716 ± 5232 ª	6406 ± 2877 °	P= 0.12

#### 5.4.2. Overall and Mean Germination by Factor Type

Table 5-5 below shows the germination results obtained for seeds collected from different study site populations. The majority of mixed seed germinated (58.2%) which was a random assortment of both green and brown seeds collected using both methods from all farms. Farm 1 had the highest overall germination result (79.2%) followed by farm 3 (46.4%), then farm 4 (27.8%) and lastly farm 2 (22.5%). The average number of net collected seeds which germinated (16.5  $\pm$  0.7) was higher than picked seeds (12  $\pm$  2.8) for farm 1. The opposite was observed for farm 2 between net collected (1  $\pm$  1.41) and picked (7  $\pm$  2.8). Farm 3 showed similar results as farm 1, net collected seed had an average germination of 9.5  $\pm$  6.3 whilst picked seed was 6.5  $\pm$  4.9. In terms of seed colour, the average number of green seed germinated was higher than brown seed for both farm 1 (15  $\pm$  1.4), (13.5  $\pm$  4.8) and farm 3 which had (12  $\pm$  2.8) and (4  $\pm$  1.4) respectively. For farm 2, green seed (3.5  $\pm$  2.1) had slightly lower germination than brown (4.5  $\pm$  6.3). Farm 4 only had picked seed and green (38.9) performed better than brown (16.7).

Table 5-5: Germination test results for seeds collected from study site farms. The labelling of each category was as follows: NG- green seeds collected by net; NB- brown seeds collected by net; PG-green seeds collected by picking; PB- brown seeds collected by picking. Mixed seeds taken from all farms had a germination percentage of 58.2% out of 98 seeds. Seeds from farm 1 germinated most successfully with 79.2%, farm 3 had the second-best germination rate of 46.4%, followed by farm 4 with 27.8% and farm 3 with 22.5%

Source	Seed Type	Sample Size (n)	Germinated	Germinated %	Mean	Mean
All farms	Mixed	98	57	58.2		
Farm 1		72	57	79.2	-	
			57	79.2		
	Net Green	18	16	88.9	16.5 ± 0.7	15 ± 1.4
	(NG)				(NG+NB)	(PG+NG)
	Net Brown (NB)	18	17	94.4		
	Picked Green	18	14	77.8	12 ± 2.8	<b>13</b> .5 ± 4.8
	(PG)				(PG+PB)	(NB+PB)
	Picked Brown	18	10	55.6		
	(PB)					
					I	
Farm 2		71	16	22.5		
	Net Green	17	2	11.8	1 ± 1.41	3.5 ± 2.1
	(NG)				(NG+NB)	(PG+NG)
	Net Brown (NB)	18	0	0.0		

	Picked Green	18	5	27.8	7 ± 2.8	4.5 ± 6.3
	(PG)				(PG+PB)	(NB+PB)
	Picked Brown	18	9	50.0		
	(РВ)					
Farm 3		69	32	46.4		
	Net Green	17	14	82.4	9.5 ± 6.3	12 ± 2.8
	(NG)				(NG+NB)	(PG+NG)
	Net Brown (NB)	18	5	27.8		
	Picked Green	18	10	55.6	6.5 ± 4.9	4 ± 1.4
	(PG)				(PG+PB)	(NB+PB)
	Picked Brown (PB)	16	3	18.8		
Farm 4		72	20	27.8		
Farm 4		/2	20	27.8		
	Picked Green	36	14	38.9	10 ± 5.6	
	(PG)				(PG+PB)	
	Picked Brown (PB)	36	6	16.7		

### 5.4.3. Colour and Collection Method

Table 5-6 below shows the results from the T-tests, for farm 2 there was a significant difference between the net and picked seed (p=0.04), whilst farm 3 had a significant difference between the green and brown seed germination (p=0.04). No other significant results were obtained. Figure 5-3 represents the data graphically, and shows more clearly the differences between farms was greater from the net collected samples than the picked samples. Germination tempo is shown in figure 5-4 per farm, samples were grouped together and an overall germination rate is given. All samples showed an increase from the first monitoring time 38 days after planting until the last, 77 days later with the exception of farm 2 which showed no further germination after the 38-day period. Farm 1 seeds germinated most between 56-77 days after planting (39 % to 79 %), farm 3 had 27.5 % at 38 days which remained the same until 56 days, then increased to 46 % at the 77-day period. Farm 4 had a similar germination tempo showing no germination from 38-56 days thereafter it increased from 14 % to 28 % until 77 days.

Table 5-6: Impact of colour and collection method on germination rates within farms. Germination differed insignificantly between netted and picked seed for farm 1, similarly green and brown seed also differed insignificantly for farm 1. Netted seed germinated significantly better than picked seed for farm 2 (P=0.04) but insignificantly between green and brown seed. For farm 3 netted and picked seed germination differed insignificantly but green seed germinated significantly better than brown seed (P=0.04)

Data	Test statistic (T value)	P(T<=t) two-tail	Actual difference in means
Farm 1 (NG+NB vs PG+PB)	T= 2.18	P= 0.27	3.5
Farm 1 (NG+PG vs NB+PB)	T= 0.33	P= 0.79	1.5
Farm 2 (Net vs Picked) (NG+NB vs PG+PB)	T= -2.7	P=0.04*	6
Farm 2 (Green vs Brown) (NG+PG vs NB+PB)	T= -021	P= 0.86	1
Farm 3 (Net vs Picked) (NG+NB vs PG+PB)	T= 0.52	P= 0.64	3
Farm 3 (Green vs Brown) (NG+PG vs NB+	T= 3.5	P= 0.04*	8

