

# Invasive Plant Science and Management

## Reestablishment Potential of Beach Vitex (*Vitex rotundifolia*) after Removal and Control Efforts

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Beach vitex is an invasive plant of coastal areas of the southeastern United States from Maryland to Georgia. Many resources have been dedicated to the control of established beach vitex stands. Successful eradication will require knowledge of this plant's ability to reestablish from seed after control efforts. To understand seed-based regenerative potential, studies were conducted to characterize the fruits and seeds, document the existence and size of seed banks, determine stratification requirements for germination, and ascertain seed dormancy mechanisms. Studies of fruit lots from three consecutive years (2003 to 2005) found that the average fruit contained 1.39 seeds, and more than 76% of fruits contained at least one viable seed. A positive correlation existed between seed number and both fruit mass and fruit diameter. A substantial soil seed bank was discovered that contained viable seeds 4 yr after vegetation removal. Stratification was required for seed germination. All stratification treatments induced germination, with highest rates realized when stratification was performed at 10 C for 8 or 12 wk. Germination rates were modestly increased (from 0 to 17%) through mild scarification in the absence of stratification. Results indicate that beach vitex has physical (fruit coat) and physiological (seed) dormancy mechanisms that are capable of delaying germination for multiple seasons, allowing development of a soil seed bank. Beach vitex can reestablish from seed after vegetation removal.

**Nomenclature:** Beach vitex, *Vitex rotundifolia* L. f.

**Key words:** Dormancy mechanism, fruit, germination, landscape plant, seed viability, invasive, seed bank.

Beach vitex (*Vitex rotundifolia* L. f.) is a deciduous, woody shrub that is native to the Pacific Rim (Neal 1965; Pope 1968). The plant has attractive floral and foliage characteristics and is tolerant of elevated salt concentrations (Dirr 1998). Beach vitex remained a rarity in the landscape trade until the mid-1980s, when it was promoted as a landscape plant for coastal areas of the southeast (Olsen and Bell 2005). It was widely planted to stabilize frontal dunes along the east coast of the United States in the aftermath of Hurricane Hugo in 1989. Established beach vitex communities are currently present in coastal areas from Maryland to Georgia and along the Gulf Coast of the United States (Cousins et al. 2010).

In coastal regions of the southeast, mature beach vitex populations dominate primary sand dune areas by forming dense stands that substantially reduce the population

density of native species (Gresham and Neal 2004; personal observation). Additionally, conservation groups fear that beach vitex impedes nesting activities of endangered, federally protected, sea turtles (CBVTF 2009). In 2005, a task force of public and private agencies in North and South Carolina was created to address concerns about the negative environmental impacts of beach vitex in coastal areas. The task force has primarily focused on education, eradication, and legislation. Since beach vitex was initially recognized as a coastal threat, many community ordinances have been enacted to prevent further planting of beach vitex in both South and North Carolina (Cousins et al. 2010). In February 2009, the North Carolina Department of Agriculture officially listed beach vitex as a Class B State Noxious Weed (NCDA&CS 2010). Regional coastal surveys are being conducted to detect populations, and control measures are being implemented on infested sites.

Current control methods involve herbicide applications to trunks and stems followed by aboveground vegetation removal. Regrowth continues to be observed at cleared sites. Beach vitex rapidly reproduces vegetatively by producing long runners (growth of up to 2 m yr<sup>-1</sup>) that root at nodes (Cousins et al. 2010; Gresham and Neal

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\* Graduate Student, Adjunct Assistant Professor, and Professor, Department of Horticulture, Clemson University, Clemson, SC 29632; Associate Professor Emeritus, Associate Professor, Department of Forestry and Natural Resources, Clemson University, Clemson, SC 29634. Corresponding author's E-mail: twhtwll@clemson.edu

## Interpretive Summary

The findings of this paper are of importance in the continued efforts to control beach vitex. The ultimate success of the eradication projects will depend on the potential for beach vitex to reestablish itself at sites where it has been removed. Vegetative invasive characteristics have been investigated, but the potential for reestablishment from fruits is not well understood.

