

**Endangered Species Act 5-Year Review**  
**Johnson's Seagrass**  
**(*Halophila johnsonii* Eiseman)**

National Oceanic and Atmospheric Administration  
National Marine Fisheries Service

November 2007

**5-Year Review**  
**Species Reviewed: Johnson's Seagrass (*Halophila johnsonii* Eiseman)**

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**5-Year Review**  
**Johnson's Seagrass (*Halophila johnsonii* Eiseman)**

**1. GENERAL INFORMATION**

**1.1. Reviewers**

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**1.2 Methodology Used to Complete the Review**

The National Marine Fisheries Service (NMFS) of the National Oceanic and Atmospheric Administration (NOAA) in conjunction with the Center for Coastal Fisheries and Habitat Research (CCFHR), of the same (NOAA), initiated a 5-year review of Johnson's Seagrass (*Halophila johnsonii* Eiseman) in September 2006. The CCFHR and NMFS solicited information from the public through Federal Register notice (71 FR 60108, October 12, 2006), as well as through personal and written communications with several educational institutions, federal and state governments, and private research organizations. To complete the review, we

evaluated all information that has become available on the species since 1997, the date of its last biological status review. Thus, the review is based upon the best scientific and commercial data available.

### **1.3. Background**

#### **1.3.1. FR notice citation announcing initiation of this review**

The notice announcing the initiation of this 5-year review and requesting information from the public was published on October 12, 2006 (71 FR 60108).

#### **1.3.2. Species status**

The status of this species is “threatened” according to the September 14, 1998, listing.

#### **1.3.3. Listing history**

FR notice: 63 FR 49035  
Date listed: September 14, 1998  
Entity listed: *Halophila johnsonii* Eiseman  
Classification: threatened

Critical habitat designation  
FR notice: 65 FR 17786  
Date of notice: April 5, 2000

#### **1.3.4. Associated rulemakings**

No associated rulemaking has occurred for this species.

#### **1.3.5. Review history**

*The Distribution, Abundance, and Ecology of Halophila johnsonii Eisemen in the Lower Indian River, Florida* by W. Judson Kenworthy, Southeast Fisheries Science Center, Beaufort Laboratory, NMFS, NOAA, Beaufort, NC. Submitted to Office of Protected Resources, NMFS, NOAA, Silver Spring, MD, 1993.

*An Updated Biological Status Review and Summary of the Proceedings of a Workshop to Review the Biological Status of the Seagrass, Halophila johnsonii Eiseman* by W. Judson Kenworthy, Southeast Fisheries Science Center Beaufort Laboratory, NMFS, NOAA, Beaufort, NC. Submitted to Office of Protected Resources, NMFS, NOAA, Silver Spring, MD, October 15, 1997.

### **1.3.6. Recovery plan or outline**

National Marine Fisheries Service. 2002. Recovery Plan for Johnson's Seagrass (*Halophila johnsonii* Eiseman). Prepared by the Johnson's Seagrass Recovery Team for the National Marine Fisheries Service, Silver Spring, MD, 120 pages.

### **1.3.7. Species recovery priority**

Johnson's seagrass is assigned a recovery priority of seven, based on a moderate magnitude of threats, a low-moderate recovery potential, and the potential for economic conflict. The moderate magnitude of threat is derived from the threats discussed in 2.3.2. The recovery potential was considered to be low-moderate, and economic conflict was considered to exist based on anticipated future in-water construction projects (i.e., dredging, dock construction, and projects that adversely modify water quality).

## **2. REVIEW ANALYSIS**

### **2.1 Application of the 1996 Distinct Population Segment (DPS) policy**

The Endangered Species Act of 1973 (ESA) defines species as including any subspecies of fish or wildlife or plants, and any distinct population segment of any species of vertebrate wildlife. This definition limits listings as distinct population segments (DPS) only to vertebrate species of fish and wildlife. Because the species under review is a plant and the DPS policy is not applicable, the application of the DPS policy to the species listing is not addressed further in this review.

### **2.2 Recovery criteria**

#### **2.2.1 Does the species have a final, approved recovery plan containing objective, measurable criteria?**

Yes  
 No

Yes, however revisions to criteria two and three should be considered to improve their clarity. Also, the plan would be improved by the addition of threats-based criteria.

#### **2.2.2 Adequacy of recovery criteria**

##### **2.2.2.1 Do the recovery criteria reflect the best available and most up-to date information on the biology of the species and its habitat?**

Yes  
 No

No, the recovery plan was written in 2002, and new data have since become available regarding the species, its genetics, distribution, and habitat. In addition threats-based criteria and action to address listing factors relevant to species should be added to the recovery plan. Criteria two and three in the recovery plan should be improved so that progress toward recovery is more measurable.

**2.2.2.2 Are all of the 5 listing factors that are relevant to the species addressed in the recovery criteria (and is there no new information to consider regarding existing or new threats)?**

**Yes**

**No**

Johnson's seagrass was listed as threatened based on a combination of the following factors, described in section 4(a)(1) of the ESA:

- Present or threatened destruction, modification, or curtailment of its habitat or range.
- Other natural or human-made factors affecting the species' continued existence.
- The inadequacy of existing regulatory mechanisms.

**2.2.3. List the recovery criteria as they appear in the recovery plan, and discuss how each criterion has or has not been met, citing information**

1. The species' present geographic range remains stable for at least 10 years or increases,
2. Self-sustaining populations are present throughout the range at distances less than or equal to the maximum dispersal distance to allow for stable vegetative recruitment and genetic diversity, and
3. Populations and supporting habitat in its geographic range have long-term protection (through regulatory action or purchase acquisition).

Criterion 1 has been met. Monitoring of the northern and southern range limits indicates there have been no significant changes in the past 10 years (see discussion below on Distribution and Abundance).

The status of progress in meeting criterion 2 is still under evaluation. The definition of self-sustaining populations needs to be clarified in the recovery plan, and the recovery plan needs to be revised to reflect the new information regarding the spatial and temporal fluctuations in *Halophila johnsonii*'s distribution, abundance, and population dynamics. Based on experimental work with clonal fragment dispersal potential, the maximum dispersal distances need to be calculated and compared to reported spatial distribution of the species to better quantify this criteria. More research is needed to identify the correspondence between genetic diversity and factors which sustain the populations.

Criterion 3 does not appear to be met. This criterion in particular requires significant re-evaluation to determine specific recovery actions that will help ensure the criterion is met and sustained. See discussion in 2.3.2.4. on adequacy of existing regulatory mechanisms.

## 2.3. Updated information and current species status

### 2.3.1. Biology and habitat

The last status review for *H. johnsonii* was conducted before the species was formally listed as threatened (Kenworthy, 1997). Since that time, new information has become available on the biology, population ecology, and habitat requirements of *H. johnsonii* including clonality, growth, life history, dispersal mechanisms, population dynamics, physiological ecology, photobiology, phylogeny, genetics, and transplanting. A combination of published and unpublished studies and surveys were reviewed indicating that: 1) there has been no significant change in the overall geographic range of the species, 2) no reports of male flowers or sexual reproduction, and 3) genetic diversity is very low. Clonal growth, unique physiological attributes, and tolerance for a range of water quality conditions and UV light enables *H. johnsonii* to grow patchily distributed in a wide range of environments.

#### 2.3.1.1. Reproduction and clonality

Like all other seagrasses, *H. johnsonii* is clonal, which refers to plants that have many semi-independent units (ramets) acting together as a single organism (Cook, 1983). Reproduction is achieved primarily by asexual means (Fig. 1a). While all other species of seagrass reproduce sexually, there is still no evidence of sexual reproduction in *H. johnsonii*. All attempts to find seeds and seedlings have failed to detect any evidence of their occurrence (Jewitt-Smith et al., 1997; Hammerstrom and Kenworthy, 2003). Likewise, despite widespread sampling and surveys throughout the entire range of the species, no male flowers have ever been reported and confirmed. Female flowers, however, have been documented in both culture and nature (Eiseman and McMillan, 1980; Heidelbaugh et al., 2000). They are common and often very abundant (Heidelbaugh et al., 2000). They have been observed throughout the entire range of the species during all times of the year, but no consistent patterns of spatial or temporal distribution have been observed or reported.

Based on morphological, anatomical, and phylogenetic information (see discussion below), *H. johnsonii* is most closely related to *Halophila ovalis*, a dioecious species which reproduces sexually (Posluszny and Tomlinson, 1990; Freshwater, 1999; Waycott et al., 2002). Based on this apparent relationship between the two species, *H. johnsonii* is presumed to be dioecious. Although male flowers have never been observed, it is not possible to completely rule out their existence and the potential for sexual reproduction. They may occur cryptically in isolation or in the vicinity of females. They may be extremely rare, or they may express themselves only at night, as was the case in a related species, *Halophila hawaiiiana* (Herbert, 1986). The uncertainty regarding the existence of male flowers coincides with the fact that *H. johnsonii* seedlings have never been observed; they should not have been missed in the many benthic surveys that have been conducted throughout the species' range.

By comparison, the congeneric *Halophila decipiens* reproduces prolifically by seed and re-establishes populations annually in the same habitat as *H. johnsonii* (Kenworthy, 2000; Hammerstrom et al., 2006). Relative abundance of *H. decipiens* is nearly always an order of magnitude higher than *H. johnsonii* (Kenworthy, 2000, 1992; Virnstein et al., 1997; Virnstein

and Morris, 2007). Since rates of asexual reproduction and clonal growth rates of *H. johnsonii* and *H. decipiens* are nearly identical, the absence or rarity of sexual reproduction is likely contributing to the large difference in abundance between the two species and to the rarity of *H. johnsonii* (Kenworthy, 1992; Bolen, 1997). An important difference between the two species is that *H. johnsonii* is perennial, while *H. decipiens* behaves as an annual plant (Kenworthy, 2000).

Despite the importance of clonality and the absence or rarity of male flowers, it seems unusual that there should be so many female clones and so much energy put into the possibility of sexual reproduction. Until recently, it was unknown whether the pistillate flowers of *H. johnsonii* could produce haploid egg cells that could potentially be fertilized by males. Megagametogenesis, the process of pistil development in sexually reproducing flowering plants, is required to generate a functional haploid gametophyte for fertilization. A recent study by York (2005) demonstrated that meiosis and megagametophyte development does occur in *H. johnsonii* and that sexual reproduction could take place in the presence of pollen. This precludes the suggestion by Eiseman and McMillan (1980) that *H. johnsonii* may reproduce by apomixis. Apomicts do not undergo meiosis and no haploid cells are formed in the ovaries. Therefore, the search for male flowers and research directed at confirming fertilization potential and viable seed formation should be a high priority in the future.