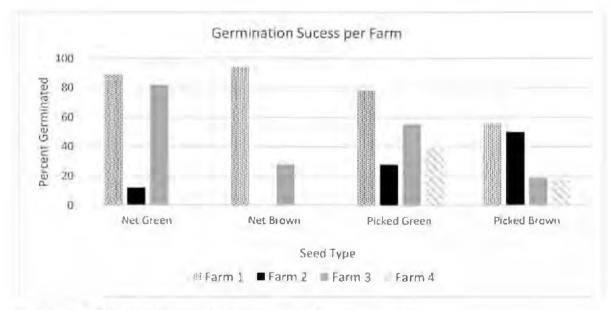


Figure 5-3: Germination (%) by colour and collection method

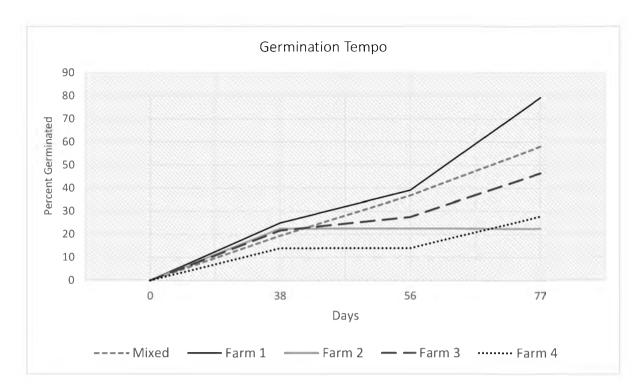


Figure 5-4: Germination tempo over the duration of the trial. Monitoring of germination ceased after 77 days

### 5.5. Discussion

Given the importance of wild *C. intermedia* populations to the honeybush industry and the potential rising demand, managing the resource is critical at this time. A component of this management is understanding population characteristics. Supplementing wild populations of plants to manage the impact of human pressure is a well-known conservation technique (Godefroid *et al.*, 2011). The problem in the context of the honeybush industry is establishing whether or not our impact on wild populations necessitates augmentation. If harvesting negatively alters plant lifespans, growth or seed production this may lead to even slower recruitment rates and eventual population decline (Tickten, 2004).

The results presented in this study are limited by scale and location and may therefore not be representative of other populations. Sample sizes were small for some of the allometric comparisons which may have influenced the results (de Winter, 2013). Further studies using large-scale surveys between different areas are still required.

It has already been recognized that resprouting species such as *C. intermedia* are heavily dependent on soil stored seedbanks and it is recommended that plants be allowed to flower and drop seed as many times as possible between harvesting intervals (Joubert *et al.*, 2011; McGregor, 2017). The impact of lowered recruitment rates and altered demographic structures may only become obvious over a long period of time, hence the need to develop monitoring programs for harvested wild populations. Research suggests that recruitment rates for resprouting species in fynbos are dynamic (Van Wilgen and Forsyth, 1992) but are generally much lower than those of reseeders (Marais, 2012). Recruitment rates for wild *Cyclopia* populations are not well understood. Another resprouter, *C. longifolia* was shown to have a rate of 3.8 seedlings per adult (Du Toit and Campbell, 1999) far less than the reseeder *C. pubescens*. Similarly, *C. genistoides* a resprouter was shown to have fewer developed and filled seed pods than *C. subternata* indicating lower recruitment potential (Motsa *et al.*, 2017).

As fire is an essential component for *C. intermedia* seed germination, it was expected that the populations which were burnt in 2016 would show some levels of recruitment. Although, seedings may have germinated and died between the time of the fire and surveying, only 1 site actually had seedlings. Two of the other sites had a small proportion of young plants, indicating that some recruitment had occurred in the past few years. Harvested populations which are able to sustain good levels of recruitment could be an indication of proper management. Determining what the appropriate levels of recruitment are for *C. intermedia* is yet to be done and may be complicated. This is due to a host of factors that influence recruitment such as disturbance history, population density and site conditions (Kraaij *et al.,* 2013). In view of the evidence that recruitment even under good management can maintain pace with impending demand, a precautionary recommendation can be made that populations which are subjected to increasing harvest pressure should be supplemented by planting seeds or seedlings sourced from the same parent populations.

This chapter also aimed to provide information about the growth characteristics from different populations. Due to a number of factors which may influence growth such as topography, rainfall and competition, ascribing possible reasons for these differences is complex. Thus, observations about variation are not presented as definitive. The average number of stems, large canopy size and large basal circumference for the WB population was expected as these plants had the most time to grow post disturbance. Increasing basal circumference and number of stems is naturally correlated for resprouters as more shoots are able to be initiated from larger bases (Keeley *et al.*, 1999). The size of basal circumference and average number of stems for mature plants displayed this growth feature for all the other populations. The only minor difference being that plants at site SK had an average of 28.7 stems with a 61.4 cm basal circumference and plants at site BK 2 had more stems (32.6) but a smaller circumference (53.3 cm). The differences in stem counts were likely due to the mature populations being older or younger. Most of the mature plants at BK 1 were younger (between 10-20 stems) than BK 2 (20-40 stems) for example. This would have also influenced overall canopy size.