Beach vitex fruits were found to contain 1.39 seeds, with more than 76% of fruits containing viable seeds. Thus, nearly all beach vitex fruits could aid in reestablishment. Large beach vitex seed banks were found and characterized. No seed bank degradation was detectable in the 2 yr after beach vitex removal. Germination occurred in all stratification treatments, indicating that the majority of seeds should meet dormancy requirements on the dunes of the southeast. The highest germination percentages were observed when fruits were subjected to stratification at 10 C for 8 or 12 wk. To explain how fruits persist in the substrate despite the apparent ease of satisfying stratification requirements, scarification studies were performed to test the existence of a physical dormancy mechanism. Germination rates increased in response to fruit ablation from 0 to 17%. This mechanism helps explain how beach vitex is capable of traveling on ocean currents. As a result of these findings, long-term seedling control practices must be implemented in areas previously inhabited by beach vitex. In the future, control methods must include surveys of sites and removal of seedlings for at least four seasons after vegetation removal.

2004). Any surviving plants can quickly repopulate areas that have been cleared, and retreatment is required for a period of several years (Cousins et al., 2010).

The potential for beach vitex to reestablish on cleared sites depends not only on its vegetative invasive characteristics but also on fruit and seed characteristics. Beach vitex produces large numbers of fruits, up to 6,000 m<sup>-2</sup> (Gresham and Neal 2004), which are botanically drupes (FOCEC 1994). Fruits contain from zero to four seeds in separate locules (de Kok 2007). Fruits and seedlings have been observed in large numbers at sites that have been cleared (Figure 1). Beach vitex seedlings have also been detected on oceanfront sites where it was not planted. Fruits have a thick, hydrophobic cuticle, readily float when placed in water, and are apparently dispersed by ocean waves and currents (Cousins et al. 2009, 2010; Munir 1987).

The goal of this research was to investigate the potential for beach vitex to reestablish from seed after vegetation removal. First, beach vitex fruits and seeds were characterized as a measure of number and viability. Second, the sand substrate of vegetated and cleared sites was surveyed to determine whether fruits and viable seeds are stored in a soil seed bank. Third, seed germination requirements and dormancy mechanisms were investigated through stratification and scarification studies. It is anticipated that these results will contribute to the development of long-term control measures designed to eradicate beach vitex from oceanfront dune systems in infested areas.

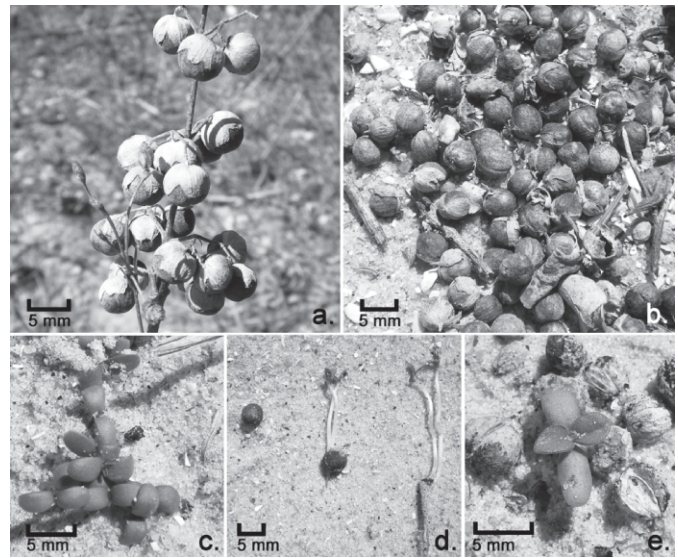


Figure 1. Fruit and seedling emergence characteristics of beach vitex. Fruits remain attached to the plant into the late winter (a) before being deposited on the surface of the sand (b). Emerged plants are typically found in large groups (c). Hypocotyls elongate to allow leaf emergence above the surface (d). First pair of true leaves develops above cotyledons (e).

