Population Demography of the Fragrant Prickly-Apple Cactus (*Harrisia fragrans*)

Contract # 012036
Final Report

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Introduction

Background

The fragrant prickly-apple cactus, *Harrisia fragrans* Small ex Britton & Rose, is a shrubby cactus endemic to Florida, which is listed as Endangered by the U.S. Fish and Wildlife Service and the State of Florida. Up until recently, all known extant occurrences of *H. fragrans* were in southeastern St. Lucie County (Rae 1995, FNAI 1997). Isolated populations have recently been discovered in Indian River and Volusia counties (Woodmansee et al. 2007), but St. Lucie County has the largest, most contiguous population. The Institute for Regional Conservation (IRC) reported *H. fragrans* as being frequent at Savannas Preserve State Park (SPSP; Bradley et al. 1999), based on surveys conducted between 1998 and 2002.

Two demographic studies of *H. fragrans* were previously conducted. The first was by Rae and Ebert (2002) who studied the population dynamics of two subpopulations in and around SPSP between 1988 and 1996. Their results showed that both subpopulations suffered serious decline (55.3% - 59.8%). They predicted the decline and eventual extinction of *H. fragrans*. The second study was conducted by The Institute for Regional Conservation (IRC) between 1999 and 2002 (Bradley et al. 2002). It entailed monitoring the entire SPSP *H. fragrans* population, including Rae and Ebert's (2002) subpopulations. The population was found to be stable, contradicting Rae's previous findings.

Bradley et al. (2002) also discovered that approximately 10% of *H. fragrans* plants died each year, while another 10% recruited. Because of the high, yearly turnover rate of plants, a negative change in any ecological process that affects mortality or recruitment would result in a substantial decline of *H. fragrans*.

The goals of this project were to continue monitoring the population status of *H. fragrans* in the SPSP, to conduct more thorough investigations of the processes that influence mortality and recruitment, collect more precise data on early seed germination and mortality rates, determine whether *H. fragrans* forms a persistent seed bank, evaluate seed germination in sand pine (*Pinus clausa*) dominated habitats, and model long-term population trends using population viability analyses. An additional goal of the project was to collect historical data on habitat types at the SPSP to better understand the current habitat for *H. fragrans* and how it may have changed in the past due to farming activities and other events.

Study Species

H. fragrans is a columnar cactus with multiple stems reaching 4 m. Stems may be 5 cm in diameter and are armed with gray spines that are 2-4 cm long. Branches arise from both the base of the plant and laterally from other stems. White to pink fragrant flowers that are ca. 20 cm long, open at night and are presumably moth pollinated (Ted Fleming, pers. comm.). The red fruits are globose and 5-6 cm in diameter and can contain ca. 1,400 seeds (Rae 1995). Few interactions with animals have been observed. A scale insect, *Diaspis echinocati* (Diaspididae) is frequently observed on plants. Birds, gopher tortoises, and raccoons may play a role in dispersal, although this has not been observed.

Study Site

H. fragrans at the SPSP is presently known from a 13 km by 0.5 km section of the Atlantic Coastal Ridge between the cities of Ft. Pierce and Jensen Beach, and between the Indian River and a large swale known as the Savannas (Figure 1). Most of the undeveloped area in this region is now located within the

SPSP. This section of the Atlantic Coastal Ridge is transected by the Florida East Coast (FEC) Railway from north to south. The SPSP, and the bulk of the *H. fragrans* population, is on the west side of these tracks. To the east of the tracks, extensive development has occurred and little is in public ownership. A smaller number of *H. fragrans* occur on private lands east of the tracks. In a survey of 52 properties, only 62 plants were found (Woodmansee et al. 2007). *H. fragrans* at the SPSP is found on St. Lucie sand with 0-8 percent slopes, at elevations between approximately 8 and12 meters (Watts & Stankey 1980). The area receives 140 cm rainfall/year, with about 62% occurring from June to October (Watts & Stankey 1980).

H. fragrans at the SPSP grows in formerly disturbed scrub. The most common habitat where the species occurs are areas dominated by fields of *Aristida gyrans* and *Polygonella robusta*, open sand, isolated or clumped *Sabal palmetto* stands associated with vines (especially *Smilax auriculata*) and stands of *Quercus* spp. and *Carya floridana*. *H. fragrans* most commonly grows along these edges of the *S. palmetto* clumps. *H. fragrans* rarely occupies areas of *Pinus clausa* scrub. In some areas *H. fragrans* grows in the interior of larger, xeric hammock communities. In addition, one large colony of *H. fragrans* occurs in an area dominated by the exotic coniferous tree *Callitris glaucophylla*. This tree forms a dense canopy at the southernmost known *H. fragrans* colony. *H. fragrans* occurs in an atypically dense population beneath the canopy of this tree.

Methods

Demographic Monitoring

In December 2003 and January 2004, three colonies of *H. fragrans* were selected for sampling (= Job 1 in contract). The area of each colony was selected to include approximately 300 plants that were alive at the end of the final sampling in 2002 (Figure 1). These colonies contained plants that had been tagged and monitored by IRC annually as early as 1999. Each subpopulation was visited three times each year[1], once in the spring, summer, and winter, starting at the end of 2003 (winter). Winter sampling was conducted to measure existing plants (total stem length and number of stems), monitor reproductive output (number of fruits and flowers), record mortalities, and search for new recruits (either seedlings or fallen stems). Spring and summer sampling focused on monitoring reproductive output by recording fruit and flower production. Sources of mortality were noted whenever possible. In addition to the plants in these three selected subpopulations, a small number of plants that IRC had previously identified as being monitored by John Rae starting in 1988 were also monitored, even when they occurred outside of the three subpopulations.

Repeated Measures Analysis of Variance was used to analyze changes in stem length and flower in fruit production. These analyses were conducted using EZAnalyze 3.0. Post-hoc tests were done with Wilcoxon Signed Ranks Tests using SPSS 13.0. A population viability analysis (PVA) will be conducted, based on demographic monitoring and seed germination data (discussed below) using Ramas Metapopulation software. Because seed germination data will not be available until 2008, the PVA will be submitted in 2008.

[1] Monitoring was not conducted in 2004 because of contract changes.

Figure 1: *H. fragrans* study colonies at the SPSP

<u>Seed Germination</u>

To quantify seed germination and early seedling survival, seed germination experiments were conducted (= Job 2). Each winter 36 seed boxes were installed. Each seed box was a wooden frame (25 cm X 25 cm X 4 cm) covered with metal hardware cloth for protection. These were placed in four sampling locations, including areas within each of the three subpopulations discussed above, and additionally in a habitat that was dominated by sand pine (*Pinus clausa*) (=Job 5). At each of the four locations three sets of three seed boxes were installed in each of three treatments - one in full sunlight, one in partial sunlight, and one in full shade. By the end of the study 108 seed boxes had been established, nine sets at each of the four locations. When each seed box was installed, 50 *H. fragrans* seeds were spread over the soil surface and lightly covered with sand or leaf litter. At each subsequent sampling period (spring, summer, and winter), each seed box was monitored for seed germination and the results were recorded. Seedlings were marked when possible, or their location was recorded in the seed box so that its status could be determined at the next sampling (alive or dead). Differences for seed germination rates were tested using Chi Square test in SPSS.