While sexual reproduction of *H. johnsonii* remains somewhat of a mystery, reproduction by asexual means and clonal growth is well understood. Asexual reproduction occurs when rhizome apical meristems divide and form new leaf pairs, flowers, or rhizome apices (Posluszny and Tomlinson, 1990) (Fig. 1a). On average, new meristems are formed on rhizomes every 2 to 4 days (Kenworthy, 1997; Bolen, 1997). The species spreads and clones expand in local space by rhizome extension and leaf pair formation, eventually forming high density “patches.” Widely spaced patches, usually on the order of 1-20 square meters in size, are the most commonly encountered feature of *H. johnsonii* meadows (Virnstein et al., 1997; Kenworthy, 1997, 2000; Virnstein and Morris, 2007; Kenworthy, 2003). Patches can expand rapidly, however, increasing in area at a maximum rate of one square meter per month (Kenworthy, 2003) leading to coalescence of adjacent patches and the formation of larger meadows. To date, the largest reported contiguous meadow was observed in Lake Worth Lagoon and estimated to be 30 acres (Kenworthy, 1997).

Patches can also disappear rapidly. Sometimes they will disappear for several years and then re-establish: a process referred to as “pulsating patches” (Heidelbaugh et al., 2000; Virnstein and Morris, 2007). In the absence of sexual reproduction, one possible explanation for the pulsating patches is dispersal and re-establishment of vegetative fragments, a process which commonly occurs in aquatic plants and has been demonstrated in other seagrasses (DiCarlo et al., 2005), and was also recently confirmed by experimental mesocosm studies with *H. johnsonii* (Hall et al., 2006). *Halophila johnsonii* is a shallow rooted species and vulnerable to uprooting by wind waves, storm events, tidal currents, bioturbation, and motor vessels. It is also vulnerable to burial by sand movement and siltation (Heidelbaugh et al., 2000); all mechanisms capable of disturbing patches and creating clonal fragments for dispersal. Hall et al. (2006) showed that drifting fragments of *H. johnsonii* can remain viable for 4 to 8 days, during which time they can settle, root, and grow. Fragments could drift several kilometers under the influence of wind and tidally driven circulation, providing potential recruits for dispersal and new patch formation.



Fragments are regularly observed either drifting or entangled in drift algae, which behave like tumbleweed and can transport the fragments long distances. In the absence of sexual reproduction, these are likely to be the most common forms of dispersal and patch maintenance.

Clonal plants are, to varying extents, physiologically integrated (Magda et al., 1988). Consequently, resource-starved ramets of clonal plants may be supported by unstressed ramets (Noble and Marshall, 1983; Slade and Hutchings, 1987; Watson, 1984; Oborny et al., 2001). *Halophila johnsonii* exhibits clonal integration; however, it does not significantly modify carbon translocation to support specific ramets (Dean, 2002; Dean and Durako, In Press). Carbon sharing among *H. johnsonii* leaf pairs (ramets) in response to shading stress suggests that plants (genets) with shaded ramets do not exhibit compensatory increases in carbon per gram dry weight compared to plants with un-shaded ramets. In addition, there is no significant directionality in photosynthate allocation to ramets with respect to age or shading. Photosynthate is allocated to ramets proportional to their proximity to the source ramets rather than because of the condition of neighboring ramets, and the amount of allocation decreases rapidly with increasing distance. In short, the physiological strategy exhibited by *H. johnsonii* indicates that it makes surplus photosynthate available to all ramets, preferentially to neighboring ones, but does not selectively supply resources to stressed ramets (Dean and Durako, In Press). This is logical because the fast turnover and short-lived deterministic leaves of *H. johnsonii* may result in no advantage being gained by selectively supporting ramets based on condition or age. Strategies employed by larger seagrasses contrast with this strategy. Larger seagrasses have slower turnover rates that exhibit facultative and directional physiological support for light-stressed ramets (Tomasko and Dawes, 1989).

Observations suggest that *H. johnsonii* exploits unstable environments or newly-created unvegetated patches, with minimal resources allocated to the holding of space (the concept of pulsating patches applies here). Thus, selective support of a stressed ramet by this fast-lived plant could be disadvantageous when new growth is critical. By exhibiting fast-growth and support for all local ramets, *H. johnsonii* may exploit areas in which it could not otherwise compete. It may quickly recruit to locally uninhabited patches and through prolific lateral branching and fast horizontal growth, move out once conditions become unfavorable. While these attributes may allow *H. johnsonii* to compete effectively in periodically disturbed areas such as shallow intertidal fringes, if the distribution of this species becomes limited to stable areas it may eventually be out-competed by more stable-selected plants represented by the larger bodied seagrasses (Durako et al., 2003). In addition, coupled with this species' low capacity for storage and limited physiological integration, vegetative growth over large unsuitable patches may be unlikely, and its ability to recover from widespread habitat loss may be limited. The clonal and reproductive growth characteristics of *H. johnsonii* combine to explain its patchy, non-contiguous, and temporally fluctuating distribution.

### **2.3.1.2. Life History and Population Biology**

The apparent absence of sexual reproduction suggests that the life history and maintenance of *H. johnsonii* populations is exclusively dependent on asexual reproduction and clonal growth dynamics. Growth and the occupation of space, as well as the dispersal of the species, depend on the division of apical meristems. The divisions and subsequent differentiation of meristems

(meristem dependence) into the various attributes of the ramets are the foundation of growth and productivity in all seagrasses (Tomlinson, 1974). *H. johnsonii* grows by division of apical meristems on horizontal rhizomes which branch, forming leaf pairs, female flowers, and new lateral branches (Fig. 1b). Apical meristem densities can reach hundreds to thousands per square meter (Kenworthy, 1997; Heidelbaugh et al., 2000). This is orders of magnitude higher than most other larger bodied species of seagrass and provides an extraordinary vegetative growth potential. While apical meristems are producing new leaf pairs, they spread horizontally at the front or outside perimeter of a patch. At the same time, older leaf pairs and their rhizomes (either at the center of a patch, or at the rear of a migrating patch) senesce, die, and disintegrate. Even though an individual leaf pair is not motile and has a relatively short life span, the growing apex of a clone continues to move along the bottom. Therefore, at any given point in space a patch of *H. johnsonii* may or may not be constantly present. Given the reported apical densities, the rapid rhizome elongation and patch expansion rates, and the short turnover time of leaf pairs, it is not surprising that there are large spatial and temporal variations in *H. johnsonii* patch distribution and abundance.

The leaf pairs are determinate and live weeks or a few months, at most, if undisturbed, but natural mortality rates are not precisely known for either the leaves or the apical meristems which are, theoretically, immortal. The lateral branches formed on the nodes either abort, remain suppressed, or grow. Physiological and environmental processes controlling branching and growth are not well understood. Rates of node formation and branching, however, are reported to be on the order of one node every 3 to 9 days with 50% to 90% of the nodes forming lateral branches, depending on whether the node is on a primary rhizome axis or a branched rhizome (Bolen, 1997; Kenworthy, 1997; Richmond et al., 2006).

Rhizomes can elongate at rates approaching  $0.5 \text{ cm} \cdot \text{d}^{-1}$  (Bolen, 1997; Kenworthy, 1997), and when combined with prolific branching, individual patches (clones) can expand at extraordinary rates, ranging from  $0.3$  to  $1.0 \text{ m}^2$  per month (Kenworthy, 1997; 2003; Greening and Holland, 2003). Whole patch disappearance (mortality) has frequently been reported, as has patch recolonization (Heidelbaugh et al., 2000; Virnstein et al., 1997; Virnstein and Morris, 2007; Kenworthy, 2003; Greening and Holland, 2003). Mortality, or the disappearance of patches, can be caused by a number of processes, including burial from bioturbation and sediment deposition, erosion, herbivory, dessication, and turbidity. *Halophila johnsonii*'s canopy is only 2-5 cm tall and may be easily covered by sediments transported during storms or redistributed by macrofaunal bioturbation during the feeding activities of benthic organisms. Mesocosm experiments indicate that clonal fragments can only survive burial for up to a period of 12 days (W.J. Kenworthy, CCFHR, NOAA, Beaufort, NC, unpublished). Therefore, any explanation for longer time intervals between patch loss and patch recolonization, years in some reports (Virnstein and Morris, 2007), must take into account other processes, including widespread vegetative dispersal of clonal fragments (Hall et al., 2006). Development of a population dynamics model incorporating all of the parameters discussed in this section is an essential requirement for obtaining a better understanding of patch dynamics and survival of *H. johnsonii*.

### 2.3.1.3. Physiological Ecology

Observations of its distribution and the results of limited experimental work suggest that *H. johnsonii* has a wider tolerance range for salinity, temperature, and optical water quality conditions than *H. decipiens* (Dawes et al., 1989; Gallegos and Kenworthy, 1996; Durako et al., 2003; Kunzelman et al., 2005; Torquemada et al., 2005). *Halophila decipiens* is more stenohaline than *H. johnsonii*. *Halophila johnsonii* has been observed growing perennially near the mouths of freshwater discharge canals (Gallegos and Kenworthy, 1996), in deeper turbid waters of the interior portion of the Indian River Lagoon (Kenworthy, 2000; Virnstein and Morris, 2007), and in clear water associated with the high energy environments and flood deltas inside ocean inlets (Kenworthy, 1993, 1997; Virnstein et al., 1997; Heidelbaugh et al., 2000; Virnstein and Morris, 2007). This species can colonize and persist in high tidal-energy environments; it has been observed where tidal velocities approach the threshold of motion for unconsolidated sediments (35-40 cm\*sec<sup>-1</sup>). Intertidal populations of *H. johnsonii* may be completely exposed at low tides, suggesting high tolerance to desiccation and wide temperature ranges.