## Materials and Methods

**Fruit and Seed Characteristics.** Beach vitex fruits were collected in December 2003, December 2004, and September 2005 at Pawleys Island, SC. The collection site had been planted with beach vitex in the mid to late 1990s and was covered by dense, mature growth. Fruits were collected before abscission and stored in plastic bags at room temperature (22 C) for less than 1 yr before analysis.

Fruits collected in 2003 and 2004 ( $n = 99$ ) were bisected equatorially to expose all four locules and placed by lots of 11 fruits into petri plates containing 25 ml of a 1% 2,3,5-triphenyltetrazolium chloride (TTC)<sup>1</sup> solution. Plates were incubated in the dark for 24 h at 22 C (ISTA 1985). The numbers of seeds per fruit and seed viability were determined. All seeds observed were plump and white, and no degradation or decay was observed. Seeds that stained pink from exposure to the TTC solution were considered viable. As a negative control, 30 fruits were autoclaved at 121 C for 20 min. Thirty fruits that were not autoclaved served as the positive control. No viable seeds were found in fruits that had been autoclaved, and all seeds were viable in fruits that were not autoclaved.

The 2005 lot contained 312 fruits. Each fruit was measured (equatorial diameter), weighed (mass), and bisected equatorially. One of the two halves was placed into a 24-well microtiter plate and covered with 1 ml of TTC solution (prepared as described above).

Seed number and viability averages were compared over years by ANOVA and linear contrasts with  $\alpha = 0.05$ . For

the 2005 sampling, number of seeds in each fruit was determined and correlated with fruit mass and diameter. Spearman correlation coefficients ( $\rho$ ,  $p$ ) were calculated to determine quality of correlation. This coefficient is preferred over Pearson's correlation coefficient ( $r$ ) because number of seeds per fruit is a discrete variable, ranging from zero to four. Analyses were performed with SAS (Version 9.1),<sup>2</sup> and graphics were produced with Microsoft Excel 2003.<sup>3</sup>

**Seed Bank Characterization.** *Seed Bank Survey over 4 yr.* The soil seed banks of two sites near Pawleys Island, SC, were surveyed in April 2006, 2007, 2008, and 2009. One site (Litchfield Beach, SC) had been purged of above-ground beach vitex vegetation by mechanical removal and herbicide applications in the spring of 2005. The second site (Pawleys Island) had actively growing beach vitex stands during the 2006, 2007, and 2008 samplings. Vegetation was subsequently removed before fruit deposition in late summer 2008. Both sites had been vegetated for more than 5 yr before the initial survey. Soil type was beach sand (carbon content: 0.3% by weight), and sampling was conducted at an elevation of 5 m above sea level. Soil cores were collected on a grid with sample spacing of 2 m. At the Litchfield site, 20 locations were sampled in 2006, whereas nine locations were sampled in 2007, 2008, and 2009. For the Pawleys site, nine locations were sampled in each of the four sampling years. Each sampling location was sampled to a depth of 15 cm. The 2006 samples were collected with a 10-cm-diam steel pipe. In 2007, 2008, and 2009, sampling was conducted with a 20-cm-diam PVC pipe. Sand cores were separated into 5-cm increments to yield three depth subsamples per sampling location, consisting of the surface (0 to 5 cm), middle (5 to 10 cm), and deepest (10 to 15 cm) layers. Samples were screened and washed on site to remove sand, and fruits were placed into plastic storage bags.

Seed number and viability were determined as previously noted. All collected fruits were tested in 2006. However, because of the large number of fruits collected in 2007, 2008, and 2009, viability tests were performed on a subsample of fruits. When more than 20 fruits were collected in a sample, a 20-fruit subsample was tested for viability. Statistical analyses were not performed on data because the experiment included only two experimental units (sites). Values presented were normalized to number of seeds and fruits ( $m^{-2}$ ).