Seed Bank

In order to determine whether *H. fragrans* forms a persistent seed bank, a seed dormancy experiment was conducted (=Job 3). In winter 2005, seed bags were buried in each of the three subpopulations, in each of three microhabitats (full sun, partial sunlight, and full shade). At each of these nine sites, nine seed bags, each containing 50 seeds, were buried, resulting in a total of 450 buried seeds. At each subsequent sampling session (spring, summer, and winter) one seed bag was retrieved from each of the nine sites. These seeds were then provided to Fairchild Tropical Botanic Garden for germination trials.

Habitat History

To assess the history of the Atlantic Coastal Ridge at and adjacent to the SPSP and factors influencing current habitat types, a variety of data sources were consulted (=Job 4). These sources included:

- historical photographs
- local accounts by St. Lucie county residents
- journal articles; books, including those with pictorial accounts of the history of the area
- published agricultural and early settler's logs, letters, and articles
- information from organizations with a focused interest on understanding the history of this area, such as the St. Lucie County Historical Society
- aerial photographs from 1943 and 1944

Aerial photographs were obtained from the State of Florida, Publication of Archival Library and Museum Materials and were subsequently georeferenced using ArcGIS.

Results

Demographic Monitoring

Over the four year period of the project, 1,542 plants were monitored: 631 in Colony A, 354 in Colony B, 540 in Colony C, and 14 outside of the colonies that were formerly monitored by John Rae. In winter 2003 there were a total of 1,094 plants across the three subpopulations, and in winter 2007 there were 739 plants (Table 1, Figure 2). In winter 2007 there were only 67.6% as many plants as at the beginning of the study. The majority of this decrease occurred between the 2003 and 2005 samplings and appears to be due to the impacts of Hurricanes Jeanne and Francis, which passed over the SPSP in 2004. Colony A, in which H. fragrans occurs under the exotic tree *Callitris glaucophylla*, showed the largest decrease, from 470 plants in 2003 to 232 (49.4%) in 2007. Colony B was at a 67.0% and Colony C was impacted the least ending 2007 with 92.1% of the pre-hurricane population. It should be noted that much of the decrease can be attributed to a large cohort of seedlings (n=195), of which only 54 (27.6%) were still alive by the next sampling session.

	Year			
Colony	2003	2005	2006	2007
A	470	253	228	232
B	270	177	157	181
C	354	222	237	326
Total	**1,094**	**652**	**622**	**739**

Table 1: Numbers of living *H. fragrans* by year

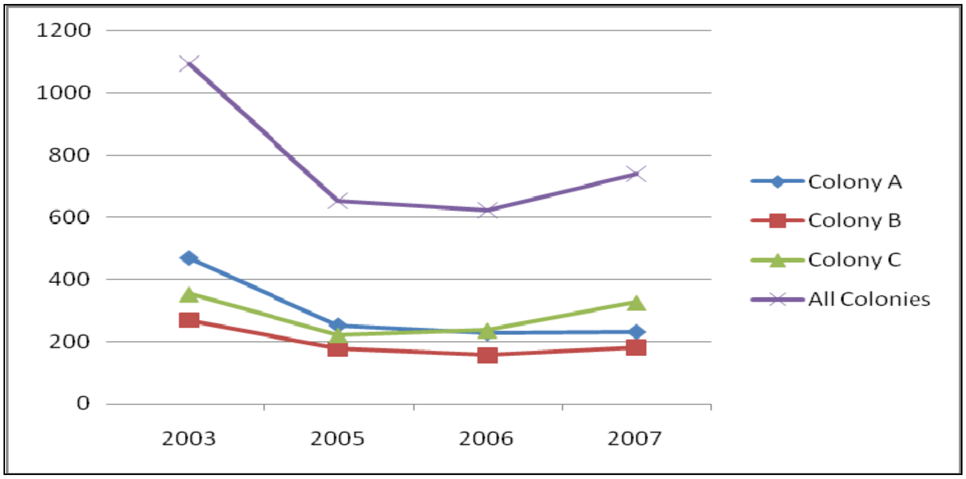

Figure 2: Numbers of living *H. fragrans* by year

For plants that were monitored throughout the study, there was a significant difference in flower production between the three seasons (F=41.319, P<0.001, N=756). Post-hoc tests between seasons showed that there were significant differences between winter and spring (Z= -10.725, P <0.001, N=756), winter and summer (Z= -2.256, P =0.012, N=756), and spring and summer (Z= -9.93, P <0.001, N=756). Flower production was highest in the spring each year and usually lowest in winter (but not in 2006, in which flower production was lowest in summer) (Figure 3). There was also a significant difference in flower production between years (F=6.173, P=0.002, N=756). Post-hoc tests showed that there were significant differences in flower production between 2005 and 2006 (Z= -7.682, P <0.001, N=756) and 2005 and 2007 (Z= -8.589, P <0.001, N=763), but not between 2006 and 2007 (Z= -1.622, P =0.105, N=756.

For plants that were monitored throughout the study, there was a significant difference in fruit production between seasons (F=74.871, P<0.001, N=756). Fruit production peaked each year in summer, and was lowest in spring (Figure 4). Post-hoc tests between seasons showed that there were significant differences between winter and spring (Z= -8.133, P <0.001, N=756), winter and summer (Z= -8.476, P<0.001, N=756), and spring and summer (Z= -12.872, P <0.001, N=756). There was no significant difference between years (F=0.915, P<0.401, N=756).

While total stem length for plants present in 2003 and alive through 2007 showed a trend toward reduction between 2003 and 2005, and an increase to near-2003 levels by 2007 (Figure 5), this observed difference was not significant using repeated measures ANOVA (F=1.66, P=0.174, N=323).