Torquemada et al. (2005) investigated the effects of salinity, temperature, and pH variations on growth, survival, and photosynthetic rates of *H. johnsonii*. While tolerance ranges are greater than those for *H. decipiens*, growth and survival are significantly affected by salinity, with maximum growth rates and survival obtained at 30 psu, a significant reduction in both growth and survival at higher and lower salinities, and no growth (i.e., mortality) at 0 and 60 psu (Torquemada et al., 2005). Similar responses to extreme variations in salinity have been observed in several seagrasses, although the extent of the salinity-tolerance ranges do vary. Salinity variations have caused significant reductions in growth (McMillan and Moseley, 1967; Walker, 1985; Walker and McComb, 1990) and survival of seagrasses (McMillan and Moseley, 1967; Pinnerup, 1980; Wortmann et al., 1997; Vermaat et al., 2000).

Torquemada et al. (2005) also found that salinity and temperature alter photosynthetic parameters in *H. johnsonii*. The parameters of photosynthetic efficiency curves, light-saturated photosynthesis ( $P_{max}$ ), and the photosynthetic efficiency at sub-saturating light ( $\alpha$ ) increase significantly up to an optimum of 40 psu, decreasing again at the highest salinities. The greatest decrease in photosynthetic activity occurs in freshwater. Dark respiration rates and compensating irradiance ( $I_c$ ) show minimum values at 40 and 50 psu, while light-saturation point ( $I_k$ ) is maximum at 30 – 50 psu. No effects of interactions between salinity and temperature were observed by Torquemada et al. (2005), although an increase of temperature alone was shown to produce an increase in  $\alpha$ ,  $P_{max}$ , respiration rates and  $I_k$ . An interaction between salinity and pH occurred only in the  $P_{max}$  response. In addition, reducing the pH increased  $P_{max}$  and  $\alpha$  significantly. There was a significant reduction in dark respiration with decreasing pH, but the opposite tendency was observed in the photosynthetic rates. Decreases in pH may lead to increased photosynthesis. Thus, recent global trends of ocean acidification and increasing CO<sub>2</sub> concentration may result in environmental conditions that are more conducive to this species (Orth et al., 2006).

While *H. johnsonii* is negatively affected by both extreme hypo- and hyper-salinity conditions, it does tolerate hypersaline conditions better than hyposaline conditions. Most other seagrasses, conversely, are thought to be more sensitive to increased salinity (Ogata and Matsui, 1965; Biebl and McRoy, 1971; Zieman, 1975; Adams and Bate, 1994; Doering and Chamberlain, 1998). The recent results of Torquemada et al. (2005) are consistent with earlier observations by Dawes et al. (1989) who observed positive responses for salinities between 15 and 35 and for temperatures between 10 and 30°C. They concluded that *H. johnsonii* showed a broader tolerance to temperature and salinity fluctuations than *H. decipiens*. The results of both studies indicate that *H. johnsonii* could be seriously affected by salinity variations produced by human activities, such as freshwater discharges through water management practices or brine discharges from seawater desalination plants. Interestingly, salinity changes do not seem to alter the tolerance of this species to other environmental factors, such as temperature or pH (Torquemada et al., 2005).

Since *H. johnsonii* grows intertidally, subtidally, and in the canopy of large bodied seagrasses, it is exposed to a range of light environments. In the subtidal environment it coexists with *H. decipiens*, making comparisons between the two species valuable. *H. johnsonii* and *H. decipiens* exhibit significant differences in photosynthetic characteristics that may, at least partially, explain the different depth distributions of these two species (Durako et al., 2003; Kunzelman et al., 2005). In situ rapid light curves (RLC) indicate that *H. decipiens* has lower maximum relative electron transport rates ( $RETR_{max}$ ) than *H. johnsonii* (Durako et al., 2003). The relatively lower  $RETR_{max}$  and  $E_k$  (the irradiance where ETR is approaching maximum) and generally higher  $\alpha$  (quantum efficiency) measured in situ for *H. decipiens* are in agreement with laboratory-based oxygen-flux measurements (Dawes et al., 1989). Both the oxygen and fluorescence results indicate greater photosynthetic light-efficiency, adaptation to lower irradiances, and inhibition by high irradiances for the more deeply-distributed *H. decipiens*, compared to the shallow-water *H. johnsonii*.  $RETR_{max}$  values decrease for intertidal *H. johnsonii* transplanted into subtidal beds, but they increase for both species when transplanted from subtidal to intertidal beds (Durako et al., 2003).

Although photoinhibition (or, more properly, down regulation in the RLC) was evident for both *H. johnsonii* and *H. decipiens* in situ (Durako et al., 2003), the irradiance levels for the onset of down regulation were much lower for the exclusively subtidally-distributed species ( $537\text{--}830 \mu\text{mol photons m}^{-2} \text{s}^{-1}$  for *H. decipiens* versus  $1785\text{--}2670 \mu\text{mol photons m}^{-2} \text{s}^{-1}$  for *H. johnsonii*). In a previous study using laboratory incubations, *H. johnsonii* did not exhibit photoinhibition at high light intensities, as did *H. decipiens* (Dawes et al., 1989). Within *H. johnsonii* populations, the deeper-growing subtidal plants exhibit greater down regulation than intertidal individuals.

Absorption spectra of leaf pigments (Fig. 2) reveal that *H. johnsonii* contains high levels of ultraviolet-absorbing pigments (UVPs) (Yakovleva and Titlyanov, 2001), which are characteristic of high-light adapted species (Franklin et al., 1996; Hader et al., 1998). UVP levels significantly increased within 4 days for subtidal-to-intertidal reciprocal transplants, indicating that photo-adaptation to higher UV radiation (or PAR) occurs rapidly in *H. johnsonii*, a distinct advantage enabling the plant to grow in shallow water. Absorption spectra for intertidal *H. johnsonii* acetone-soluble leaf pigments exhibited a dominant peak near 345 nm; this UV peak was 30% lower for subtidal plants. Pigment absorption spectra for *H. decipiens* lacked the 345 nm peak, and absorbances, normalized to leaf pairs, were lower across the

spectrum. The UVP levels in reciprocal transplants (Durako et al., 2003) also responded to decreasing irradiances in a manner similar to the patterns exhibited by other subtropical seagrasses, decreasing in response to reductions in PAR and UV (Dawson and Dennison, 1996; Detres et al., 2001). *Halophila decipiens* has generally lower Fv/Fm (photosynthetic efficiency, or potential quantum yield, Durako et al., 2003) values compared to *H. johnsonii*, and very low UVP absorbance (Fig. 2). The lack of UVPs may contribute to *H. decipiens*' high mortality when transplanted to shallow sites.

The absorption spectra for acetone extracts of *H. johnsonii* leaves exhibited a peak of 343-348 nm, which could be indicative of Mycosporin-like amino acid (MAA) or flavonoid absorption. Flavonoids are known to protect vascular plants from UV radiation and MAAs are thought to serve this same function in lower organisms (Sinha et al., 1998). MAAs have not, however, been isolated from any vascular plant. Flavonoids are the largest class of naturally occurring UV protecting compounds found in plants. The ability of flavonoids to absorb UVB light can prevent the DNA damage and photosystem damage induced by ultraviolet light (Hollosoy, 2002). Anthocyanins can serve a dual purpose by providing not only UV protection to plants but also are the pigments responsible for flower color. The UVP compounds found in *H. johnsonii* are a mixture of two flavone glycosides and three flavone acetylglucosides (Krzysiak, 2006). The production of flavones by angiosperms is not unusual. In fact, flavones are abundant in the plant kingdom, but the largest flavone-containing taxon is the angiosperms (Martens and Mithofer, 2005). Furthermore, the presence of flavone compounds within the *Halophila* genus has previously been observed. Sulphated flavones have been noted in *H. ballonis*, *H. engelmannii*, *H. stipulacea*, and *H. ovalis* (McMillan et al., 1980). In addition to *H. johnsonii*, glycosylated flavones have been observed in smaller leaved members of *Halophila* (McMillan et al., 1981). In support of their photo-protection role, the production of flavonoids appears to diminish in *H. johnsonii* when it is transplanted into deeper waters (Durako et al., 2003).

Decreases in Fv/Fm at high irradiance in *H. johnsonii* are due to decreasing Fm (the maximum fluorescence for dark acclimated tissue) rather than increases in Fo (the fluorescence for dark acclimated tissue) (Durako et al., 2003). These changes are indicative of non-photochemical quenching and photo-protection and possible changes to leaf optical properties under increased PAR conditions rather than destruction of photosystem II (PSII) reaction centers (photo-inhibition), and they are consistent with the observed increase in UVPs in the subtidal-to-intertidal transplants (Dawson and Dennison, 1996; Franklin et al., 1996; Gorbunov et al., 2001; Major and Dunton, 2002).

The results of Durako et al. (2003) indicate that photosynthetic tolerance to higher irradiances and presence of UVPs in *H. johnsonii* may allow this species to exploit the shallowest waters without competition from the closely-related, but UVP-lacking, *H. decipiens*. Survival of the shallowest *H. johnsonii* populations, however, may be threatened by other perturbations associated with intertidal fringe areas such as exposure to breaking waves, desiccation at low tides (Björk et al., 1999), and shoreline development activities. While *H. decipiens* may not be able to survive intertidally, its depth range extends much deeper than the 3-4 m maximum depth of occurrence observed for the threatened *H. johnsonii* (Kenworthy, 2000; Hammerstrom et al., 2006). At the lower depth limits, turbidity and other factors affecting light attenuation, such as

increased chlorophyll, are more critical. Thus, degradation of water quality due to human impacts, which would result in a more narrow depth range, may pose a more significant threat to *H. johnsonii* than continued increases in UV radiation.

#### 2.3.1.4. Genetics and Phylogeny

Detailed molecular studies of the genetic diversity of *H. johnsonii* have used DNA markers including: 1) Randomly Amplified Polymorphic DNA (RAPDs) (Jewitt-Smith et al., 1997; Freshwater, 1999) and, more recently, 2) Amplified Fragment Length Polymorphisms (AFLPs) (Wilson Freshwater, University of North Carolina Wilmington, Center for Marine Science, Wilmington, NC; and Michelle Waycott, James Cook University, Townsville, AU, unpublished). Freshwater (1999) compared *H. johnsonii* and *H. decipiens* from two locations within *H. johnsonii*'s geographic range and found that *H. decipiens* has much more genetic variation than *H. johnsonii* (Fig. 3a). He demonstrated that there was little or no genetic diversity for *H. johnsonii* within or between isolated patches at Sebastian Inlet and that there was also very little diversity detected among samples taken from seven other locations, ranging from the northernmost limit of the species at Sebastian Inlet to approximately 120 km south at Boca Raton (Fig. 3b). These findings are in contrast to the high level of RAPD banding pattern variability reported from an earlier study of *H. johnsonii* (Jewitt-Smith et al., 1997). In the Freshwater study, RAPDs detected the most diversity at two sites, Boynton Beach and Boca Raton, in the south central range of the species. Five unique RAPD phenotypes were recorded at these two locations and the genetic variation between phenotypes was highest at the Boynton Beach site. Because these two southern sites may represent unique and genetically distinct semi-isolated populations, both areas were selected and designated as critical habitat in an attempt to protect as much of the known genetic variation as possible.