*Seed Bank Persistence after Vegetation Removal.* A second study was conducted to document soil seed bank dynamics and persistence after vegetation removal. Nine sites on Pawleys Island were sampled in April 2009. At the time of sampling, three sites had active beach vitex infestations, three sites had been cleared before fruit production in 2008, and three sites had been cleared before fruit

production in 2007. Beach vitex had been established or previously established at all sites for at least 5 yr. Each of the nine sites was sampled at five locations. Sampling was performed to a depth of 15 cm with the use of a 20-cm-diam PVC pipe, and sand was partitioned by depth as previously described. Viability and seed number were determined as previously described.

Fruit numbers and seed viability values from the sites and depths were subjected to ANOVA and linear contrasts with  $\alpha = 0.05$ . A log transformation was conducted on number of fruits and number of viable seeds before analysis. Transformations were used to minimize heteroscedasticity of the variances. Analyses were performed with SAS (Version 9.1).<sup>2</sup>

**Seed Germination Requirements.** *Stratification Study.* The source of the fruits used in the stratification studies was a mature oceanfront planting of beach vitex at Pawleys Island, SC, that had been vegetated for more than five seasons before fruit collection. In December 2005, fruits were harvested after leaf fall had occurred, placed in plastic bags, and stored at room temperature (22 C) until experiments commenced.

Experimental design was a 3 by 3 factorial with three stratification temperature treatments (5, 10, and 15 C) and three stratification duration treatments (4, 8, and 12 wk). Planting dates (February through April 2007) were staggered four weeks apart so that all treatments simultaneously concluded. Preceding planting, fruits (approximately 300) were placed in 1 L of distilled water for 48 h at room temperature (22 C). Hydration requirements were indicated by an earlier stratification study in which only 11% germination was realized when dried fruits were planted and stratified. A subset of the soaked fruits were planted in 72-cell (12, 6 packs) planting flats that had been filled with Fafard 4-B media.<sup>4</sup> Fruits were planted at a rate of 1 fruit  $cell^{-1}$  at a depth of 1.5 cm. On each planting date, three flats were planted, watered to saturation, allowed to drain, and sealed with plastic wrap. One planted flat was placed in each of three temperature treatment coolers set to 5, 10, or 15 C. Substrate moisture was maintained at field capacity through periodic water addition to correct for limited amounts of water loss. Relative humidity of the coolers was held at 65%.

Twelve weeks after the first planting date, all flats were removed from the coolers (May 2007). Individual six-packs from each temperature by duration treatment condition were randomly placed into plant trays, with each tray containing one six-pack from every treatment condition. One six-cell pack served as an experimental unit, and each treatment was replicated 12 times. As a negative control for stratification, fruits that had been soaked for 48 hours and not subjected to stratification were planted as above. Flats were placed in a greenhouse (27/21 C day/night

Table 1. Beach vitex fruit characteristics (mean ± standard error) were evaluated over three seasons.

Year	No. of seeds	No. of viable seeds	Viable seeds	Fruits w/viable seeds
	fruit <sup>-1</sup>		%	
2003	1.32 ± 0.09	1.20 ± 0.07	91.2 ± 4.8	81.8 ± 4.6
2004	1.29 ± 0.06	1.27 ± 0.06	98.4 ± 0.8	76.8 ± 3.6
2005	1.57 ± 0.07	1.43 ± 0.08	90.7 ± 2.0	84.9 ± 3.0
Contrasts			Pr > F	
2003 vs. 2004	0.7694	0.5207	0.0741	0.3259
2003 vs. 2005	0.0130	0.0276	0.8898	0.4600
2004 vs. 2005	0.0068	0.1101	0.0284	0.0701

temperatures) and watered as needed. The study was repeated with stratification treatments ending 2 wk after the first study. Germination data were recorded weekly beginning 2 wk after placement in the greenhouse and terminating in week 8. Seeds were considered to have germinated when cotyledons were visible above the media surface.

Two hundred unstratified fruits were tested for viability as previously described after being soaked for 48 h in distilled water. Additionally, fruits from the unstratified control treatment were removed from the media at the end of the experiment and tested for viability.