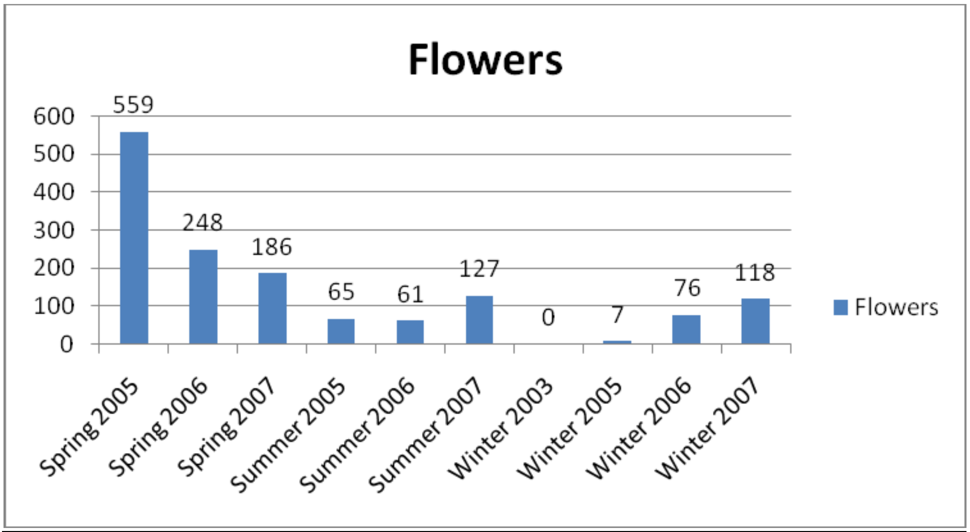

Figure 3: Total number of flowers by sample season

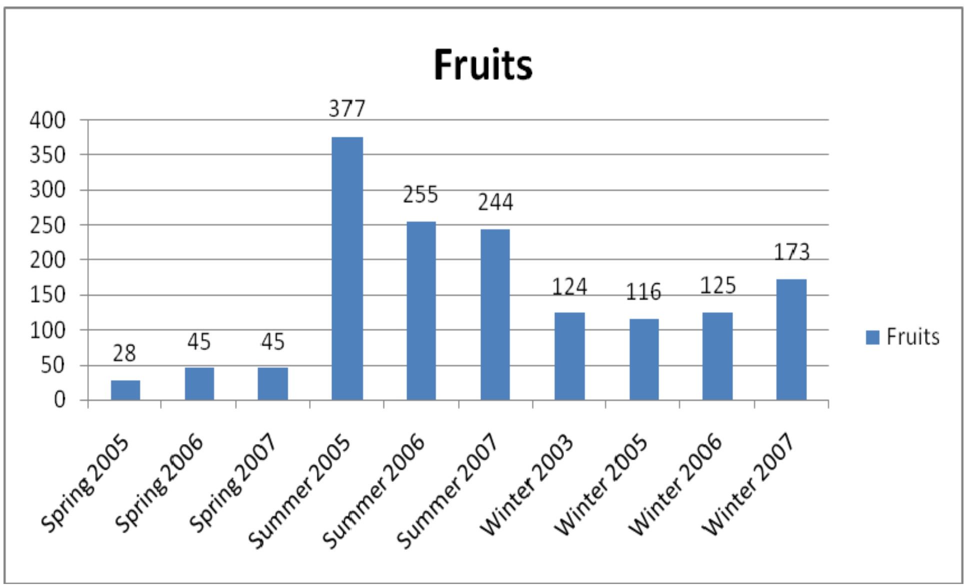

Figure 4: Total number of fruit by sample season

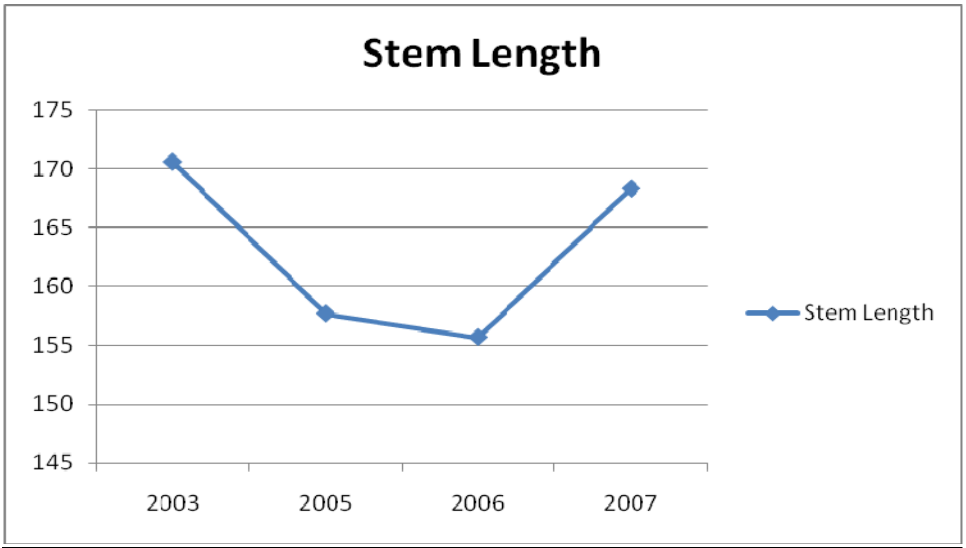

Figure 5: Total stem lengths for plants alive from 2003 to 2007

In 2003, 14 plants that IRC had identified as having been monitored by John Rae between 1988 and 1996 were still alive. These were plants where we could positively associate a plant with one of Rae's tag numbers, because the tag was still in the ground at the base of the plant. Four of these plants were tagged as adults by Rae in 1988, and two of these were still alive in 2007, demonstrating that *H. fragrans* can survive for at least 19 years.

Between 2003 and 2007 an effort was made to identify causes of mortality for *H. fragrans* plants that were found dead. We found that it was generally difficult to determine cause of mortality unless there were extenuating circumstances. Nonetheless, eight types of mortality were identified for mature plants:

- ATV or other vehicle damage – 1 plant
- Buried by sand – 3 plants
- Vandalism (chopped with a machete) – 1 plant
- Herbicide damage – 1 plant
- Over shading (e.g. covered by *Cassytha filiformis,* but not parasitized) – 1 plant
- Feral hogs – 1 plant
- Toppled by hurricane winds – 6 plants
- Killed by treefalls – 12 plants

<u>Seed Germination</u>

By winter 2007, 81 seed boxes had been installed in the three colonies. Between winter 2006 and spring 2007 four of the seed boxes in Colony C that were installed in winter 2005 had been vandalized, including three in full sun and one in partial sunlight, all from the original seed box array established in winter 2005. These boxes have been removed from the study. Results are not yet available for the 27 seed boxes installed in winter 2007. These boxes will be surveyed in spring, summer, and winter 2008. In the remaining 50 seed boxes (representing 2,500 seeds), 44 seedlings were found, a germination rate of 1.76%. Seedlings were found in 9 (18.0%) of the boxes. As many as 14 seedlings were found in a single box (28% of the seeds in the box). Seedlings appeared between 12 months and 24 months, with a mean time to discover of 15 months. Seed boxes in Colony A produced an average of 1.00 seedlings, Colony B 4.67, and Colony C 3.33 seedlings. There was no significant difference between colonies (x^2 = 17.08, p = 0.252). Boxes in full sun produced an average of 0.25 seedlings, in partial sunlight 2.80 seedlings, and in shade 4.83 seedlings. There was no significant difference between treatments (x^2 = 15.167, p = 0.367).