Studies have shown that the AFLP method detects variation at a different level than that which can be detected using RAPDs. Since the earlier studies with RAPDs, attempts to use AFLPs have resulted in further confirmation of the relatively low genetic diversity in *H. johnsonii*. To date, more than 1,000 AFLP loci and 25 RAPD loci have been screened in a combined analysis (Wilson Freshwater, University of North Carolina Wilmington, Center for Marine Science, Wilmington, NC, and Michelle Waycott, James Cook University, Townsville, AU, unpublished). All show low variability compared to *H. decipiens* (an obligately sexual species-seeder); 80% of the bands of *H. decipiens* were variable and less than 25% of the bands were variable in *H. johnsonii*. There was no clear geographic pattern to the low variability observed in *H. johnsonii*. Using an unrooted genetic distance Neighbor Joining Tree, the combined data set suggests that there are "core" genotypes found in different locations representing a colonizing form of *H. johnsonii* (Fig. 4). Over time, the newer colonizing forms are accumulating a small amount of variation. This is supported by the observation of the population at Johns Island, just south of Sebastian Inlet. On the Neighbor Joining Tree, the Johns Island site has a quite distinct genotype cluster, despite physical proximity to the other sites that were sampled. It is hypothesized that this population is a re-colonizing population after being "lost" due to a disturbance which was documented during a concurrent study of transplanting and patch dynamics (Heidelbaugh et al., 2000). Current data supports a single or very limited origin to the

population and subsequent vegetative recruitment, over a very long time, evidenced through the accumulation of somatic mutations (variability) and only able to be detected using high resolution genetic markers.

Phylogenetically, *H. johnsonii* is located in what is being referred to as the *H. ovalis* complex (Waycott et al., 2002; Freshwater, 2004). The DNA loci sequenced thus far include the internal transcribed spacer regions of the nuclear-encoded ribosomal RNA genes (ITS) and the chloroplast-encoded *trnL* intron, intergenic spacer regions flanking the *trnL* gene, 5' intergenic region of the *matK* gene, and *matK* intron. Freshwater's (2004) results suggest that the species with complex phyllotaxy (*H. engelmannii*) terminate multiple early diverging lineages within the genus, while all species with simple phyllotaxy (e.g., *ovalis*, *decipiens*, *johnsonii*) are resolved in a single monophyletic clade (Fig. 5). Within the clade of species with simple phyllotaxy, there appears to be three major evolutionary lineages: 1) *H. capricorni*/*H. decipiens*; 2) *H. stipulacea*; and 3) *H. ovalis* complex (*H. ovalis*; *H. australis*; *H. hawaiiiana*; *H. johnsonii*, and possibly *H. minor*). The closest relative to *H. johnsonii* in this tree is an *H. ovalis* sequenced from Zanzibar. This is an indication that these lineages may have diverged over an evolutionarily short period and consequently there was not enough time for mutations to have accumulated between the two different divergence events that created the three lineages. This is one possible explanation for the overall *H. ovalis* complex as well. Freshwater postulates that what he is resolving in the *H. ovalis* complex is the relatively rapid and recent divergence of a number of different species. If this is the case, then there may not have been sufficient time for mutations to accumulate between the different lineage divergences, or for mutations to have accumulated in the particular loci sequenced, making the different lineages clearly distinct. The lack of homology between microsatellite loci in *H. ovalis* and both *H. hawaiiiana* and *H. johnsonii*, as well as the large amount of variation in RAPD banding patterns found when *H. ovalis* complex taxa are compared, suggests that the *H. ovalis* complex most likely represents distinct species.

In summary, *H. johnsonii* populations sampled thus far exhibit a very low level of genetic diversity and a high degree of clonality. This is consistent with the fact that there are no reports of male flowers or evidence of sexual reproduction, a major process responsible for genetic diversity in plant populations. The sources for the small amount of variation may be a combination of: 1) a founder effect left over from the original colonizing population, 2) past sexual reproduction events if males were ever present, 3) somatic mutations, and 4) vegetative fragment dispersal of clonal diversity. Although low diversity was detected, and given the fact that entire populations disappear and reappear at time scales of months to several years, even the smallest amount of diversity may be at risk of being lost from the species. Likewise new forms of diversity can be introduced into areas by clonal fragment dispersal. These mechanisms need to be confirmed with more sophisticated and precise co-dominant genetic markers. Analysis of dates of origin of the species are now underway using molecular clock calibrations of the genetic distance data and potential dates of origin of the genetic stock (Michelle Waycott, James Cook University, Townsville, AU, study in progress). The information provided by these advanced studies of genetic diversity will be valuable for the development of conservation and management strategies for this species and should be considered in future recovery efforts.

### 2.3.1.5. Distribution and Abundance

*Halophila johnsonii* is found only in southeastern Florida from near Sebastian Inlet (27.855906°, -80.453130°) to Virginia Key (27.747142°, -80.144286°) (Fig. 6). Since the last status review (Kenworthy, 1997), there have not been any reported reductions in the geographic range of the species. Two survey programs, one in the northern range of the species, between Sebastian Inlet and Jupiter Inlet, conducted by the St. Johns River Water Management District (SJRWMD) (Virnstein and Morris, 2007) and a second, recently initiated survey in the southern range of the species between Jupiter Inlet and Biscayne Bay (Kunzelman, 2007), have confirmed previous observations of *H. johnsonii*'s distribution and abundance (Kenworthy, 1997). Recently, however, the SJRWMD observed *H. johnsonii* 3 km north of the Sebastian River mouth on the western shore of the lagoon (27.884942°, -80.502986°) – a discovery that slightly extends the species' known northern range. *Halophila johnsonii* grows opportunistically in a patchy, disjunct distribution from the intertidal zone down to depths of approximately 3-4 meters in a wide range of sediment types, salinities, and in variable water quality conditions.

Since the listing, additional surveys funded by NMFS include a random point survey of Hobe Sound and the Jupiter Inlet designated critical habitat area, a random point survey in the region of the Indian River Lagoon that was subjected to intensive hurricanes between Sebastian Inlet and St. Lucie Inlet, and a random point survey of Biscayne Bay. All three of these surveys employed restricted random sampling designs to assess seagrass and macroalgal abundance and all found *H. johnsonii* was present consistently in only 2%-6% of sampling locations. In addition to these surveys, a 'data mining' project designed to obtain as much information as possible about *H. johnsonii* distribution from the records of local, county, state, and federal agencies and non-government organizations, has also been completed.

The following discussion of the species' distribution and abundance is broken into three sections. The first section includes information resulting only from the SJRWMD transects in the Indian River Lagoon and is designated as the "Northern Range Distribution." Data from the Hobe Sound/Jupiter Inlet random survey and the post-hurricane survey are not discussed further in any detail because analysis of the dataset is still underway. The second section includes information primarily resulting from the recently initiated transect study between Jupiter Inlet and Biscayne Bay, but also draws from the Biscayne Bay random point survey, and is designated as the "Southern Range Distribution." The third section discusses distribution information from the data mining project and is designated as such.

#### 2.3.1.5.a. Northern Range Distribution

Since 1994, the SJRWMD has monitored 73 permanent transects in the Indian River Lagoon in both summer (June-July) and winter (January-February) (Virnstein et al., 1997; Virnstein and Morris, 2007). Despite extensive ground-truthing since 1986 and monitoring all 73 transects throughout the Indian River Lagoon beginning in the summer of 1994 (a total of about 25,000 quadrats), *H. johnsonii* has never been found more than 3 km north of the Sebastian Inlet area. Thirty-five of the 73 permanent SJRWMD transects are located south of Sebastian Inlet, and data from this extensive monitoring effort show that *H. johnsonii* was found at 31 of those 35 transects (between Sebastian Inlet and Jupiter Inlet) during 1994-2007. Where it does occur, its



distribution is patchy, both spatially and temporally. It occurred in 7.1% (733 of the 10,387 quadrats derived from the 35 transects) of the 1- m<sup>2</sup> quadrats (Table 1). It was never observed at more than 23 of the 35 sites during any one season, and it was observed as infrequently as only once. At no single site was it present for all sampling periods. It frequently disappeared from transects only to reappear several months or several years later.

Along transects, *H. johnsonii* was routinely observed to be patchy, and percent cover varied along the length of the transects. It averaged only 4.3% cover over all sampling dates on the 35 transects within its range, and only 0.6% cover when averaged Lagoon-wide over all 73 transects monitored since 1994. Leaf pair density ranged up to 3,813 leaf pairs/ m<sup>2</sup> and most of the patches were smaller than 1 m<sup>2</sup>, and average shoot density was 52.3 shoots/ m<sup>2</sup>. These values are overestimates of the species' true relative abundance because 8 transects were located specifically in areas where *H. johnsonii* was known to be present.

*Halophila johnsonii* is a perennial species showing no consistent seasonal or year-to-year pattern in these surveys. Although perennial, it exhibited some winter decline. However, during exceptionally mild winters, as in the winter of 2004, *H. johnsonii* can maintain or even increase its abundance from summer to winter.

Although it is more commonly found in monotypic patches, *H. johnsonii* can also grow among low to moderate densities of *Halodule wrightii* and *Syringodium filiforme*, and in deeper water mixed with *H. decipiens* (Kenworthy, 1993, 1997, 2000; Virnstein et al., 1997; Virnstein and Morris, 2007). During monitoring in the northern range, *H. johnsonii* was observed to occur both in monotypic stands and mixed with other species. It co-occurred most commonly with *H. wrightii* (38% of quadrats) and *H. decipiens* (15%), rarely with *S. filiforme* or *H. engelmannii*, and never with *T. testudinum* or *R. maritima* alone. Its percent cover was not well correlated with the percent cover of any other species, the highest correlation being with *H. decipiens* ( $r^2 = 0.17$ ).