The GLIMMIX Procedure for generalized linear mixed models was used to analyze logit-transformed germination data. Treatment comparisons were carried out on data points collected 4 wk after placement in the greenhouse. In 4 wk, maximum germination had been attained for all treatments. Data were analyzed by ANOVA and linear contrasts with  $\alpha = 0.05$ . Analyses were performed with SAS (Version 9.1),<sup>2</sup> and graphs were constructed in Microsoft Excel 2003<sup>3</sup> with nontransformed data.

*Preliminary Scarification Studies.* Fruits collected in 2007 at a site on Pawleys Island that had been vegetated for more than 5 yr were subjected to a scarification experiment in spring 2008. There were three scarification treatments: scalpel fruit ablation to expose a single seed, 30 s in concentrated sulfuric acid,<sup>1</sup> and sandpaper removal of the entire exocarp (but leaving mesocarp tissue). Each treatment lot contained 30 fruits, and an additional 30 fruits, not subjected to scarification, served as a control. The fruits were placed in groups of 10 into petri plates with 25 ml of distilled H<sub>2</sub>O. Plates were sealed with Parafilm and incubated in the dark at 27 C for 4 wk (time selected on the basis of results of germination study). At the end of the study, germinated seeds were counted. A second experiment compared germination of 30 seeds that had been excised from 30 fruits with 30 unscarified fruits. Incubation protocol and conditions were as above.

## Results and Discussion

**Fruit and Seed Characteristics.** Over 3 yr (2003, 2004, and 2005), 76 to 85% of the fruits contained at least one viable seed, and more than 90% of seeds observed were viable (Table 1). Fruits from 2003, 2004, and 2005 contained 1.32, 1.29, and 1.57 total seeds with 91%, 98%, and 91% seed viability, respectively. Fruits from 2005 contained more seeds than those from 2003 or 2004, and seeds from 2004 were more likely to be viable than those from 2005, whereas 2003 seed viability was not different from 2004 or 2005 viability. Approximately 20% of fruits had no seeds, whereas nearly all other fruits lacked a complete seed complement (Table 2). Most fruits contained one or two seeds (69%), whereas only a small percentage contained four seeds (2%).

Table 2. The percentages of beach vitex fruits (mean ± standard error) containing zero, one, two, three, or four seeds for fruits collected over 3 yr.<sup>a</sup>

Seeds per fruit	2003	2004	2005
	% of total fruits		
0	18.2 ± 4.6	23.2 ± 3.6	15.1 ± 1.8
1	41.4 ± 5.3	38.4 ± 1.0	32.7 ± 1.9
2	31.3 ± 5.3	27.3 ± 3.5	35.6 ± 1.4
3	8.1 ± 0.02	8.9 ± 0.01	13.5 ± 1.2
4	1.0 ± 0.01	3.0 ± 0.02	3.2 ± 0.9
Contrasts		Pr > F	
0 vs. 1	0.0025	0.0015	< 0.0001
0 vs. 2	0.0466	0.2751	< 0.0001
0 vs. 3	0.1114	0.0015	0.4560
0 vs. 4	0.0141	0.0002	< 0.0001
1 vs. 2	0.1114	0.0099	0.1835
1 vs. 3	0.0002	< 0.0001	< 0.0001
1 vs. 4	< 0.0001	< 0.0001	< 0.0001
2 vs. 3	0.0025	0.0003	< 0.0001
2 vs. 4	0.0004	< 0.0001	< 0.0001
3 vs. 4	0.2496	0.1795	< 0.0001

<sup>a</sup>The year by number of seed interaction term was significant.