In addition to the seed boxes in the three colonies, 27 seed boxes have been established in *P. clausa* scrub following the same microhabitat treatments. Three sets of boxes were established each winter in 2005, 2006, and 2007. Results are available for the 12 boxes installed in 2005 and 2006. The boxes installed in 2007 will be monitored in the spring, summer, and winter of 2008. As of winter 2007, no seedlings had appeared in any of the previously installed seed boxes.

<u>Seed Bank</u>
Seed dormancy bags were buried at each study area in November 2005 for collection at each monitoring. One bag was collected from each site during each of the six subsequent monitoring sessions and the bags were delivered to Fairchild Tropical Botanical Garden (FTG) for seed germination testing. Since the last seed bags were collected in winter 2007, results will not be available until winter 2008. The preliminary report from FTG is included in *Appendix A*. The results to date indicate that *H. fragrans* is able to form a seed bank that shows little decrease in viability over a 19 month period. Germination rates remained at 64-100% for the duration of the study, with 7 days to first germination and 25 days to maximum germination. There was no consistently significant decline in germination with increased time in the seed bank.

<u>Habitat History</u>

Background on the area
H. fragrans occurs primarily in St. Lucie County at the SPSP, and in order to better understand the history of the habitat of this endangered plant, it is critical to have a general knowledge of the history of St. Lucie County. St. Lucie County was established in 1905, although the area had been previously designated as its own entity under various other names dating back to 1844. This county has had a long history of human activity within its borders, with Native Americans occupying the area for quite some time before Spaniards and Europeans arrived (Rights 1994). Such early white travelers and settlers have also been in the area for some time, with intriguing early accounts of the area even being available from the famed Jonathon Dickinson dating back to 1696. Many other accounts from settlers and early pioneers range from after that time, through the 1800’s and the turn of the century.

The region’s long history of human habitation, although of invaluable cultural significance, does in fact complicate the effort to understand the natural history of the area because it is difficult to understand the myriad ways in which humans have modified the natural landscape over time. To begin with, the Ais Native Americans were known to have lived along the Indian River since at least 400 years ago (Rights 1994, Van Landingham 1988, Williams 1963). In addition, other Native American groups such as the Jeagas and Santa Luceros inhabited adjacent lands (Van Landingham 1988). These Native Americans depended upon the nearby Indian River for much of their food supply, and were known to have gathered wild plants and made use of native wildlife for their livelihoods as well. No records of any uses of native cacti by the Native Americans were uncovered, although clearly much remains to be learned about the ancient ethnobotany of the region. The numbers of the Native American inhabitants declined rapidly with the introduction of European and even Cuban diseases (Williams 1963), wars with new settlers, and finally Native American removal campaigns by the US government. By the early 1700s the Ais Native Americans, the native inhabitants of the region, were thought to have numbered only around 200 (Williams 2003). The newly arrived Creek Native Americans (later referred to as Seminoles) from north of modern-day Florida, Georgia, and Alabama were also in the St. Lucie County area at that time, in much larger numbers than the Ais people, but also declining due to similar reasons (Williams 2003).

From what can be inferred based upon different sources, there remained Native American activity in the area for a long period despite the aforementioned declines of these peoples. In the early 1800s there began the heated Indian Wars, with the First and Second Seminole Wars heavily impacting human habitation and activity in the area. Following the relative conclusion of the Second Seminole War around 1842, white settlers began further encroaching on historical Native American lands. The Act that truly caused the first real permanent settlement by American whites occurred later in 1842: the influential Armed Occupation Act, instituted by the US government. This act helped provide the impetus for the first settlement of the region that is now close to *H. fragrans* habitat. This critical year brought new settlers and pioneers began to truly make their mark on the banks of the Indian River. In addition to increasing settlement in the area, the Armed Occupation Act also inspired land-clearing as it required pioneers to clear and cultivate at least five acres of their claimed lands (Van Landingham 1988).This first settlement of pioneers was known as the Indian River Colony. They were recorded to have vacated the area due to an "Indian scare" in 1849, though some returned in 1850.

Early Explorers, Settlers and Pineapple Plantations

Evidence revealing the original vegetation communities of the Savannas PSP is scarce. Only a few brief descriptions from early settlers and two military expeditions were found. A few photographs were also located that are somewhat helpful.

In 1854 and 1855 the Savannas region was traversed by two military expeditions led by Major Prince and Lieutenant Hill, respectively. Descriptions of the area made by these expeditions are the first and best observations on the original vegetation of the area.

> "The growth on the shore, between the two forts [Fort Capon and Fort Pierce], is pine. South of the latter post the ridge is covered with hammock growth, and, two miles below, rises to the height of forty feet, where is the old Indian Garden….Nine miles south of Indian Garden, there is a high point of the ridge, which is bare and dotted with patches of white sand, ("bleach yard,") and, two miles below, at Mount Elizabeth, pine growth again appears upon the shore; the ridge receding towards the St. Lucie River. Between Mount Elizabeth and Fort Pierce, it borders the river, forming a steep high bank, covered with cabbage, palmetto, and cultivable ground, from which there is, a rapid descent, inland, to an open country, covered with flagponds, savannas, sawgrass, marshes, and palmetto flats, with a few scattered pines." (Ives 1856).

The Indian Garden, two miles south of Fort Piece, is still nearly three miles north of the Savannas PSP. Mount Elizabeth is in Martin County just south of the Savannas. So, two important observations were made. Between Mount Elizabeth and Fort Pierce, in which the length of the Savannas lies, much of the area was dominated by hammock, between the Indian River and the wetland swale now known as the Savannas. The mention of the patches of white sand or "bleach yards" appears to be in the vicinity of

Walton, a community established along the ridge adjacent to the Savannas. No mention is made of pine forests in the vicinity of the Savannas.

An interesting comment was made by a Floridian during the frenzy of building activity that sheds some more light on the status of the natural area before clearing. Wenzel J. Schubert commented “Seventy years ago (1898) I stood on Grandfather Tancre’s front porch at Ankona … and watched a gang of laborers chopping down and grubbing out several fine old trees as they cut the new “county road” right across the middle of our front yard. This narrow, single lane, dirt road that tunneled through the dense river bank hammock growth was Florida’s first East Coast throughway.” This account described the construction of the Indian River Drive, right along the western shore of the Indian River. A few photographs from the Ft. Pierce area and to the north were found that were taken along the west edge of the river. Based on this evidence, it appears that hammocks dominated the eastern edge of the Atlantic Coastal Ridge along the bank of the Indian River.