Average plant height appears to increase rapidly in the year following a disturbance and then growth tapers down. This is evidenced by the 1-year post fire plants at TW being slightly taller than the 4-year-old populations at BK 1 and SK. Similar results were shown in a separate study when C. intermedia plants ranging from 1 to 4 years post-harvest were compared, no significant difference was found between the heights, and in some instances the 1-year plants were actually taller than the older populations (Barnardo, 2013). Taller honeybush may be a result of surrounding community competition forcing the plants to grow upwards for sunlight. The TW population was located on a moist south facing slope near a drainage line and the surrounding community was comprised of taller shrubs (particularly *Cliffortia linearifolia*). Site BK 1 has been described as an 'untypical' honeybush area by the landowner (G.K. McGregor, pers. comm. 2021) as the slope is almost due west facing and the plants appear to be naturally stout but abundant. The population at site BK 1 was harvested a few months before being burnt in 2016. Given what we know about the ecology of resprouters two consecutive disturbance without enough recovery time could lead to a depletion of energy in the lignotuber resulting in poor resprout vigour, growth, reproduction and even mortality (Canadell and Lopez-Soria, 1998).

The analysis of average canopy sizes shows that out of the populations which burnt in 2016, the mature plants at BK 1 were smaller in the 6-20 stem class and the >41 range, but that this difference was insignificant. Only the 1-year post fire population at TW had significantly

smaller canopy sizes for the 21-40 stem class compared to the other populations and this may have been expected since it had less recovery time. However, this same population had notably larger canopy sizes than the older sites BK 1 and BK 2 for the >41 stem class. The variability in regrowth observed was certainly influenced by the disturbance history of the site, but other site characteristics such as slope aspect and moisture interact in complex ways to influence morphology. For example, site BK 2 which is located approximately 700 m from BK 1 and is unharvested for 20 years appears to be an ideal control site to test the influence of harvesting. However, the site is south east facing and soil analysis shows that it has higher organic matter and moisture than BK 1 (McGregor, unpublished PhD data, 2020). These two factors are known to be beneficial for plant growth and health (Jangir *et al.*, 2019) and may therefore partly account for why these plants are larger. In addition, the naturally stout feature of the population at BK 1 may have been accentuated by two consecutive disturbances resulting in even more of a difference between the populations.

Whilst this study did not aim to determine if harvested populations differ from unharvested, previous research suggests that on a regional scale C. intermedia populations which are harvested do differ from unharvested populations, specifically harvested have more stems than unharvested populations (Barnardo, 2013). In theory more stems should be beneficial as they accommodate more reproductive structures. The potential concern is that recovery from harvesting is energy intensive and the stored carbohydrates in the lignotuber that would have been used for production of flowers are instead used for vegetative recovery. On the other hand, honeybush plants have been anecdotally observed to flower prolifically in the second and third years following a disturbance, including harvesting (McGregor, 2017). However, it must be mentioned that experienced honeybush farmers have observed the plants to grow and flower more vigorously after a fire when compared to harvesting (likely due to fertilization effect) (McGregor, 2017). After approximately the third year, flowering and consequentially seed set rapidly declines, the potential implication of this from a management perspective is that populations which are not subjected to disturbances will have less potential to replenish soil seedbanks and recruit new seedlings. Given that site conditions may influence plant morphology in unique ways for each population, human pressures such as harvesting could be studied by monitoring and comparing harvested plants with unharvested plants within the same populations. This would reduce the effect of site conditions. Overall, the Kouga populations studied showed typical growth features that have been observed for resprouters in general and for *C. intermedia*.

If augmentation is to be used as a tool for expanding wild populations, then seed must be collected directly from parent populations to preserve genetic integrity as discussed. This constraint requires that we understand what aspects may affect germination success. This study used two different collection methods which are both acceptable for collecting wild *C. intermedia* seed and tested their effects over germination. In addition, because seed dimorphism is known to influence germination (Koen *et al.*, 2017), the study tested if there was a difference between green and brown seed germination levels. Whilst the results are not presented as conclusive, the trial does hold some potentially insightful findings for future research or augmentation projects. The results must be interpreted within the context of factors which both naturally and artificially influence germinability. Only some of these factors which are relevant to the study will be mentioned.

The large differences observed with regard to germination success of seeds collected from different farms may have been due to the prevailing conditions during the time of pod development (Motsa *et al.*, 2017). During the summer of 2020 the entire region was experiencing hotter and dryer than usual conditions which translates to less resources being available for the plants, affecting flower and seed production. Whilst it is known that resource availability affects reproduction, there is very limited understanding to what extent *Cyclopia* respond to this. Even under similar conditions, there are natural fluctuations present in seed germination success from year-to-year due other factors such as genetics (Slabbert *et al.*, 2019). Seeds sourced at farm 1 are from populations which grow in a more arid climate than the other farms and therefor may have been less susceptible to dry conditions, whilst this may explain to some extent the success of farm 1, when looking at farm 2 which also has a substantially drier climate than farms 3 and 4, the germination success was the lowest.

Some of the artificial factors which may have influenced the results are choice of scarification method, water content of soil and choice of medium. Considering the above, ascribing possible reasons for why there was such a large difference between germination success both between and within farms is challenging. Overall seed colour and collection method only influenced germination significantly in two instances. The observed large discrepancy between some of the results such as farm 1 and farm 2 for example, suggest that location of seed collection is more of a determining factor for germination success.