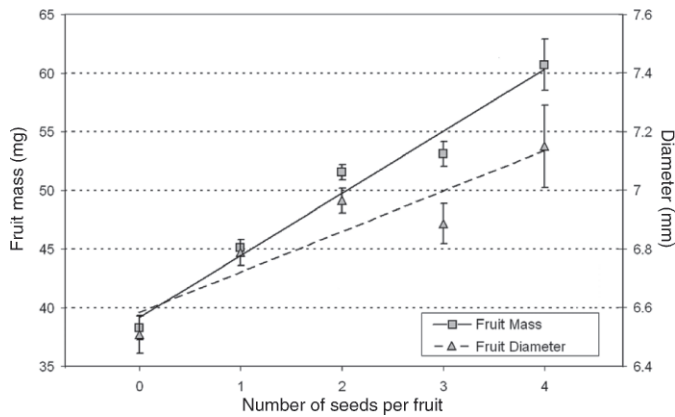


Figure 2. Correlation of mass and diameter with number of seeds per fruit. Error bars = average  $\pm$  standard error. Fruit mass by number of seeds regression equation:  $y = 5.2843x + 39.165$  ( $\rho = 0.61$ ). Fruit diameter by number of seeds regression equation:  $y = 0.1382x + 6.5828$  ( $\rho = 0.27$ ).

Fruit mass and diameter were correlated with number of seeds for the 2005 fruit lot ( $\alpha = 0.05$ ; Figure 2). In general, as seed number increased, the mass and diameter of the fruit increased. Fruits containing zero, one, two, three, and four seeds weighed 38.3, 45.1, 51.5, 53.1, and 60.7 mg, respectively. Fruits containing zero, one, two, three, and four seeds measured 6.5, 6.8, 7.0, 6.9, and 7.2 mm diam, respectively. Mass (Spearman coefficient [ $\rho$ ] = 0.61) correlated with seed number better than diameter (Spearman coefficient [ $\rho$ ] = 0.27).

The majority (80%) of fruits contain at least one viable seed. Seed abortion before maturation (de Kok 2007) might explain the absence of seeds in approximately 20% of fruits. The data indicate small yet significant differences in seed and viable seed production over years; however, because fruits are produced in large quantities, these differences are unlikely to factor into the overall potential for reestablishment.

**Seed Bank Characterization.** *Seed Bank Survey over 4 yr.* Large numbers of fruits and viable seeds were found in the substrate of both the site cleared in 2005 (Litchfield) and the site cleared in 2008 (Pawleys) over the four sampling years (2006, 2007, 2008, and 2009) (Figure 1; Table 3). At the Litchfield site, the number of fruits (data not shown) and number of fruits with viable seeds (Table 3) declined between the measurement taken one season after removal (2006) and the measurement taken four seasons after removal (2009). For the Pawleys site, an increase in numbers of fruits (data not shown) and fruits with viable seeds (Table 3) occurred through 2008. The 2009 data indicate a decrease in numbers of fruits and numbers of viable seeds after vegetation removal in summer 2008. The majority of fruits were found in the surface subsamples at both sites. Large standard errors indicate large spatial variability in fruit deposition in the sand. High levels of fruits and viable seeds found in the substrate demonstrate the presence of a soil seed bank at these two sites. This seed bank remained viable for four seasons after vegetation removal.

*Seed Bank Persistence after Vegetation Removal.* Length of time after beach vitex removal did not affect the numbers of fruits and viable seeds in the seed bank (Table 4). An average of 1,833 viable seeds  $m^{-2}$  was found in the soil seed bank over all sampled sites. Significant depth effects were present for all sites sampled, with fruit numbers decreasing by roughly an order of magnitude with each successive 5-cm increase in depth (Table 4). Number of viable seeds in the substrate followed a similar depth distribution pattern.

Large numbers of fruits and seeds are present in the substrate of vegetated and cleared beach vitex sites. The fact that no significant changes were noted after 2 yr without fruit deposition indicates the existence of a stable and persistent seed bank. Because the distribution of fruits within substrate layers remained similar through 2 yr after

Table 3. Viable beach vitex seeds in the seed bank (mean  $\pm$  standard error) for the Litchfield (vegetation removed in 2005) and Pawleys (vegetation removed in 2008) sites.