Beginning around the early 1880s, settlers began having large-scale impacts on the natural habitats of the area as extensive lands were cleared for important cash crops. One of the first and most significant crops to be cultivated in this area was pineapple. Pineapple was first brought to the region by Capt. Thomas E. Richards in 1879 (Van Landingham 1988) and eventually became the most important cash crop, dominating much of the Indian River bank landscape. According to various reports, pineapple plants were thought to number around 2 million within ten years after the first introduction (Williams 2003, St. Lucie Historical Museum website). One description illustrates the extent of the pineapple plantations:

> “This is a country of pineapple plantations. They cover that ridge next to the Indian River, clothing it in prickly green lances from the river banks to the savanna behind it, for miles on miles, running north and south (Packard 1912).”

One photograph was found that shows evidence of sand pines along the Atlantic Coast Ridge in the vicinity of the Savannas. This photograph (Figure 6) shows a group of men clearing vegetation with grub hoes in preparation for a pineapple plantation. A small stand of sand pines is seen in the background.

Figure 6: Workers clearing vegetation, with sand pines in background (Rights 1994)

In addition, other species were known to have been cultivated and established for agricultural purposes, with other key cultivated crops including tomatoes and citrus. Fertilizers were used in the area from early on (Watts and Stankey 1980), thus modifying the soil chemistry, but their use declined drastically with the coming of World War 1 and the associated scarcity of products. It was not discovered through our research exactly what fertilizers were commonly used in the area, but people were known to have used seaweed as well as the scraps from their food (such as fish, turtles, etc.) (Rights 1994).

Additional disturbance came to the area with the construction of the Florida East Coast Railroad in 1894 and the subsequent influx of settlers. It can be clearly understood that the construction of the railroad impacted the area that *H. fragrans* is native to, as even today many plants grow only meters away from the railroad. According to reports (Van Landingham 1988, Rights 1994), railway stations existed in areas noted as St. Lucie, Ft. Pierce, White City, Eldred, Walton, Eden, Jensen, and Stuart.

By the 1920s, pineapple plantations were declining and being closed. Competition from Cuban growers, scarcity of fertilizer, and a pest caused the farming to be unprofitable. Agricultural production, when it did occur later, shifted into growing citrus and tomatoes, but extensive groves or farms were probably not used on the Atlantic Coastal Ridge because of the xeric conditions. Thus, much of the acreage formerly devoted to pineapple plantations was probably abandoned. Some stands of the tree grand eucalyptus (*Eucalyptus grandis*) are present in the Savannas PSP, but it is unknown whether this is due to recruitment from seed or a relic from some type of plantation.

Aerial Photography

Aerial photography of the Savannas was taken by the U.S. Department of Agriculture in 1943 and 1944. This is the first aerial imagery we have seen from this area. The imagery was of course taken over two decades after the pineapple plantations failed, 50 years after the railway was constructed, and some 60 years after the permanent settlers arrived and started to farm pineapples. Still, the photos do show some important features. It appears that some vegetation was never cleared, or was cleared and allowed to regenerate very quickly. It is clear from these forest fragments that hammock vegetation was present along the banks of the Indian River, east of what is now the railway. The hammock succeeded into sand pine scrub to the west, until the ridge sloped down to the Savannas wetland. In one clear example, the hammock extends west about 130 meters from the river at a point where the ridge is about 590 meters wide. Not enough forest fragments appear on the aerial to determine if this vegetation pattern continued the entire length of the ridge and the range of *H. fragrans.* Some areas where *H. fragrans* now grows that are xeric hammock were formerly citrus groves. No clear evidence on the ground indicated this.

Discussion

Demographic Monitoring and Mortality

At the end of this study, there were 32.4% fewer living *H. fragrans* plants than at the beginning of the study in winter 2003. Most of this decline occurred between the winter 2003 sampling and the next sampling in spring 2005, during which time there was a 40.4% reduction in the population. Hurricanes Francis and Jeanne in 2004 can probably be implicated in much of this mortality. Some of the decrease can be attributed to a large cohort of seedlings (n=195), of which only 54 (27.6%) were still alive by the next sampling, and this appears to be within the normal mortality range for seedlings. While high seedling mortality is expected, the 2004 hurricane season caused greater than normal mortality to adults.

Removing the seedling cohort from the 2003 data, there was still a 33.5% decline in mature plants. Bradley et al. (2002) found an average annual mortality rate of about 10%, so the 2004 hurricane season more than tripled expected mortality rates. This is also much higher than the mortality rate of 5% reported by Rae (1995). By winter 2007 the population had started to recover. This trend is expected to continue as fallen stems continue to root. It is also possible that plants that we have not been able to find because they were covered with hurricane debris will be found in the future.

In addition to hurricane mortality, additional sources of mortality were discovered. Three plants were killed directly by people, including vehicle damage, off-target herbicide damage, and vandalism (chopping with a machete). One mortality was attributed to feral hog damage. In most cases, plants of all age classes died with no apparent cause. The latter two categories are probably much more significant than is indicated above. After the 2004 hurricane season many plants were covered by trees and other debris and could not be found for several more years or never to be positively identified as dead. Others probably suffered from root and stem injuries in the storms and died in later years. The fact that only 59.6% of plants that were alive in 2003 were alive in 2005, after direct hits by Hurricanes Jeanne and Francis, suggests that hurricanes have a significant impact on mortality. This was particularly important in Colony A, where plants grew under the canopy of the exotic tree *C. glaucophylla.* These trees did not withstand the hurricane winds very effectively, and many trees toppled or lost many of their branches. Many *H. fragrans* plants were covered by *C. glaucophylla* debris and were never relocated.

<u>Seed Germination and Seed Dormancy</u>
Final results for seed germination and seed dormancy are still pending as sampling will continue for another year. In terms of the seed box experiments, no seedlings have been observed in the *P. clausa* scrub habitat arrays to date. While the reasons for this lack of germination remain unknown, this finding reinforces the notion that this habitat type is not ideal for *H. fragrans*. Plants are rarely found within this habitat-type even though it is the dominant plant community at the SPSP. Future research should focus on environmental variables such as soil moisture and pH that may impact *H. fragrans* growth in this type of habitat. Seedlings were discovered in all the other locations, though only 1 seedling was found in seed boxes in Colony A. This is surprising because many seedlings have been found in the colony in the past, including 195 in winter 2003. More seedlings were found in colonies B and C, although there was no signification difference in numbers of seedlings found between the three colonies. No significant difference was found among sunlight treatments either, though the highest numbers of seedlings were observed in the shade treatments and almost none in full sunlight treatments. Despite a lack of statistical significance, we believe that shady conditions are preferred for seedlings as natural seedling clusters are also consistently observed in these areas as opposed to areas of full sunlight.