Depth of occurrence ranged from 0.03 to 2.5 m within transects. When data from all transects were combined, there was no correlation of *H. johnsonii* abundance with depth, despite observations that at some sites *H. johnsonii* occurred in either very shallow or very deep water. However, the deep edge at some transects was only 0.1 m; at other transects, it was 2.5 m. When all depths of occurrence were standardized (as percent of maximum depth of a transect), *H. johnsonii* was more abundant in the deeper parts of the transects. Most (78% or 574 out of 733) occurrences of *H. johnsonii* were at >70% of maximum transect depth; half were at >90% of maximum depth.

Although it can grow throughout a wide depth range, it often appears to be out-competed in the mid-depth ranges by the larger canopy-forming seagrass species (e.g., *H. wrightii*). Where the larger, canopy-forming species are absent, *H. johnsonii* can grow throughout the full seagrass depth range for the Indian River Lagoon.

*Halophila johnsonii* is rare but gregarious. It occurs in a wide variety of habitat types throughout the northern range of its distribution. It was found on intertidal wave-washed sandy shoals, on the flood deltas near inlets, in deep water, in soft mud, and near the mouths of canals and rivers where presumably water quality is sometimes poor and where salinity fluctuates widely.

### **2.3.1.5.b. Southern Range Distribution**

Prior to this review there was no detailed and systematic information on the distribution and abundance for most of the area in the species' southern range (Jupiter Inlet to Biscayne Bay). The 2002 survey of Biscayne Bay, the only large-scale random survey south of Jupiter Inlet, was designed specifically to provide a detailed assessment of *H. johnsonii*'s abundance and distribution near and at the southern limit (Durako, 2002), and to determine if the exact location of the southern limit at Virginia Key had changed significantly since 1974 (Eiseman and McMillan, 1980). In this study, 99 random sampling station locations and 17 additional locations where *H. johnsonii* had previously been observed were visited. *Halophila johnsonii* was only present in 6% of the random sampling stations and in only 29% of the repeat visit sites. One population was encountered south of Norris Cut (the longest record of continuous population persistence) and this location was only 0.6 km to the south and found in a small cove in Virginia Key.

In the summer of 2006, a permanent transect monitoring program was implemented in the southern half of *H. johnsonii*'s distribution, and was a collaboration between NMFS and the Florida Fish and Wildlife Research Institute (FWRI). The survey was designed specifically to monitor *H. johnsonii*. Thirty sites were selected from a list of potential locations based on the following criteria: 1) Transects were implemented only where *H. johnsonii* had been observed at least once in the past (suggesting that the environmental conditions were suitable), 2) transects were to be distributed as equally by distance from north to south as possible, and 3) transect sites were preferentially selected in an effort to represent as many different habitat types as possible, though they were not necessarily represented equally.

Of the 30 sites where transects were established, 14 were located in wider areas of the lagoon, 12 were located in narrower canal locations, and 5 were located near inlets. There was a fairly even distribution among the east and west bank of the Intracoastal Waterway (ICW) and sites located on islands in the ICW. Transect sites also varied by shoreline type, including rocky shore, mangrove, rip-rap, and seawall. One transect site was also set-up in each of the designated critical habitat areas located within the southern range.

Transects were sampled in summer 2006, and again in the winter 2007. In summer 2006, *H. johnsonii* was present at 97% of the sites sampled (Fig. 7). The mean frequency of occurrence over all transects sampled was 37% and the mean Braun-Blanquet value was 0.40. *Halophila johnsonii* was only absent from one site in summer 2006. There was little difference in the species' frequency or abundance encountered between the summer and winter sampling period. The lower frequencies for *H. johnsonii* occurred at those sites where larger-bodied seagrasses like *T. testudinum* and *S. filiforme* were more abundant.

Neither mean abundance nor the frequency of occurrence of *H. johnsonii* varied significantly between wide lagoon sites and the narrow canal sites. And though there was a trend of higher frequency and higher abundance at inlet sites compared to others, these differences were not significant at the 95% confidence level (non parametric ANOVA equivalent). Also surprising, no significant trend was detected in the frequency or abundance of *H. johnsonii* among sites that differed by shoreline orientation (east bank of ICW, west bank of ICW, island in ICW) or shoreline type (seawall, rip-rap, sand, mangrove). No significant relationships were apparent between the physical parameters sampled and the abundance of *H. johnsonii*.

The southern range transect data support some of the conclusions drawn from previous studies and other surveys. This is a rare species; however, it can be found in relatively high abundance where it does occur. Based on the results of the southern transect sampling, it appears that although it is disjunctly distributed and patchy there is some continuity in the southern distribution, at least during periods of relatively good environmental conditions and no significant large-scale disturbances.

#### **2.3.1.5.c. Data Mining Project**

The Johnson's seagrass data mining project was designed to identify, collect, and compile both survey and biological data on *H. johnsonii* for the development of a GIS database to be used for tracking its distribution and abundance. The project had two objectives: 1) to catalogue and document as many known occurrences of *H. johnsonii* as possible by obtaining information from various federal, state, and county permit files, academic institutions, and environmental consulting agencies; and 2) to begin production of a detailed baseline distribution map using GIS (Hall, 2005). In addition to an exhaustive search for records of *H. johnsonii* through state and county permit files (Palm Beach County and Dade County Departments of Environmental Resource Management), the 2004 Data Mining Project compiled data from environmental consulting companies (PBS&J, Dial Cordy, and Miller Legg), as well as academic institutions and federal agencies. Most of the state, county, and federal permit files located, as well as some information received from environmental consulting agencies, contained incomplete data, particularly in regard to the exact locations of *H. johnsonii* observations. It was determined necessary to locate and visit as many of the sites as possible. One hundred and seventy-three sites were chosen for re-surveying and 167 of these sites were actually visited in November 2004. Exact coordinates, Braun-Blanquet estimations of seagrass cover, and a number of water quality parameters were recorded at each site.

The results from the data mining project (which included the data compiled from outside sources as well as data collected during the re-surveying) were combined with the updated northern transect survey data, the southern transect survey data, and all other distribution data from *H. johnsonii* studies that have taken place since the data mining project was completed. All data are illustrated in a detailed baseline distribution map for *H. johnsonii*. The 11 panels depict all of the compiled occurrences of *H. johnsonii* throughout its range to date (Figures 8-19). Please note that distribution data received from PBS&J includes their own data as well as data compiled from outside sources during a similar exercise PBS&J completed in 2000 (Gelber et al., 2000).

Any data seen on the maps labeled as “PBS&J” is data that was collected by them for the purpose of their study. Any data seen on the maps labeled as “PBS&J Areas and Transect Lines” are data they compiled from outside sources.

Map Figures 8 – 19 will be used in future gap analyses and combined with a population dynamics model to examine the continuity of the species distribution, gap distances, and species dispersal potential, and to evaluate what constitutes a definition of self sustaining populations for recovery criteria 2. The maps of the Johnson’s seagrass critical habitat areas are provided for illustrative purposes only. For the precise legal definition of Johnson’s seagrass critical habitat, please see the description in the final rule (65 FR 17786).

### 2.3.1.6. Habitat and Functional Value

Seagrasses have recently received increasing attention from scientists and managers because of the valuable functional roles they play in coastal ecosystems (Costanza et al., 1997; Larkum et al., 2006). Functions associated with seagrasses include nutrient recycling, detrital production and export, sediment stabilization, and provision of food and habitat for many stages of numerous marine species. Very little work has been done on the functional value of *H. johnsonii*, therefore, the functional roles of its closest relative, *H. ovalis*, and other *Halophila* spp. are also considered in this discussion.

The most well-known function of seagrasses is their role as habitat for numerous fishes and invertebrates. Some species spend their entire lives within seagrass beds and others utilize it only during certain stages of their life cycle (usually the postlarval and juvenile stages). Heidelbaugh (1999) conducted one of the only studies that examined benthic fauna associated with *H. johnsonii*. In this study, differences in benthic fauna among *H. johnsonii*, *H. wrightii*, and bare sand were compared on the flood tidal delta just inside Sebastian Inlet, FL. *Halophila johnsonii* beds yielded a total of 126 species (69 epifauna and 57 infauna), while 117 species were collected from *H. wrightii* beds and 99 species from bare sand. The most abundant infaunal organisms belonged to Nematoda while the most abundant epifaunal species were amphipods and tanaids. The majority of macrofaunal organisms consisted of decapod crustaceans (*Callinectes sapidus*), fishes (*Eucinostomus* sp.), and some gastropods (especially *Bursatella leachii*). Three hundred and twenty macrofaunal organisms were collected from *H. johnsonii* beds compared to 690 from *H. wrightii* beds and 78 from bare sand. These results reveal its resource value in that they demonstrate that *H. johnsonii* faunal communities are more ecologically similar to other seagrass species than to bare sand.

Habitat value studies have also been carried out for other species of *Halophila*. One study compared nekton densities among *H. engelmannii*, *H. wrightii*, and nonvegetated habitats and, similar to the results of the Heidelbaugh (1999) study, found higher densities in the seagrass habitats (King and Sheridan, 2006). Naked goby (*Gobiosoma bosci*), code goby (*Gobiosoma robustum*), bigclaw snapping shrimp (*Alpheus heterochaelis*), and blue crab (*Callinectes sapidus*) were particularly abundant in *H. engelmannii* beds. A study in Thailand examining the community structure and abundance of benthic animals in *H. ovalis* beds found 77 different taxa including 33 annelid polychaetes, 25 mollusks, 12 arthropods, and 4 echinoderms (Nakoaka et al., 2002). Pipefish (*Stigmatopora* spp.) were found in deeper waters (12 – 16 m) of Australia

where *H. ovalis* was present (Kendrick and Hayes, 2003). Scorched mussel (*Brachidontes exustus*), brown crown conch (*Melongena melongena*), mojarra (*Ecinostomas melanopterus*), permit (*Trachinotus falcatus*), and nurse sharks (*Ginglymostoma cirratum*) have all been found associated with *H. baillonii* beds in Belize (Short et al., 2006).