Green seed had higher germination than brown seed in every instance other than for farm 1 netted brown seed and farm 2 picked brown seed, both of which were only marginally more. This result may reflect that the scarification method used is more effective on green seed which have harder seed coats. In terms of maximum germination, the brown seed netted sample from farm 1 had the best rate of 94 %, this result is interesting in that other research with seed of similar age (approximately 1 year) attained maximum germination rates of 77% and 88% for a reseeding species (*C. subternata*) which are thought to have higher seed germination rates than resprouting species such as *C. intermedia* (Du Toit and Campbell, 1999; Mbangcolo, 2008; Koen *et al.*, 2017). Overall germination success was very high compared to what is observed under wild conditions. For example, in this study seed germination for the field trials reached a maximum of 23%, and in other research 16% (Beyers, 2016).

This suggests that given favourable conditions, *C. intermedia* has high seed germinability but under more natural conditions this decreases drastically, just as seedling survival does too.

Understanding germination success is important for augmentation projects since it provides a proportional estimate of seeds that are likely to germinate it will aid in nursery management by knowing how many extra seeds to plant in order to hedge against the ones which don't germinate to produce the desired number of seedlings. The latter point may be particularly important since nursery space is valuable and planting two seeds into each tray instead of one could be a critical decision if germination levels are poor. In view of the sheer variability from the results it is also recommended that a preliminary germination trial be carried out to understand what kind of germination levels are to be expected. In terms of collection method, there are advantages and disadvantages to both which need to be considered. Nets can be purchased as cheap or expensive material and therefore the costs are contextual, however, they do require more work since each plant needs to be individually wrapped at the right time and then revisited and carefully removed to prevent losing the seed. The advantage of this method as already mentioned is that in theory the seeds will be of optimally ripe condition.

An interesting point to make is that seeds collected by net had a higher green to brown ratio than those picked by hand (pers. obs. 2021). Picking seed pods by contrast may be less effort given there is a high abundance of fruiting plants, less seeds are also likely to be lost using this method. Some of the main disadvantages are picking unripe pods, seed predation and choosing the correct time to pick. Due to the variation in timing for pod development within the same population some pods will mature at later stages and therefore the population would need to be revisited for picking at a later stage. A final note is that seed germination tempo in this study seemed to reflect a slower rate than what has been observed for *C. intermedia*, 38 days after planting approximately only a third of the total seeds which would end up germinating had done so. This result compared to Schutte and Van Wyk, (1995) which demonstrated a germination percentage of almost 90% after 40 days.

#### 5.6. Conclusions

Recruitment levels and demographic structure of wild C. intermedia populations may be negatively affected by harvesting pressure and the extent of this impact will rise as more demand is placed on these populations. The natural recruitment levels from 5 populations surveyed in this study suggest good seedling to parent recruitment for 1 harvested population. Only 2 young plants were found for another harvested site and 1 at an unharvested. One of the sites was burnt 1 year prior and the other 2 sites 4 years prior to surveying which is an important mechanism in seed germination and this should be kept in mind when assessing the demographics of a population. Considering the high levels of success observed in the germination trials it would be expected that more seedlings would be present. Thus, seed germination success and post germination survival appear to be low naturally. In light of this, using seeds and seedlings to boost population numbers to accommodate increased harvesting may be a viable solution. Variability in growth characteristics is influenced by a combination of site conditions and disturbance history. Overall features were similar to those that have been observed for the species and resprouters in general. Plants increase in size rapidly after a fire and then growth appears to slow. Comparing harvested and unharvested plants within the same sites (preferably very near) would reduce the impact of site conditions on plant morphology. Similarly mature plants should be further categorized into stem count classes to ensure that age discrepancies are not too large.

Further germination studies specifically focused on wild harvested seed from honeybush populations are needed. Until this is accomplished only preliminary remarks can be made about the variability observed in germination success. Green seeds germinated more successfully the majority of the time although overall this was only significant in one instance. Similarly, the net collected seeds germinated slightly better than the picked seed but was only significantly better in one instance. The large difference between farms suggests that source location plays more of a role. As a personal observation, there were less damaged seeds collected in the net compared to picking the pods. On the same note more green seeds were collected inside the nets, suggesting that a large proportion of naturally mature seeds ejected from the pods are in fact greener. Collection method choice is entirely contextual, and the only recommendations that can be made are to ensure that pods are mature enough.

If a site is hard to access and may not be visited regularly then collection by net is recommended to avoid the possibility of losing the seed from the pods opening naturally. In terms of nursery planning and augmentation, a seed germination trial is recommended to establish average rates and experimentation with alternative scarification methods that have been used and are accepted is encouraged.

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# Chapter 6: Synthesis and Preliminary Recommendations for the Augmentation of Wild *C. intermedia*

#### 6.1. Synthesis

This thesis investigated some of the pertinent aspects related to the use of augmentation as a means to increase wild populations of the honeybush species C. intermedia. Chapter 2 consisted of identifying research gaps and reviewing literature surrounding the main topics of the study. Research gaps relevant to this study were ideal habitat conditions for growing C. intermedia, the main economic components and field trials testing the potential of augmentation and natural population characteristics and seed germination trials. It was outlined in this chapter that wild C. intermedia resources are currently what the industry relies upon the most, hence, in the face of impending industry growth, these wild population may be put under more pressure than what is ecologically tenable. Increasing cultivation of other species and the development of harvesting guidelines have contributed positively to reducing this pressure. However, for many landowners there are barriers to cultivating the species such as lack of suitable land and slow growth. Abandoning or reducing wild harvesting may also be unviable since landowners and harvesters may depend largely or even solely on this source of income. A possible solution similar to that employed in the wildflower industry is to augment these populations by planting seeds and seedlings directly into wild fynbos. Chapter 2 argued that although augmentation has been used as tool for conservation, in the context of expanding an indigenous crop for commercial purposes there are risks involved and therefore an agroecological approach should be taken. Principles such as the preservation of genetic integrity, biodiversity and ecosystem processes have all been acknowledged as vital components for agriculture in fynbos systems. In the realm of wild honeybush augmentation this would translate into using seeds sourced only from parent populations, knowing and protecting vegetation communities or sensitive species that share the same preferred ecological conditions and avoiding high disturbance techniques such as ploughing to prepare wild fynbos.