In the seed box experiments we observed a 1.76% germination rate. Fairchild Tropical Garden has shown that in the lab setting a mean germination of 94% can be achieved. Their germination tests do include treatment with 1N sulfuric acid and soaking in water. Their treatments also include daily observations, whereas we can only look for seedlings every four months. It is likely that seedlings have emerged in the field that were dead before they were seen. It is also probable that acid scarification of the seeds and subsequent soaking in water increased germination rates, so inferences from lab germination rates cannot be made. Most seedling clusters that have been found in the field since 1999 have been in very small areas, most likely the result of seeds dispersing from single fruit dropped directly on the ground under a parent plant. Germination rates may be higher if seeds are consumed by birds or other vertebrates.

Seed bank studies indicate that *H. fragrans* seeds maintain high viability over a 19 month period, losing very little viability over that period. This should serve to make *H. fragrans* more resilient after stochastic events, such as the two hurricanes in 2004 which directly killed many adult individuals.

For final results, seed boxes will be monitored through 2008 so that a full 3 years of germination data and dormancy data can be collected, which is required by population viability analysis models we anticipate using. The final seed dormancy bags were collected in winter 2007, as specified in the grant agreement, but results will not be available until later in 2008. An addendum to this report, including final results and the population viability analysis, will be submitted when data are collected.

<u>The Habitat of *H. fragrans*</u>

The results of the historical study are unfortunately inconclusive. Not enough historical data on the original vegetation of the Savannas PSP was found, despite extensive searching. More importantly, no data was found that links *H. fragrans* itself to any particular habitat-type prior to pineapple farming.

The geography of the Atlantic Coastal Ridge at the Savannas may have created a unique environment for *H. fragrans* that is different than other parts of the ridge to the north or south. The large wetland swale immediately behind the ridge and the proximity of the ridge immediately on the Indian River are unique. The geography may have resulted in unique habitats in the area. It is possible that fires along this section of the ridge were rarer than expected. Typically, scrub has a fire frequency of 50-100 years and is necessary for the regeneration of *Pinus clausa*. With the ridge being so narrow (ca. ¼ mile wide), and bordered by a deep wetland and the Indian River, natural lightning strikes igniting fires may have been rare. Thus, *P. clausa* scrub may have been limited in its extent, with xeric hammock being the climax community.

In addition, it has been suggested that the Atlantic Coastal Ridge at the Savannas may have originally been part of a barrier island with the Savannas wetland representing a saltwater lagoon (DEP 2003). If so, then prehistoric habitats here may have been similar to those where *H. aboriginum* grows in southwest Florida, including shell mounds and coastal berm. This may be one reason why *H. fragrans* is limited in distribution. The species may be persisting in this small area despite changes in sea level and changes in vegetation.

More recently, prior to European settlement, the habitat of *H. fragrans* may have been limited to the areas directly within and adjacent to hammocks. These hammocks certainly occurred along the Indian River, and may have extended across the ridge in places, replacing *P. clausa* scrub in the absence of fire. The species could well have grown in the partially and fully shaded areas within such hammocks below cabbage palms and oaks. At least some *P. clausa* scrub did occupy portions of the ridge at the Savannas, probably occupying the highest, most central portion of the ridge all the way across the SPSP, with hammocks on the eastern and western slopes.

The mosaic of vegetation communities on the Atlantic Coastal Ridge at the Savannas PSP, *i.e.* some areas dominated by sand pines, others by *Aristida gyrans*, and still other areas dominated by xeric hammocks, may be the result of different site histories in those areas. Several parts of the pineapple farming process may have influenced post-abandonment vegetation succession:

- Original vegetation

- Site preparation
- Period of use
- Fertilizer use
- Random, post-abandonment recruitment of seeds

Whether the extensive *Aristida gyrans* communities lacking sand pines occurred in the area prior to farming is unknown. The observation of “bare and dotted with patches of white sand, ("bleach yard,")” in the 1850s in the vicinity of what became Walton is unusual. No pines were mentioned. This could have either been the *Aristida gyrans* community, but is more likely a rosemary (*Ceratiola ericoides*) bald. The *Aristida gyrans* community is probably a post-farming plant association, not one that was historically at the Savannas. These areas do appear to have denser vegetation now than they did on 1940s aerials, probably because of more hardwoods recruiting into the areas, and a denser cover of herbaceous species.

We do not believe that *H. fragrans* was typically associated with the *P. clausa* scrub community. Further evidence of this now comes from seed germination studies discussed above. We have not found any seedlings in seed germination studies in *P. clausa* scrub after two years of observations. Management of the SPSP should serve to maintain the population of *H. fragrans.* Very little *H. fragrans* was found on private property to the east of the SPSP in surveys conducted in 2006 and 2007 (Woodmansee et al. 2007). While Woodmansee et al. (2007) did find new colonies of *H. fragrans* outside of St. Lucie County, the SPSP still maintains the majority of the global population of the species. Management activities where *H. fragrans* occurs should focus on exotic plant removal. Fire management, while critical to the SPSP as a whole, including *P. clausa* scrub areas, is detrimental to *H. fragrans*, killing adult plants and not promoting recruitment (Bradley et al. 2002). The ideal solution for management of the species would be restoration of coastal hammocks along the eastern slope of the Atlantic Coastal Ridge. This was probably the most occupied habitat for *H. fragrans* before human activities caused radical landscape level changes in the region. Unfortunately, almost the entire hammock area has been destroyed and is on hundreds of privately owned lots, so restoration options are limited. Maintenance of the *Aristida gyrans/ Sabal palmetto* habitat that *H. fragrans* now occupies remains the best option for the long-term existence of the species. This is particularly significant since the persistence of many of the populations found by Woodmansee et al. (2007) outside of St. Lucie County is unlikely in coming decades because of sea level rise.

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Appendix A: *Harrisia fragrans* seed dormancy

Germination of *Harrisia fragrans*
Seed Bank Trials
Second Annual Report

Submitted to: Institute for Regional Conservation

Submitted by: Don Walters and Joyce Maschinski
Center for Tropical Plant Conservation
Fairchild Tropical Botanic Garden
11935 Old Cutler Rd.
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November 1, 2007

Objective

The purpose of this experiment was to determine the longevity of *Harrisia fragrans* seed in its natural seed bank.

Methods

Using *Harrisia fragrans* seeds collected by The Institute for Regional Conservation (IRC) from the field in November 2005, March 2006, August 2006, November 2006, March 2007 and July 2007, FTBG conducted six germination trials. Seeds were freshly collected, buried for 3.5 months, 8 months, 11 months, 16 months and 19 months, respectively. Each germination trial had a total of 450 seeds; 5 replicates of 10 seeds collected from one plant growing in one of three conditions (sun, partial sun, or shade) in one of three sites (South, North and Middle).

We soaked seeds in 1N sulfuric acid (H_2SO_4) for 5 minutes, rinsed twice with water, and soaked in clean water for 15 to 20 minutes. We sowed seeds in Petri dishes filled half-way with coarse sand and water. The dishes were set in a growth chamber with the temperature set for 25^o C (77^o F), 50% relative humidity and 12/12 h photoperiod. Water was added to the dishes as needed after each observation (2-3 times per week). Seeds were determined to have germinated when the protruding radicle was visible. Each trial was run for 42 days.