Depth	No. of viable seeds			
	2006	2007	2008	2009
cm	$m^{-2}$			
Litchfield				
0–5	738 $\pm$ 160	977 $\pm$ 363	390 $\pm$ 236	469 $\pm$ 233
5–10	102 $\pm$ 70	94 $\pm$ 51	0 $\pm$ 0	32 $\pm$ 15
10–15	32 $\pm$ 32	3 $\pm$ 3	0 $\pm$ 0	0 $\pm$ 0
Pawleys				
0–5	2,363 $\pm$ 714	1,816 $\pm$ 275	5,061 $\pm$ 1,842	771 $\pm$ 207
5–10	71 $\pm$ 48	593 $\pm$ 217	438 $\pm$ 186	45 $\pm$ 21
10–15	127 $\pm$ 127	131 $\pm$ 35	181 $\pm$ 133	32 $\pm$ 24

Table 4. Beach vitex fruits and viable seeds (mean ± standard deviation) in the soil of sites that had beach vitex fruit set in the season of sampling or had been without beach vitex fruit set for the previous one or two seasons.<sup>a</sup>

Depth	Fruits		Viable seeds	
	m <sup>-2</sup>			
cm				
0–5 <sup>b</sup>	3,644 ± 580		1,687 ± 228	
5–10	258 ± 41		138 ± 7	
10–15	28 ± 8		8 ± 2	
Contrasts	Pr > F			
0–5 vs. 5–10	0.0001		0.0002	
0–5 vs. 10–15	< 0.0001		< 0.0001	
5–10 vs. 10–15	0.0002		< 0.0001	

<sup>a</sup> A log<sub>10</sub> transformation was used prior to ANOVA. Non-transformed data are presented.

<sup>b</sup> Data were pooled by depth because seed banks were not different among vegetated and cleared sites.

beach vitex removal, downward migration of fruits due to sand accretion does not appear to be occurring. When taken alongside the data from the previous smaller scale seed bank study, these findings support the conclusion that seeds from the beach vitex seed bank could potentially produce seedlings even 4 yr after vegetation removal.

**Seed Germination Requirements. Stratification Study.** Data from the repeated experiments were analyzed separately because there was a significant experiment effect when data were pooled. However, both experiments yielded similar results by treatment comparison (i.e., there was no significant treatment by experiment interaction). Differences were generally magnitude-based, with all values significantly lower in the second experiment. The only recognized discrepancy in experimental conditions is that the first study was moved to a smaller greenhouse after 2 weeks when the second study was removed from treatment coolers. Light quantity might have been initially higher for the first experiment than the second. Data presented are for the first experiment.

Analysis of stratification data indicated no interaction between temperature and duration of stratification. As a result, data were pooled by treatment factors for analysis (Table 5). The numerically highest and numerically lowest germination curves along with the experiment average curve are presented to illustrate the shape of the logarithmic germination curves generated over the 6-wk data collection period (Figure 3). The highest germination percentages were realized with 10 C stratification. Differences were not detected between the 5 C and 15 C treatments. Germination rates elicited by 4, 8, or 12 wk of stratification were not different. The pooled germination for the first

Table 5. Germination (mean ± standard deviation) 4 wk after stratification at 5, 10, or 15 C for 4, 8, or 12 wk.

Stratification factor	Germination	
	%	
Temperature <sup>a</sup>		
5 C	71.3 ± 3.3	
10 C	81.0 ± 2.6	
15 C	69.4 ± 3.8	
Duration <sup>a</sup>		
4 wk	76.4 ± 2.9	
8 wk	76.9 ± 3.5	
12 wk	68.5 ± 3.5	
Contrasts	Pr > F	
5 vs. 10 C	0.0308	
5 vs. 15 C	0.7132	
10 vs. 15 C	0.0124	
4 vs. 8 wk	0.7834	
4 vs. 12 wk	0.0978	
8 vs. 12 wk	0.0580	

<sup>a</sup> The temperature by time interaction was not significant. Stratification factors were analyzed separately.

experiment was 73%, similar to the 71% germination rate that was previously noted in a laboratory setting (Chong-Min and EulSoo 2001). In the nonstratified control treatment, germination was limited to only one seed, whereas 93% of fruits were later found to contain viable seeds (data not shown).