In chapter 3 a desktop GIS analysis using environmental variables characterizing wild C. intermedia habitats and a multi criteria selection (MCS) process was applied to identify broad scale distribution of the population. The same process was applied at a larger farm scale in the Kouga to identify areas of optimal growth and study sites. The output was compared with known wild honeybush occurrence to test 'model' accuracy and showed a satisfactory overlap indicating that the method can be used to identify areas for augmentation on farms, although ground truthing should be undertaken to verify site conditions. The vegetation and soil sampling results from chapter 3 confirm small scale habitat heterogeneity, typical of fynbos. Areas close to wild populations of *C. intermedia* that have the same general topographic conditions do not show a consistent pattern of being different with regards to vegetation and soil conditions, other than having slightly lower species richness. The high number of species found at all sites was expected as a normal trait of fynbos though this may have been compounded by the south facing aspect, and communities differed considerably between plots. Overall, plots at each site contained species of the Proteaceae and Ericaceae families that have a protected status. The implication is that communities which share habitat preferences with C. intermedia are vulnerable to changes and should be surveyed before and after an augmentation trial is done to monitor impacts. On the same note these co-occurring species may be benefiting C. intermedia in ways that are unknown such as supporting microbial communities or maintaining other ecological processes and should therefore be preserved as far as possible.

Post fire age of the soil sample impacted average nutrient levels more often than the presence or absence of honeybush. Veld aged 4 years had significantly lower levels of P, K and Ca than 7-month-old veld. The newly burnt site showed significant increases for the nutrients P, K and Ca from before to after burning. The magnitude and timing of these changes were significantly greater for the absent plot in most instances, most notably P levels (mg) were almost double that of the present plot at the time of the final sample. The controlled burn conducted for this study was small (<1 ha) and site conditions during winter at the time of burning (cool and moist) were conducive for a low intensity fire. The drastic increase shown for some of the nutrients may also be an indicator of fire intensity because there does not seem to be a loss due to volatilization which happens at higher temperatures.

Bearing in mind the complex effects of fire, further research is needed on using fire for veld preparation before it can be put forward as a recommended practice.

In chapter 4 augmentation field trials and a cost benefit analysis (CBA) were used to assess the survival of supplemented seeds and seedlings as well as the main economic inputs and outputs. Due to the amount of veld which shares similar conditions as those where wild C. intermedia is already growing, the potential to augment these areas was tested by setting up plots on the same slopes which were not located in wild populations. On the same note, due to C. intermedia being ecologically dependent on fire the influence of this was tested by planting on different post fire aged veld. After a year of monitoring, the results for farm 1 showed that absent plots had only slightly higher seedling survival for 2 of the sites and for the third site it had significantly better survival. Similarly seed germination was better in the absent plots for 2 sites. The influence of site age was insignificant over both seedling growth and survival but overall, the newly burnt site had the best seedling growth. In the two older sites, absent plots had significantly bigger seedlings. Seed germination appeared to be considerably better in the newly burnt site and germinated seed survival was insignificantly different between all plots except in one instance at the present plot at the 7-month-old site. A variety of factors may have influenced results and therefore it cannot be conclusively stated that areas adjacent to wild populations are better suited than areas within wild populations only that they appear to meet the ecological requirements for survival and can be considered candidate areas for augmentation. The seedling survival results from the farm 1 (Kouga farm) were overall better than the other farms except in one instance. This is unexpected as the Kouga farm has a considerably drier climate. Farm 1 received lower than usual rainfall and therefore the results may be different in years with better rain. Due to the variability in the results extrapolating future success rates based on these findings is not advisable.

The CBA results favoured the use of seeds compared with growing or buying seedlings and planting them out mainly due to the high input costs associated with seedling production. The cost of setting up a basic nursery was used as an example of what a honeybush landowner may opt to use. This initial expense is high and compounded by the seedling planting labour makes this option costly.

Despite the augmentation trials demonstrating that survival is substantially better with seedlings, the required survival rates to obtain a worthwhile return on investment after a discounting rate was applied was not achieved in most cases.

Chapter 5 dealt with the characteristics of 5 wild populations on farm 1, specifically focusing on recruitment rates and the potential impacts of harvesting on growth. In conjunction seeds were collected from wild populations on 4 farms spread across the study region using two alternative collection methods namely picking by hand and netting. The seeds from each collection method were then sorted into green and brown and a germination test was run.