Seagrass beds are one of the primary nursery habitats because of their abundance of prey items as well as the protection they provide from predators (Zieman and Zieman, 1989; Heck et al., 2003). In Queensland, Australia, postlarval and juvenile stages of three commercially important species of prawn (*Metapenaeus bennettiae*, *Penaeus plebejus*, and *P. esculentus*) were all found associated with seagrass beds that included *H. ovalis* (Masel and Smallwood, 2000).

Rapid growth, high turnover rates, and labile tissues make *Halophila* spp. a good source of nutrition for several marine herbivores (Kenworthy et al., 1989; Lanyon, 1991; Preen, 1995; Bolen, 1997). In areas such as Thailand and Moreton Bay, Queensland, dugongs (*Dugong dugong*) preferentially feed on *H. ovalis* (Nakaoka et al., 2002; McMahon, 2003). The Florida manatee (*Trichechus manatus latirostris*) has been observed grazing on *H. johnsonii* near a power plant in Palm Beach, FL (J. Reid, Sirennia Project, U.S.G.S., Gainesville, FL, personal observation). Green turtles (*Chelonia mydas*) are known to eat several species of *Halophila* including *H. ovalis* in the Arabian Gulf (Hasbun et al., 2000; Kannan and Rajagopalan, 2004), *H. decipiens* and *H. hawaiiiana* in Hawaii (Russell et al., 2003), and *H. ovalis* in Queensland, Australia (Whiting and Miller, 1998). *Halophila* also provides nutrition for herbivorous fish. Through consumption, the stareye parrotfish (*Calotomus carolinus*) has the ability to control the abundance and distribution of short lived seagrass species such as *H. stipulacea* in Kenya (Mariani and Alcoverro, 1999). Even invertebrates such as the queen conch (*Strombus gigas*) (Thayer et al., 1984) and various species of harpacticoid copepods (Shimode and Shirayama, 2006) have been observed feeding on *Halophila* species.

Seagrasses have long been recognized for their ability to stabilize sediments. It was once assumed, however, that due to its small size and sparse biomass, *Halophila* spp. were not capable of stabilization (den Hartog, 1970). Fonseca (1989) proved this assumption incorrect using a surface-supplied, inverted seawater flume. He found the cumulative effect of *H. decipiens* in reducing sediment erosion was significantly greater than adjacent, unvegetated sand. The degree of sediment stability as compared to bare sand was equivalent to many of the larger seagrass species and was well above that of *S. filiforme*. It is hypothesized that the allocation of leaf biomass and rhizomes at the sediment-water interface is the primary physical basis for the significant sediment stabilization effects of *H. decipiens*. However, being close to the sediment surface also means that it can be buried more quickly. Therefore, even though sediment stabilization by *H. decipiens* is significant, it may only occur in a narrow range and duration of velocities relative to other larger seagrasses. The persistent presence of high density elevated patches of *H. johnsonii* on flood tidal deltas near inlets suggests that it is capable of sediment stabilization.

Seagrasses play an important role in nutrient cycling within systems and can act as both a source and sink for nutrients (Hemminga et al., 1991). Processes that lead to a loss of nutrients from the system include: exudation/leaching from living and dead plant material, export of sloughed leaves and leaf fragments, nutrient transfer by foraging animals, denitrification, and diffusion

from sediment. Processes that result in an increase of nutrients include: nitrogen-fixation, sedimentation, and nutrient uptake by leaves. It is the fluctuation of these processes that leads to interannual variations in net losses or net gains of nutrients, and, therefore, fluctuations in the productivity of seagrass meadows (Hemminga et al., 1991). Connell and Walker (2001) examined nutrient cycling associated with *H. ovalis* in the Swan-Canning estuary in Australia. They discovered that *H. ovalis* takes up and exploits nutrients from the system in the spring when external nutrient resources are in abundance. It then has the ability to store these nutrients and, when external nutrients in the water column are insufficient during the summer to support growth, translocate and utilize these stored internal resources. During these times of year, *H. ovalis* acts as sink for nutrients in the estuary. In contrast, there were large losses of *H. ovalis* biomass in the winter which provides a good source of nutrients in the estuary. The plant material then becomes nutritionally available to consumers after undergoing decomposition to either morphous particulate organic detritus or amorphous detrital aggregates (Robertson et al., 1982).

Bacteria mediate the recycling of nutrients and may be important in regulating the flow of energy from seagrass detritus to consumer organisms (Robertson et al., 1982). Studies in the Salt River Submarine Canyon at St. Croix, U.S. Virgin Islands show that *H. decipiens* is an important source of organic matter and detritus for the Canyon (Josselyn et al., 1983; Josselyn et al., 1986; Kenworthy et al., 1989). Despite its production being less than other seagrasses, *H. decipiens* has a fast turnover time and is a major source of primary production on the floor of the Canyon (Kenworthy et al., 1989). Disturbance and burial of plant material are important mechanisms influencing the disposition of organic matter (Williams et al., 1985; Josselyn et al., 1986). Burial of *H. decipiens* through wave action and animal activities increases the rate of detrital input and retains the detritus within the Canyon (Kenworthy et al., 1989).

Given the similarities between the morphology of other *Halophila* spp. and *H. johnsonii*, it is reasonable to assume that *H. johnsonii* has the same capabilities as these other species to provide important ecological functions and services to the coastal ecosystem of southeastern Florida. Conservation of *H. johnsonii* will not only maintain the diversity of the seagrass communities, but also the important biodiversity and biophysical characteristics of the entire ecosystem.

### **2.3.2. Five Factor Analysis:**

#### **2.3.2.1. Present or threatened destruction, modification or curtailment of its habitat or range:**

With the exception of trampling, all of the threats to the species identified in the original listing are still present. These include dredging and filling, siltation, shoreline construction and modification, prop scarring, altered water quality, and storm events. According to survey reports, the present geographic range of the southern and northern limits of the species has been stable for at least 10 years. The species distribution throughout its geographic range is extremely rare, patchy, disjunct, and temporally fluctuating. Temporal fluctuations suggest that self-sustaining populations are maintained by a complex process of patch dynamics and dispersal, but the factors influencing patch dynamics on temporal and spatial scales which maintain self-sustaining populations are still not well understood; especially with regard to dispersal,

recruitment, and the role of genetic diversity. There are still no male flowers reported and genetic diversity is very low. The absence of sexual reproduction, patchy discontinuous growth, low genetic diversity, and its small size make the isolated populations vulnerable to stochastic events and natural and anthropogenic disturbances. Although the trend analysis for the northern range of the species suggests that the populations are relatively stable and resilient to natural perturbations, there is insufficient monitoring data for the southern range to fully assess the status and trend of self sustaining populations and the species overall in at least 50% of its distribution.

### **Trampling**

Originally, trampling was considered a threat but since the last status review there has been no evidence presented to support this activity as a serious threat to the species.

### **Dredging and Filling; Siltation; and Construction**

Sediment resuspension, and siltation associated with coastal construction activities and filling negatively affect seagrass by decreasing water transparency, physically burying plants, and modifying tidal current flow such that the plants experience excessive currents beyond their threshold for erosion. Dredging also may increase water depth such that the benthic plant communities are unable to receive enough light to sustain net primary productivity and growth. The Army Corp of Engineers (COE) has federal authority over the issuance of dredge and fill permits. The COE's State (Florida) Programmatic General Permit Program (SPGP) authorizes permits for the construction of docks, boat ramps, piers, maintenance dredging, and the construction of other minor over-water structures. The SPGP has seen an increase in the number of permits authorized between 2000 and 2006 (based on data provided by the COE), except for periods when the U.S. Fish and Wildlife Service (FWS) was involved in litigation over the manatee (*Trichechus manatus latirostris*). Based on the continued increase in permits issued within the range of *H. johnsonii*, we believe dredge and fill, siltation associated with dredging and coastal construction activities, and shading and physical impacts from in- and over-water structures associated with SPGP permitted activities have increased and continue to be a threat.

### **Prop Scarring**

Prop scarring and propeller dredging are one of the most severe injuries seagrasses experience because they disturb the sediments and uproot seagrasses, damaging the leaves and the root/rhizome systems, as well as the apical meristems which are responsible for the growth and maintenance of a seagrass meadow. *Halophila johnsonii* is especially vulnerable to these disturbances because it is so shallow rooted. In 2000, there were 131,759 vessels registered within the range of *H. johnsonii* between Indian River County and Miami-Dade County, Florida (<http://www.hsmv.state.fl.us/dmv/vslfacts.html>). In 2006, the DMV registered 200,187 vessels, an increase of 52% in 6 years. We expect these numbers will continue to increase based on Florida's projected population growth of 18 million in 2006 to 25 million in 2025 ([www.propertytaxreform.state.fl/docs/eo06141.pdf](http://www.propertytaxreform.state.fl/docs/eo06141.pdf)). This projected increase in the population will likely lead to an increase in the number of registered vessels and therefore an increase in impacts caused by prop scarring, anchoring, and other associated motor vessel related impacts such as dock construction and maintenance, marina expansion, inlet maintenance dredging, and erosion and sediment resuspension from boat wakes.

### **Altered Water Quality**

Turbidity (suspended solids), color, nutrients and chlorophyll are water quality constituents which affect the penetration of light in coastal waters and are major factors controlling the distribution and abundance of seagrasses (Dennison et al., 1993; Kenworthy and Haunert, 1991; Kenworthy and Fonseca, 1996). Most of the increased color and turbidity values in *H. johnsonii*'s range are being delivered by high flows of fresh water discharged from water management canals, which also reduce the salinity of the lagoon. Turbidity and nutrients are also derived from wastewater and stormwater discharges, as well as from land runoff and subterranean sources. Storm events, such as hurricanes, and variations in climate (wet seasons vs. dry seasons and wet years vs. dry years) both affect water quality and the potential for impacts from stochastically driven events. Unless they are curtailed by water management and land use practices that curb or eliminate discharges and minimize inputs of sediments and nutrients into the lagoon, it is expected that many unfavorable water quality parameters have the potential to increase in concentration corresponding with future population growth and land use practices.