Results demonstrate that beach vitex seeds require stratification for germination, indicating the presence of a physiological dormancy mechanism. A similar dormancy mechanism (endosperm) was found in *Vitex agnus-castus* seeds (Belhadj et al. 1998). Seed dormancy in beach vitex can be satisfied over a wide range of temperatures and times of stratification (experiment average, 73% germination

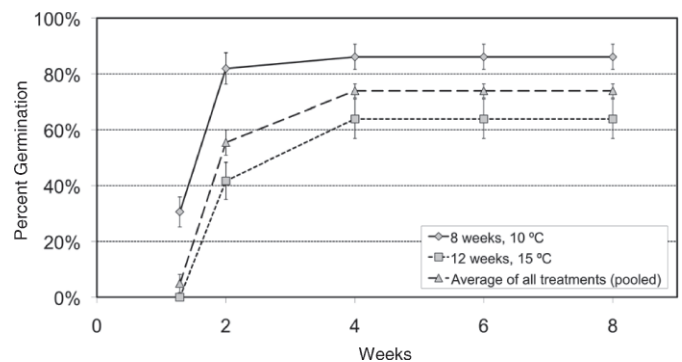


Figure 3. Germination percentages for the treatments that yielded the highest (8 wk, 10 C), and lowest (12 wk, 15 C) germination percentages are presented in addition to the pooled germination percentage for all treatments. Error bars = average ± standard error.

after stratification). This is further supported by the fact that beach vitex thrives over a wide latitude range (0 to 38°) (Lee et al. 2007; Munir 1987).

*Preliminary Scarification Studies.* Ablation of fruit coverings resulted in low levels of immediate germination independent of stratification. Scalpel scarification of fruits that exposed the seeds contained within resulted in 17% germination, whereas sulfuric acid-scarified, sandpaper-scarified, and control-unscarified fruits did not germinate (data not presented). In the follow-up study, 13% germination resulted when seeds were removed from fruits. These results indicate that the fruit enforces a portion of the seed dormancy mechanism. This interpretation is supported by the existence of a physical dormancy mechanism in *Vitex parviflora* Juss. (Umali-Garcia 1980) and *Vitex agnus-castus* L. seeds, which show both a seed coat and endosperm dormancy (Belhadj et al. 1998). Additionally, the existence of a physical dormancy mechanism is supported by the fact that a 48-h hydration period before stratification increased germination percentages from 11 to 73%. This soaking period appears to have been required to allow time for water to penetrate the hydrophobic exocarp (Cousins et al. 2009).

We have demonstrated that the fruits of beach vitex have potential to aid in the reestablishment of beach vitex after initial site clearing. The majority of fruits (approximately 80%) contain viable seeds, and these fruits are stored in a substantial and long lived (as long as four seasons) soil seed bank. Seeds will meet their stratification requirements along the coast of the southeast as noted by the wide range of temperatures and times that elicited appreciable germination. Finally, the apparent contradiction between the existence of a soil seed bank and the apparent ease of meeting stratification requirements was reconciled with the identification of a physical dormancy mechanism capable of preserving fruits in the soil for many seasons. In addition to protecting seeds from decay and slowing germination, such a dormancy mechanism could also prevent seeds from prematurely germinating while traveling to new locations on ocean currents. Although it is unknown whether this fruit coat dormancy would be overcome in sand dune habitats, there appears to be a high probability that beach vitex will re-establish from seed after vegetation control. Scouting and seedling control measures should be developed and included in management protocols for at least four seasons after beach vitex removal.

### Sources of Materials

<sup>1</sup> Sigma-Aldrich Corporation, 3050 Spruce Street, St. Louis, MO 63103.

<sup>2</sup> SAS Institute, 100 SAS Campus Drive, Cary, NC 27513.

<sup>3</sup> Microsoft Corporation, 1 Microsoft Way, Redmond, WA 98052.

<sup>4</sup> Conrad Fafard Inc., 1471 Amity Road, Anderson, SC 29621.

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