Only one population had seedlings at the time of surveying and a small proportion of young plants were found for 2 other sites, indicating that recruitment had taken place over the last few years. Importantly, populations are recruiting even when subject to harvesting and young plant to mature plant ratios for one unharvested site was only marginally higher. The scale of the data collected is too small to draw solid conclusions, considering that recruitment is a complex process in fynbos wider scale studies are needed. In terms of growth characteristics, average stem numbers for the unharvested populations were higher and canopy sizes were larger. This difference however was insignificant. In terms of the germination results, when compared within farms picked seed germinated significantly better that netted seed for farm 2, similarly green had a significantly better result than brown seed for farm 3. Overall seed germination rates were most different compared between farms suggesting that location played a bigger role in the results.

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## 6.2. Preliminary Recommendations

In conclusion a set of preliminary recommendations have been drawn up and presented as a 'flyer'. This format is intended to be used as a convenient way of distributing information and informing landowners about the aspects of augmentation learned from this research.

## Honeybush (Cyclopia Intermedia) Augmentation Recommendations

## Site Choice



- Areas near or containing wild honeybush on south to west facing slopes are both suitable.
- Soil conditions change with veld age and newly burnt veld shows an increase for some important nutrients such as phosphorus.
- Other fynbos species share the same habitat and may benefit honeybush growth, for this reason augmented areas should be monitored.

## Choice of Seeds vs Seedlings

- The survival rates from seeds which germinated were very low; between 0 and 3 seeds out of every 64 planted.
- Augmenting with seeds can be done cheaply and projects still show good economic returns under low survival.
- Seedlings show higher survival rates; between 4 and 10 plants out of every 24 planted.
- The disadvantage is that they are far more expensive and require high survival rates for good economic returns.

## Further Recommendations and Considerations

- It is vital to protect the genetic integrity of honeybush; only use seeds which are sourced from the same populations in the areas which are being augmented.
- Test augmentation site suitability with seeds first before using seedlings.
- Germination levels appear to fluctuate depending on where the seeds were collected, it is advisable to run a simple germination test for your area.
- Lastly it is recommended that populations which are subject to harvesting pressure be monitored.



# 7. Appendix:

# 7.1. Multiple criteria data sets

# Table 7-1: GIS data sets used during the MCS process for habitat distribution, optimal growth areas and study site selection

Variable	Source	Usage	Scale of data
<i>C. intermedia</i> population locations (across range)	G.K McGregor	Define the area within which environmental parameters are used to predict occurrence	1: 50 000
<i>C. intermedia</i> population locations (Melmont farm)	G.K McGregor and Q. Nortjé (Expert map)	Determine what percentage of the area the optimal site model identified within honeybush bearing land	1: 10000
Aspect and Slope	SRTM 1sec DEM NASA EOSDIS Land Processes DAAC. (2013). NASA Shuttle Radar Topography Mission Global 1 arc second. Available: https://doi.org/10.5067/MEaSUREs/SRTM / SRTMGL1.003. Accessed June 2020.	Environmental parameter	30m res
Landcover, Elevation, Rainfall and Vegetation type	NLC 2018, DEA Department of Environmental Affairs (DEA). (2018). SA National Land-Cover. DEA, Pretoria. Available: https://www.environment.gov.za/projects programmes/egis_landcover_datasets Date accessed June 2020.	Environmental parameter	20m res
MODIS fire data	University of Maryland	Determine areas which burnt over the past 5 years for the Kouga farm	

## 7.2. Vegetation Survey Species List

Table 7-2: Full species list for the Kouga farm vegetation surveys (Chapter 3). Status; EC means protected by Eastern Cape Environmental Conservation Bill as introduced 23 November 2003 and D means declining status as given by The Red List of South African Plants (2017)

SteilKop Site (1) IN			
Family	Genus	Species	Status
Asteraceae	Gerbera	piloselloides	EC
Asteraceae	Vernonia	sp. 2	
Asteraceae	Selago	dolosa	
Bruniaceae	Brunia	noduliflora	
Commelinaceae	Cyanotis	speciosa	
Cyperaceae	Ficinia	gracilis	
Ericaceae	Erica	demissa	EC
Euphorbiaceae	Clutia	laxa	
Euphorbiaceae	Clutia	alaternoides	
Fabaceae	Argyrolobium	sp.1	
Fabaceae	Cyclopia	intermedia	D, EC
Fabaceae	Rafnia	racemosa	
Fabaceae	?	?	
Gentiaceae	Chironia	baccifera	
Geraniaceae	Pelargonium	candicans	
Iridaceae	Bobartia	orientalis	
Iridaceae	Watsonia	fourcadei	EC
Malvaceae	Hermannia	sp.1	
Malvaceae	Hermannia	sp.2	

Poaceae	Diheteropogon	filifolius	
Poaceae	Ehrharta	ramosa	
Poaceae	Themeda	triandra	
Proteaceae	Leucospermum	cuneiforme	EC
Restionaceae	Elegia	vaginulata	
Restionaceae	Restio	triticeus	
Rosaceae	Cliffortia	sp.1	
SteilKop Site (1) OUT			
Family	Genus	Species	
Asteraceae	Disparago	tortilis	
Brassicaceae	Heliophila	elongata	
Bruniaceae	Brunia	noduliflora	
Ericaceae	Erica	pectinifolia	EC
Fabaceae	Otholobium	careua	
Geraniaceae	Pelargonium	unknown	
Hyacinthaceae	Albuca	possibly A. cooperi	
Malvaceae	Hermannia	hyssopifolia	
Oxalidaceae	Oxalis	?	
Poaceae	Pentaschistis	tortuosa	
Poaceae	Tristachya	leucothrix	
Proteaceae	Leucadendron	salignum	EC
Proteaceae	Protea	vogtsiae	EC
Restionaceae	Elegia	vaginulata	
Restionaceae	Ischyrolepis	capensis	
Restionaceae	Ischyrolepis	sp. 1	