Based on a Trophic State Index (TSI) of ambient water quality obtained in the northern and central region of the *H. johnsonii* geographic range provided in a long-term monitoring program implemented by the St. Johns River Water Management District, overall estuarine water quality was assessed as mostly good (67%) (Winkler and Ceric, 2006). Only 28% of the stations sampled had fair water quality, while 6% had poor quality. Fifty percent of the sampled estuarine sites were improving, while 6% were degrading, so many more sites were improving than were degrading. Forty-two percent of the lagoon sites had an insignificant trend while 3% had insufficient data to determine a trend. There is a strong positive correlation between seagrass depth distribution and water quality which enables managers to predict where seagrasses will grow based on water quality and the availability of light. As water management experts have now become confident in the correspondence between water quality and seagrass depth distribution, they have begun establishing water quality targets for the Indian River Lagoon based on seagrass as an indicator (Steward et al., 2005). Given that at least half of the stations were indicating long-term improvements in water quality, it can be assumed that seagrass abundance should not be negatively impacted if water and land use management programs continue to be effective. For example, carefully controlling or reducing water flows from discharge canals will moderate salinity fluctuations and reduce turbidity, color, and light attenuation values. However, there may be localized degradation near urbanized sites with multiple water quality problems that are more difficult to manage, such as the vicinity of the St. Lucie Inlet where the discharges from Lake Okeechobee have had significant impacts on water quality and seagrasses (Becky Robbins, South Florida Water Management District, West Palm Beach, FL, personal communication).

There has not been a comprehensive assessment of water quality published or reported for the southern range of *H. johnsonii* similar to the SJRWMD study. However, personal communication with water quality experts at the South Florida Water Management District (SFWMD) (Dan Crean, SFWMD, West Palm Beach, FL) confirm that efforts are underway to synthesize water quality information and to gain a more comprehensive understanding of the long-term status and trends of water quality in *H. johnsonii*'s southern range. Of particular



concern is an assessment of the impacts of fluctuations in water quality corresponding with variation in climate, especially “wet years” versus “dry years” variation. Future recovery efforts should include close coordination with the SFWMD and county environmental management agencies in Palm Beach and Dade counties to evaluate the status and trends of water quality in these regions of the species distribution.

### **Storms**

Storms, especially tropical storms and hurricanes, can significantly affect estuarine water quality (Steward et al., 2006) and are thus a potential threat to *H. johnsonii*. However, while hurricanes can generate runoff conditions that decrease water quality (e.g., increase turbidity and color), they also produce conditions (wind setup and abrupt water elevation changes) that can increase flushing rates. Thus, the effects of storms can be complex. Between August 14 and September 26, 2004, four tropical storm systems (Charley, Frances, Ivan, and Jeanne) impacted the central Indian River Lagoon. The lagoon received between 72 and 83 cm of rainfall during a 2 month period which generated high stream and canal discharges, wind driven suspended sediments, and significantly reduced salinities and water transparency. In September, salinities in the central Indian River Lagoon segments where *H. johnsonii* occurred dropped from 30 psu or more to 15 psu, color increased from a low of 10 pcu to 100 pcu, and turbidity increased from 3 NTU up to 14 NTU. Evidence of the hurricanes’ physical effects on seagrasses (burial, no scour) was limited to just one of the more than 25 sites inspected. Within 2 to 3 months following the hurricane period, most parameters related to water transparency returned to or showed improvement over their pre-hurricane values (February–July 2004). Unseasonably low salinities (20 psu) and moderately high color (20 pcu) were observed through spring 2005, largely attributable to a relatively long residence time and a wetter-than-average spring season in 2005. By the end of the study period (July 2006), the central Indian River Lagoon showed two opposite seagrass trends that began before 2004: an increase in depth limit coverage, but a decline in coverage density. Also, within a limited reach of the central Indian River Lagoon, *R. maritima* increased as *H. wrightii* decreased. It is likely that the persistently low salinities (not color) in 2004–2005 affected the species composition and coverage density. The authors (Steward et al., 2006) concluded that seagrasses are resilient to the acute effects of hurricanes and underscored the need to reduce chronic anthropogenic effects on seagrasses. Furthermore, the post-hurricane random survey in the region of the Indian River Lagoon affected by the four hurricanes indicated the presence of *H. johnsonii* was similar to that reported by the SJRWM district transect surveys prior to the storms. While the species may disappear initially, it returns quickly (Virnstein and Morris, 2007). We expect that hurricanes and other storm events will continue to be a potential threat for the species, but it is uncertain as to whether the frequency and strength of storms will increase and what long-term impacts this can have on *H. johnsonii*. It does appear, however, that *H. johnsonii* is resilient to potential hurricane impacts observed thus far.

In summary, the threats consisting of present or threatened destruction, modification, or curtailment of its range, identified for *H. johnsonii* at the time of listing still exist today and we do not foresee an elimination of the threats.

#### **2.3.2.2. Overutilization for commercial, recreational, scientific, or educational purposes:**

Not applicable.

### **2.3.2.3. Disease or predation**

Although disease has been reported to affect other species of seagrass there are no documented reports of disease that would threaten the geographic range, abundance, and survival of the species by affecting dispersal, recruitment, or genetic diversity. Although herbivory is documented, predation pressure (herbivory) is still not considered a significant threat to the species.

### **2.3.2.4. Inadequacy of existing regulatory mechanisms**

It is unclear whether or not existing regulatory mechanisms are adequate because there are no quantitative and comprehensive data bases and programs to track and assess the effects of all local, state, and federal regulatory actions. Federal, state, and local laws, regulations, and conservation management plans provide for the protection and conservation of seagrasses and their habitats. The Fish and Wildlife Coordination Act (FWCA) and the National Estuary Program are just two of the federal conservation measures that protect seagrasses and their habitats. The FWCA provides the basic authority for United States Fish and Wildlife Service (FWS) and NMFS involvement in evaluating proposed water resource development projects. The FWS and NMFS coordinate and consult with various federal agencies to avoid, minimize, and mitigate impacts to seagrasses and their habitats from water development projects. The state of Florida's Environmental Resource Permit (ERP) program regulates dredging, filling, and construction activities in wetlands and other surface waters. The ERP program is designed to ensure that alteration of uplands, wetlands, or surface waters does not degrade water quality, cause flooding, or diminish habitat quality or quantity. Seagrasses are also specifically identified as essential fish habitat (EFH) pursuant to the Magnuson-Stevens Fishery Management and Conservation Act, and are incorporated into fisheries management plans which the South Atlantic Fisheries Management Council, NMFS, and the state of Florida use to manage and conserve fisheries habitat.

The ESA is currently the only law that provides specific protection for *H. johnsonii*. The purpose of the ESA is "to provide a means whereby the ecosystems upon which endangered species and threatened species depend are conserved, to provide a program for the conservation of such endangered and threatened species, and to take such appropriate steps to achieve the purposes of the treaties and conventions set forth in subsection (a) of section 2 of the Act." Section 7 of the ESA requires federal agencies to consult with NMFS and FWS to insure that any action authorized, funded, or carried out by the agency is not likely to jeopardize the existence of any endangered or threatened species or result in destruction or adverse modification of critical habitat. Plants, and especially threatened plants, are not afforded the same level of protection as fish or other wildlife species listed under the ESA (e.g., compare sections 9(a)(1) and 9(a)(2) of the ESA). In addition, no quantitative methods have been developed to assess extinction risk for use in the ESA section 7 context.

The major problem in assessing the effectiveness of regulatory measures is the fact that there are no quantitative and comprehensive programs which compile and track regulatory activities in a manner that can be used to assess the status and trends of impacts and the success of the required

mitigation projects for seagrasses in general, including *H. johnsonii*. Although these federal and state conservation measures are in place to protect and conserve seagrasses and their habitats, losses of seagrasses and their habitats are still being reported (Orth et al., 2006) and their long-term effects for the species are not fully understood. Methods should be developed to fully quantify the effectiveness of the existing regulatory measures to determine their relevance in regard to the species status.

#### **2.3.2.5. Other natural or manmade factors affecting its continued existence**

Comprehensive range-wide surveys of *H. johnsonii* suggest that it has a limited capacity to co-exist with other larger bodied seagrasses. This suggests that natural and manmade factors which affect the distribution and abundance of *T. testudinum*, *H. wrightii* and *S. filiforme* could also influence the distribution, abundance, and existence of *H. johnsonii* populations. Thus, regulatory and conservation measures intended to promote other seagrass species could have a direct effect on *H. johnsonii*.

At the time of the last status review and the formal listing, no consideration was given to global climate change or sea level rise. Since then, there has been considerable scientific, political, and public discussions about the potential impacts of climate change and sea level rise on coastal marine environments and seagrasses which have not been incorporated into the recovery plan. Recent scientific information suggests that these topics and their application to seagrass conservation and management should be addressed. Once the effects of climate change are evaluated, the agency can determine the relevance of this factor on the species status.

#### **2.4. Synthesis**

There has been no significant change in the northern or southern range limits of *H. johnsonii*. It continues to have the most limited geographic distribution of any seagrass in the world. The species distribution is rare, patchy, and disjunct throughout its range, and it has very low genetic diversity. No male flowers have ever been reported, and there is no evidence of sexual reproduction. However, female flowers are common and morphologically and physiologically capable of being fertilized if male pollen was available. It appears that populations are maintained exclusively by a complex process of vegetative fragmentation and dispersal. Populations disappear and reappear on both short- (months) and long-term (years) time scales. Factors influencing the process of population maintenance and dispersal are still not well understood and a population dynamics model coupling biology, ecology, and environmental factors affecting growth, reproduction, and mortality is needed in order to better understand how self sustaining populations are maintained.

Presently, it appears that the populations in the northern range of the species (Sebastian Inlet to Jupiter Inlet) are stable and capable of sustaining themselves despite stochastic events related to severe storms and fluctuating climatology. Longer-term monitoring data are needed to confirm the stability of the southern distribution of the species from Jupiter Inlet to Biscayne Bay. This comprehensive monitoring program is now underway and should be supported in the future.

There have not been any significant changes in regulatory actions or conservation measures in the past 5 years; however, there are no comprehensive databases available to assess whether conservation and regulatory actions are adequately protecting the species.

Based on our review, we conclude that *H. johnsonii* remains vulnerable to natural and anthropogenic factors and the species still meets the definition of “threatened” under the ESA because it is still likely to become an endangered species within the foreseeable future throughout its range. With the exception of trampling, all of the threats to the species identified in the original listing are still impacting the species’ status. These include dredging and filling, shoreline construction and modification, prop scarring, altered water quality, siltation, and storm events. There has been no improvement in the species’ status in terms of its risk of extinction since its listing. Finally, no state or local efforts to protect Johnson’s seagrass are ameliorating the impacts and threats to the species, even given Florida’s rigorous permitting program regarding projects that impact seagrasses generally. Florida has not listed or otherwise identified Johnson’s seagrass for specific protections.