We analyzed differences in percent germination, number of days to first germination, and number of days to maximum germination in two tests. We compared differences among the storage durations (3.5, 8, 11, 16, and 19 months), growing conditions, and sites using a two-way repeated measures analysis (GLM, Systat 10.2, SYSTAT Software Inc., 2002). For the fresh seeds (0 month storage), the seeds we received were not identified by plant or condition, therefore they could not be included in the test.

Results

Fresh seeds germinated readily (Mean 94%; 7 days to first germination and 25 days to maximum germination). Over 19 months all sites had high percentages of germination ranging from 100% to 64% (Figure 1). Mean percent germination significantly differed across sites ($F_{2,36} = 40.87$, P = 0.0001) and trials ($F_{4,144} = 3.718$, P = 0.007), but the differences across the trials were dependent on the site, condition, and trial ($F_{16,144} = 2.407$, P = 0.004). In general, the North site had lowest percent germination, and South had the highest germination. Importantly, there was not a consistent or significant decline in germination with increased time in the seed bank at any site except for the North Shade site. Although there was a marked decline in germination at the Middle Partial Shade and North partial Shade site after 11 months in the seed bank, subsequent trials at 16 and 19 months regained germination of approximately 90% of seeds (Table 1).

The number of days to the first germination significantly differed across sites ($F_{2,36} = 40.17$, P = 0.0001), condition ($F_{2,36} = 4.267$, P = 0.02) and trials ($F_{4,144} = 46.64$, P = 0.0001) and the differences across trials were dependent on the site and condition ($F_{16,144} = 1.992$, P = 0.02). At the Middle and North sites, germination was significantly slower in the 3.5, 11 and 19 month trials than in either the 0, 8 or 16 month trials. Averaged across all trials, time to first germination increased from South, North, to Middle site, respectively. Over 19 months, there has been a periodicity to the rate of first germination (Table 2; Fig. 2). Most sites and conditions had more rapid germination after 8 months (March 2006) and 16 months (November 2006) than during any other trial time (Fig. 2).

The number of days to maximum germination significantly differed across sites ($F_{2,36}$ = 72.13, P = 0.0001; Table 3), condition ($F_{2,36}$ = 3.957, P = 0.03) and trials ($F_{4,144}$ = 19.606, P = 0.0001), however maximum germination depended upon the condition and time of trial (Table 1, $F_{8,144}$ = 4.74, P = 0.0001). The Middle site had the longest period to maximum germination (28 d) in comparison to the South (20 d) and North sites (24 d). In all sites, the days to maximum germination was lowest for seeds that had been buried for 8 and 16 months. For shady conditions, days to maximum germination did not vary with duration of seed burial (24 d vs. 23 d in 3.5 and 6 mo, respectively), yet seeds in partial sun and sun conditions achieved maximum germination in significantly fewer days in the 6-month trial than in the 3.5 month trial.

Conclusions

The 5 minute sulfuric acid treatment appears to be an effective scarification method for germinating *Harrisia fragrans*.

Little loss in viability of *Harrisia fragrans* seeds appears to have occurred over the nineteen month trial period. The studies so far indicate that *Harrisia fragrans* is able to form a persistent seed bank for at least nineteen months.

The periodicity of germination rate is a curious pattern in the data. It will be interesting to see if this pattern persists in the 2008 trials.

We recommend that additional studies of soil moisture be conducted to determine why seed longevity and germination rate varies across conditions, sites, and trials.

Appendix A: *Harrisia fragrans* seed dormancy

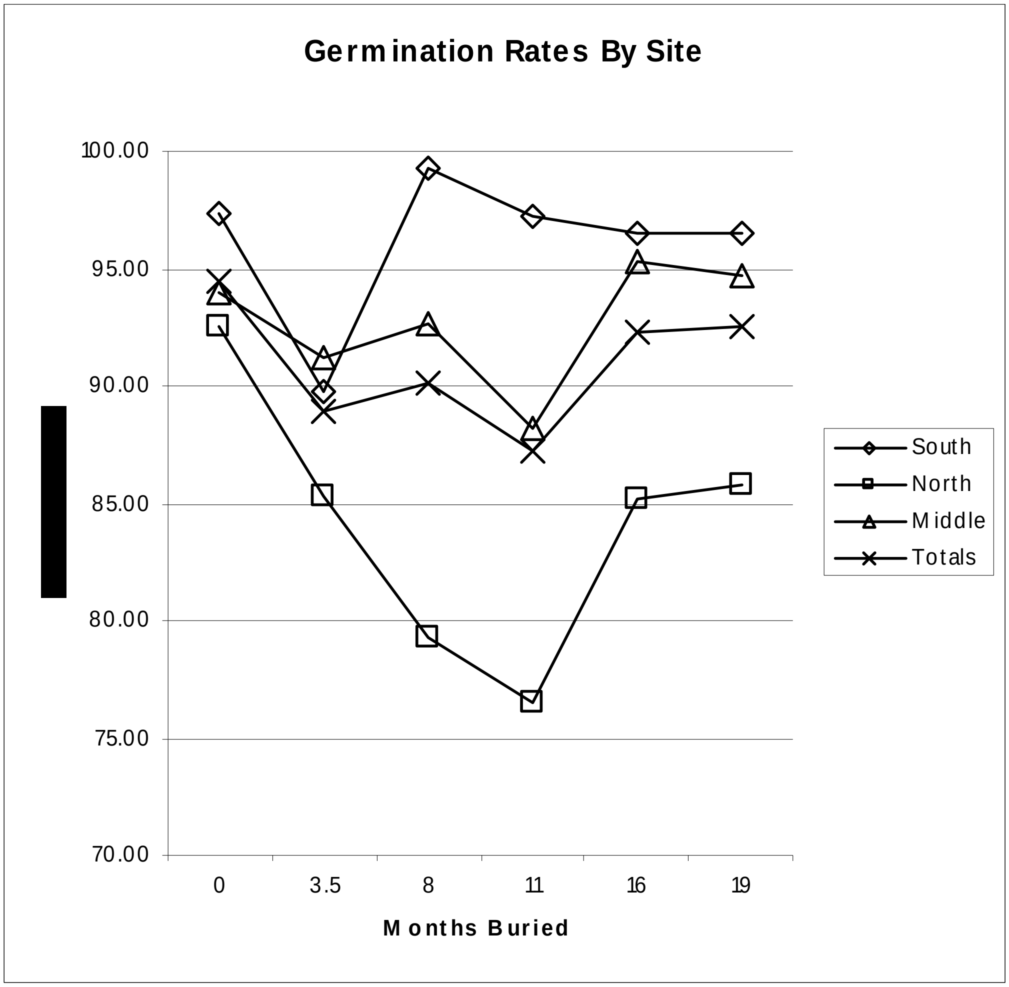

Figure 1. Total germination rates of *Harrisia fragrans* seeds buried in the seed bank for 0, 3.5, 8, 11, 16 and 19 months by site.