Rutaceae	Agathosma	mundtii	EC
Rosaceae	Cliffortia	linearifolia	
Thymalaceae	Gnidea	juniperifolia	
	Thesium	?	
Tweeling Site (2) IN		•	
Family	Genus	Species	
Asteraceae	Euryops	rehmannii	
Asteraceae	Osteospermum	glabrum	
Asteraceae	Vernonia	sp. 1	
Brassicaceae	Heliophila	elongata	
Bruniaceae	Brunia	noduliflora	
Caryophyllaceae	Montinia	caryophyllacea	
Ericaceae	Erica	demissa	EC
Euphorbiaceae	Clutia	laxa	
Fabaceae	Cyclopia	intermedia	D, EC
Geraniaceae	Pelargonium	myrrifolium	
Poaceae	Diheteropogon	filifolius	
Poaceae	Festuca	sp.1	
Proteaceae	Leucadendron	salignum	EC
Proteaceae	Leucospermum	cuneiforme	EC
Proteaceae	Protea	nitida	EC
Restionaceae	Rhodocoma	capensis	
Restionaceae	Willdenowia	glomerata	
Rhamnaceae	Phylica	sp.1	
Rosaceae	Cliffortia	linearifolia	

Rutaceae	Agathosma	capensis	EC
Tweeling Site (2) OUT			
Family	Genus	Species	
Asteraceae	Helichrysum	felinum	
Asteraceae	Stoebe	plumosa	
Ericaceae	Erica	demissa	EC
Fabaceae	Argyrolobium	sp.1	
Geraniaceae	Pelargonium	myrrifolium	
Iridaceae	Bobartia	orientalis	
Myrsinaceae	Myrsine	africana	
Plantaginaceae	Plantago	sp. 1	
Poaceae	Diheteropogon	filifolius	
Proteaceae	Leucadendron	salignum	EC
Proteaceae	Leucospermum	cuneiforme	EC
Proteaceae	Protea	nitida	EC
Ranunculaceae	Knowltonia	vesicatoria	
Restionaceae	Willdenowia	glomerata	
Rhamnaceae	Phylica	sp.1	
Rutaceae	Agathosma	capensis	EC
Rutaceae	Agathosma	mundtii	EC
Rosaceae	Cliffortia	linearifolia	
Witberg Site (3) IN	•		
Family	Genus	Species	
Aclepidaceae	Asclepias	?	
Anacardiaceae	Searsia	sp.1	

Asteraceae	Gerbera	serrata	
Asteraceae	Euryops	munitus	
Asteraceae	Vernonia	sp.3	
Campanulaceae	Wahlenburgia	tenerrima	
Cyperaceae	Ficinia	gracilis	
Euphorbiaceae	Clutia	laxa	
Fabaceae	Cyclopia	intermedia	D, EC
Fabaceae	Indigofera	sp. 1	
Fabaceae	Rafnia	racemosa	
Geraniaceae	Pelargonium	myrrifolium	
Malvaceae	Hermannia	flammula	
Malvaceae	Hermannia	hyssopifolia	
Orchidaceae	Satyrium	pallens	
Poaceae	Themeda	triandra	
Poaceae	Trachypogon	spicatus	
Poaceae	Tristachya	leucothrix	
Poaceae	Eragrostis	superba	
Restionaceae	Ischyrolepis	capensis	
Restionaceae	Rhodocoma	fruticosa	
Rosaceae	Cliffortia	ruscifolia	
Thymalaceae	Gnidea	juniperifolia	
	Lanaria ?	lanata	
Witberg Site (3) OUT			
Family	Genus	Species	
Anacardiaceae	Searsia	sp.1	

Cyperaceae	Ficinia	gracilis	
Ericaceae	Erica	umbelliflora	EC
Euphorbiaceae	Clutia	laxa	
Fabaceae	Indigofera	sp.1	
Fabaceae	Argyrolobium	sp.2	
Fabaceae	Aspalathus	sp.1	
Geraniaceae	Pelargonium	myrrifolium	
Hypoxidaceae	Hypoxis	hemerocallidea	
Iridaceae	Aristea	?	
Malvaceae	Hermannia	flammula	
Malvaceae	Hermannia	hyssopifolia	
Poaceae	Ehrharta	ramosa	
Poaceae	Eragrostis	superba	
Poaceae	Themeda	triandra	
Poaceae	Trachypogon	spicatus	
Poaceae	Tristachya	leucothrix	
Poaceae	Pentaschistis	pallida	
Proteaceae	Leucadendron	salignum	EC
Restionaceae	Rhodocoma	fruticosa	
Restionaceae	?	sp.1	
Rhamnaceae	Phylica	cf. axillaris	
Thymalaceae	Gnidea	juniperifolia	

## 7.3. Information Sources used in CBA

Table 7-3: Summary of the informants and the information they provided to derive the approximate costs and benefits used in the CBA

Source	Cost	Benefit
M. Sephton	Nursery items	
(Living Lands)	<ul> <li>Seed planting time/ha</li> </ul>	
	Large scale augmentation details	
Q. Nortjé	Seed collection	
G.K. McGregor	Harvesting loads per day	• Wet material price
		• Yield per plant

## 7.4. Nursery Layout

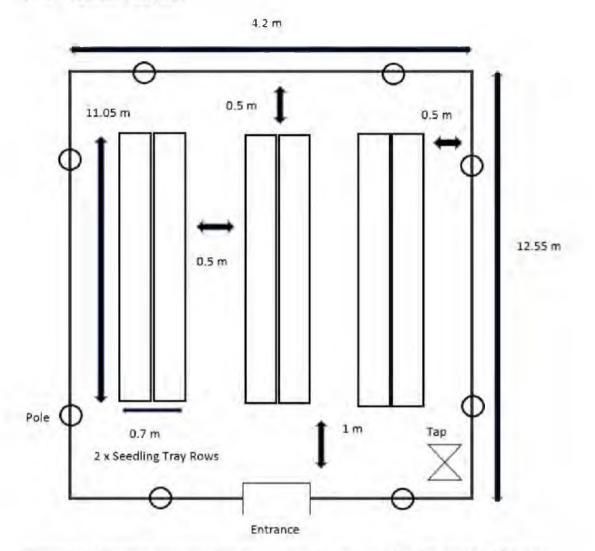


Figure 7-1: Nursery construction layout and items used for the study which can produce approximately 10000 seedlings per annum.

Table 7-4: Construction items and costing for a basic nursery setup

Total area: 4.2 x 12.55 = 51.45 m <sup>2</sup> (top coverage)
Tray dimensions for Unigrow 98 plug tray: 350 mm x 650 mm (0.24 m²)
98 x 102 trays = 9996 seedling plugs
9996 x 0.24 m <sup>2</sup> = ~ 24 m <sup>2</sup> (growing area)
Shade cloth = ~ 1x Roll (25 m x 3 m) R 1700 (Wholesale)
102 trays x R91.50 = R 9333 (DelaPlast wholesale)
Transport and building fees = ~ R1000 (Own vehicle and staff)
Poles (2.4 m) (x 8) = R 640 (Builders/BKB)
Tap, wire, nylon and entrance = ~ R 500 (Online ordering and BKB etc.)
Cement/ brick raised grow stands x 3 = R 1200 (Wholesale)

# 7.5. Cost Benefit Analysis Spreadsheet

А	B	C	D	E	F	G	н	1	J	ĸ	L	м	N	0	p	Q	R	5	т	U
Project: C. Intermedia Augmentation												1						1		-
Option: Seedlings (2A)																				
																			Crop yield-	
																			harvesting	
		-																Total	costs	B:C rat
Cost (Rands)	Year 0	Year 1	Year 2	Year 3	Year 4	Year 5	Year 6	Year 7	Year B	Year 9	Year 10	Year 11	Year 12	Year 13	Vear 14	Year 15	Year 16			
Construction+ Seedling production																				
	32510					-				-						0	0	0 32510		
eedling planting	11250									-						0		0 11250		
larvsting costs	0											0 0	5760	0		0	0 S76	0 23040	)	
ncreased yield	0	0	0	(	48000	0	0	•	48000	0	0	) (	48000	0		0	0 4800	0 192000	)	
let (benefits - costs)	-43760		0	(	42240					0	C	0 0	42240	0		0	0 4224	0 125200	)	
IPV @ 5%	-43760		~	(	34750.9525				28589.6947	0	C	) (	23520.8125	0		0	0 19350.6	3 62452	10621:	2
IPV @ 8%	-43760	0	0	(	31047.661	0	0	1	22820.9577	0	0		16774.0852	0		0	0 12329.4	5 39212	8297.	2
								4	8											
	Plants (10000)	Harvest	Vield (0.4	Price (R15	4 harvest	Harvest	4 harvest	Actual crop	Initial	NPV	B:C (A/B)	NPV @ 5%	B:C (5%)	NPV	B:C (8%)					
		Guideline	kg/plant)	wet/kg)	cycles (crop	expense	cycles	value (wet	Expenses					8%						
		180% 01					(harvesting	material-harves	t											
		plants)					costs)	expense)												
iurvival (%)																				
						_														
0.05			400	2400							011	-38449	0.12	-39611	0.0	29				
0.07			44.4	3360							- d. 1	- 36325	0.17	-37952	0.1	13				
0.1			~LV	4800	19200	576			43760	-26864	0.39	-33139	0.24	-35463	0.1	19				
0.12			384	5760	23040	691.2	2764.8	20275.2	43760	-23484.8	0.46	-31015	0.29	-33803	0.3	23				
0.2		1600	640	9600	38400	1152	4508	3379	43760	-9968	0.77	-22518	0.49	-27166	0.1	38				
0.25	2500	2000	800	12000	48000	1440	5760	42244	43760	-1520	0.97	-17207	0.6	-23017	0.4	18				
0.35	3500	2800	1120	16800	67200	2016	8064	5913	43760	15376	1.35	-6586	0.84	-14720	0.0	56				
0.5	5000	4000					11520	8448	43760	40720										
0.6	5000	4800	1920				13824	10137	43760	57616										
	7500	6000				-	17280	12672	43760	82960										
0.75																				
0.9		7200				-	20736	15206-	43760	108304			2.18	30915	1.3	76				

Table 7-5: CBA calculation for seedling option 2A as an example. Link to spreadsheet: https://docs.google.com/spreadsheets/d/1pvKbFTPR\_Ci-

b1CA9zavJbYDoiMncRo-/edit?usp=sharing&ouid=113787606297576584185&rtpof=true&sd=true