Appendix A: *Harrisia fragrans* seed dormancy

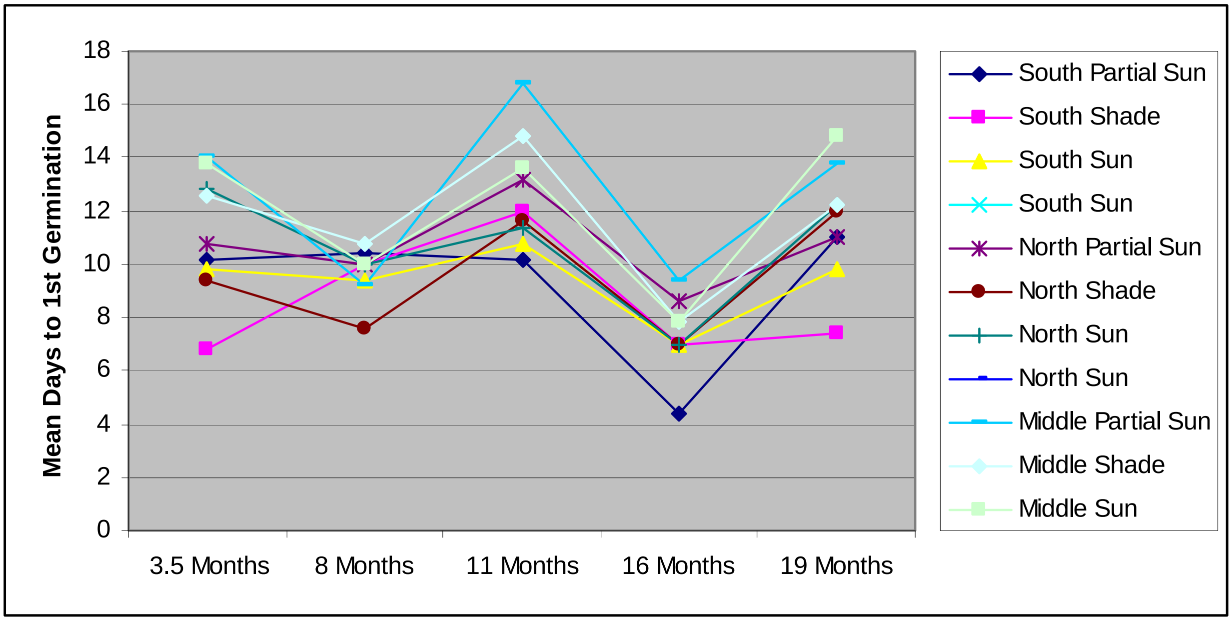

Figure 2. Comparison of mean days to first germination after 3.5 months (March 2006), 8 months (August 2006), 11 months (November 2006), 16 months (March 2007), and 19 months (July 2007) for *Harrisia fragrans* seeds buried in three locations and three conditions.

Appendix A: *Harrisia fragrans* seed dormancy

Site	Condition	Mean % Germ After 3.5 Months	SE	Mean % Germ After 8 Months	SE	Mean % Germ After 11 Months	SE	Mean % Germ After 16 Months	SE	Mean % Germ After 19 Months	SE
South	Partial Sun	94	2	100	0	100	0	95	3	98	2
South	Shade	94	4	100	0	96	3	98	2	94	4
South	Sun	80	3	98	2	96	2	96	4	98	2
North	Partial Sun	82	2	80	6	66	9	92	4	89	3
North	Shade	90	4	80	5	84	7	78	8	73	7
North	Sun	84	5	78	6	80	3	84	2	96	3
Middle	Partial Sun	88	4	100	0	64	15	90	3	92	3
Middle	Shade	92	2	88	4	90	4	100	0	100	0
Middle	Sun	96	2	90	3	94	4	98	2	92	2

Table 1. Mean percent germination for *Harrisia fragrans* seeds buried in the seed bank for 3.5, 8, 11, 16 and 19 months. SE = Standard Error

Site	Condition	Mean # Days to 1st After 3.5 Months	SE	Mean # Days to 1st After 8 Months	SE	Mean # Days to 1st After 11 Months	SE	Mean # Days to 1st After 16 Months	SE	Mean # Days to 1st After 19 Months	SE
South	Partial Sun	10.2	1	10.4	1	10.2	1	4.4	1	11	0
South	Shade	6.8	1	10	0	12	1	7	0	7.4	1
South	Sun	9.8	1	9.4	1	10.8	1	7	0	9.8	1
North	Partial Sun	10.8	1	10	0	13.2	2	8.6	1	11	0
North	Shade	9.4	1	7.6	1	11.6	1	7	0	12	1
North	Sun	12.8	1	10	0	11.4	2	7	0	12.2	1
Middle	Partial Sun	14	1	9.2	1	16.8	2	9.4	1	13.8	2
Middle	Shade	12.6	1	10.8	1	14.8	1	7.8	1	12.2	1
Middle	Sun	13.8	1	10	0	13.6	1	7.8	1	14.8	1

Table 2. Mean number of days to first germination for *Harrisia fragrans* seeds buried in the seed bank for 3.5, 8, 11, 16 and 19 months. SE = Standard Error

Appendix A: *Harrisia fragrans* seed dormancy

Site	Condition	Mean # Days to Max After 3.5 Months	SE	Mean # Days to Max After 8 Months	SE	Mean # Days to Max After 11 Months	SE	Mean # Days to Max After 16 Months	SE	Mean # Days to Max After 19 Months	SE
South	Partial Sun	25.4	4	13.4	1	20.2	4	13	1	17.4	1
South	Shade	19.2	1	20	1	26	3	15.2	2	18.2	1
South	Sun	31.8	3	15	1	22.6	2	13.2	1	28.8	3
North	Partial Sun	26	4	17.6	2	24.8	3	30.4	3	25.2	4
North	Shade	21.4	4	17.8	4	22.8	3	14	3	20.2	1
North	Sun	28.8	3	16.2	2	22.8	4	18	3	27.6	3
Middle	Partial Sun	28.8	4	20.2	2	36	3	28.2	3	35.6	1
Middle	Shade	32.4	2	31.6	3	32.8	2	18.6	3	28.6	3
Middle	Sun	31.4	2	21.4	5	34.2	2	25	3	35.6	1

Table 3. Mean number of days to maximum germination for *Harrisia fragrans* seeds buried in the seed bank for 3.5, 8, 11, 16 and 19 months. SE = Standard Error