We believe our review has complied with the statutory requirement of section 4 (c) (2) of the ESA.

### 3. RESULTS

#### 3.1. Recommended classification

- Downlist to Threatened
- Uplist to Endangered
- Delist (indicate reasons for delisting per 50 CFR 424.11):
  - Extinction
  - Recovery
  - Original data for classification in error
- No change is needed

#### 3.2. New Recovery Priority Number 7

**If applicable, indicate the Listing and Reclassification Priority Number (FWS only)**

- Reclassification (from Threatened to Endangered) Priority Number: \_\_\_\_\_
- Reclassification (from Endangered to Threatened) Priority Number: \_\_\_\_\_
- Delisting (removal from list regardless of current classification) Priority Number: \_\_\_\_\_

### 4. RECOMMENDATIONS FOR FUTURE ACTIONS

Many of the “Actions Needed” identified in the recovery plan on page IX have either been initiated or completed. Future research related actions should include: 1) continuing long-term monitoring in the southern range of the species distribution; 2) continuing development of a spatially articulated population model for evaluating patch dynamics, dispersal distances, and habitat requirements of self sustaining populations; 3) continuing experimental studies to gain a better understanding of the interactions between *H. johnsonii* and other seagrasses and evaluate

how the interactions may be affected by water management, regulatory actions, and land use practices in the species range; 4) continuing research to verify the potential for sexual reproduction in *H. johnsonii*; 5) continuing studies to further examine the genetic diversity of *H. johnsonii* and the implications of diversity indices for long-term conservation and management of the species; and 6) continuing experimental studies to evaluate seagrass transplantation as a tool to assist in the relocation and restoration of *H. johnsonii* populations.

In addition to the research needs, future actions should include a comprehensive evaluation of the existing regulatory mechanisms directly or indirectly applicable to *H. johnsonii*. Efforts should be directed toward coordinating with local, state, and federal agencies to develop a quantitative database to be used to assess how various regulatory actions (e.g., permitting overwater structures, regulating water discharges, and effectiveness of stormwater controls) are affecting the distribution and abundance of *H. johnsonii*. In conjunction with these activities, the Johnson’s Seagrass Implementation Team should evaluate if management practices and techniques are adequately protecting *H. johnsonii* habitat.

Recovery Criteria 1 has been achieved; however, the Johnson’s Seagrass Implementation Team should re-evaluate Recovery Criteria 2 and Criteria 3 to determine if they warrant revisions and if any new information suggests alternative criteria are more appropriate. Threats based criteria should be added to the Recovery Plan.

The Team should consider actions to develop an outreach program to better inform private and public sectors of the status of *H. johnsonii* and the required actions needed for long-term conservation of the species. Such actions can include an up-to-date Web page on the status of *H. johnsonii* available to the public and private sectors.

## 5. TABLES

**Table 1.** Total number of sites and quadrats with *Halophila johnsonii* (*Hj*) from 1994 to 2007. The average percent cover is calculated as the average of all sites within *H. johnsonii*’s range (not the seasonal average). Bottom panel shows summary of summer-winter comparisons of frequency of occurrence at transect sites and within quadrats from transects within *H. johnsonii*’s range from 1994-2007 (# = number of transects or quadrats with *H. johnsonii* present and **n** = total sample size).

| SEASON / YEAR | Total sites with <i>Hj</i> (out of 35) | Total quadrats sampled | Total quadrats with <i>Hj</i> | Average % cover | % Occurrence of <i>Hj</i> within quadrats |
|---------------|--|------------------------|-------------------------------|-----------------|---|
| Summer 1994   | 12                                     | 460                    | 31                            | 3.8             | 6.7                                       |
| Winter 1995   | 7                                      | 419                    | 8                             | 1.0             | 1.9                                       |
| Summer 1995   | 7                                      | 399                    | 9                             | 1.0             | 2.3                                       |
| Winter 1996   | 4                                      | 348                    | 5                             | 0.2             | 1.4                                       |
| Summer 1996   | 9                                      | 490                    | 17                            | 3.3             | 3.5                                       |
| Winter 1997   | 10                                     | 487                    | 29                            | 4.4             | 6.0                                       |
| Summer 1997   | 15                                     | 529                    | 56                            | 4.7             | 10.6                                      |
| Winter 1998   | 15                                     | 525                    | 30                            | 4.1             | 5.7                                       |

| Summer 1998          | 16  | 543                           | 58                                   | 8.2                    | 10.7   |
|----------------------|---|-------------------------------|--------------------------------------|------------------------|--|
| Winter 1999          | 9   | 525                           | 26                                   | 2.3                    | 5.0  |
| Summer 1999          | 10  | 483                           | 38                                   | 1.5                    | 7.9  |
| Winter 2000          | 7   | 429                           | 19                                   | 1.4                    | 4.4  |
| Summer 2000          | 14  | 504                           | 42                                   | 3.6                    | 8.2  |
| Winter 2001          | 9   | 441                           | 29                                   | 3.3                    | 6.6  |
| Summer 2001          | 14  | 519                           | 29                                   | 5.1                    | 5.6  |
| Winter 2002          | 11  | 410                           | 25                                   | 4.1                    | 6.1  |
| Summer 2002          | 12  | 457                           | 32                                   | 1.8                    | 7.0  |
| Winter 2003          | 8   | 69                            | 13                                   | 7.9                    | 18.8   |
| Summer 2003          | 14  | 483                           | 47                                   | 3.8                    | 9.7  |
| Winter 2004          | 11  | 70                            | 19                                   | 12.8                   | 27.1   |
| <b>SEASON / YEAR</b> | <b>Total sites with <i>Hj</i> (out of 35)</b> | <b>Total quadrats sampled</b> | <b>Total quadrats with <i>Hj</i></b> | <b>Average % cover</b> | <b>% Occurrence of <i>Hj</i> within quadrats</b> |
| Summer 2004          | 23  | 513                           | 82                                   | 7.4                    | 16.0   |
| Winter 2005          | 1   | 65                            | 1                                    | 0.9                    | 1.5  |
| Summer 2005          | 10  | 458                           | 21                                   | 3.0                    | 4.6  |
| Winter 2006          | 5   | 109                           | 7                                    | 3.9                    | 6.4  |
| Summer 2006          | 14  | 513                           | 45                                   | 6.0                    | 8.8  |
| Winter 2007          | 9   | 139                           | 15                                   | 9.3                    | 10.8   |
| <b>SUMMARY</b>       | <b>276 of 910</b>                             | <b>10,387</b>                 | <b>733</b>                           | <b>4.3</b>             | <b>7.1</b>                                       |

| <b>PARAMETER</b>                   | <b>SUMMER</b> |          |          | <b>WINTER</b> |          |          |
|------------------------------------|---------------|----------|----------|---------------|----------|----------|
|                                    | <b>#</b>      | <b>n</b> | <b>%</b> | <b>#</b>      | <b>n</b> | <b>%</b> |
| Transects with <i>H. johnsonii</i> | 170           | 455      | 37.4     | 106           | 455      | 23.3     |
| Quadrats with <i>H. johnsonii</i>  | 507           | 6,351    | 8.0      | 226           | 4,036    | 5.6      |

**Table 2.** Summary of surveys and studies included in the baseline distribution map shown in Figures 8 through 19.

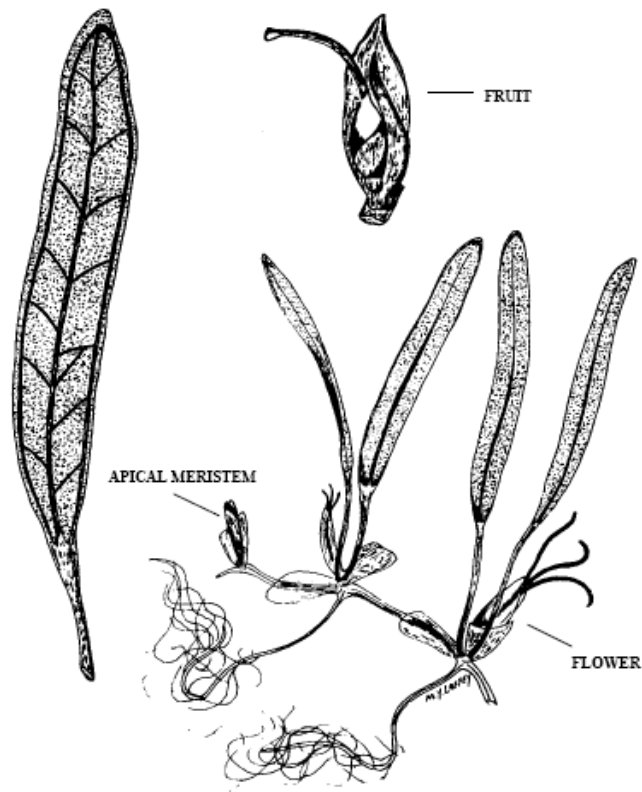
| ORGANIZATION AND PROJECT  | AS SEEN ON LEGENDS IN FIGURES 8-19 | SURVEY METHOD and YEAR   |
|---|------------------------------------|--|
| National Oceanic and Atmospheric Administration and Florida Fish and Wildlife Conservation Commission / Fish and Wildlife Research Institute - Hobe Sound and Jupiter Island Study performed to develop monitoring methodology for <i>H. johnsonii</i> .        | NOAA-FWRI(Hobe Sound/Jupiter Is)   | Random quadrat sampling using Braun-Blanquet to measure seagrass cover (2003)              |
| National Oceanic and Atmospheric Administration and Florida Fish and Wildlife Conservation Commission / Fish and Wildlife Research Institute - Indian River Lagoon Survey   | NOAA-FWRI (Northern Random)        | Random quadrat sampling using Braun-Blanquet to measure seagrass cover (2005)              |
| National Oceanic and Atmospheric Administration and Florida Fish and Wildlife Conservation Commission / Fish and Wildlife Research Institute - Southern Range Monitoring Project (Southern Transects)   | NOAA-FWRI (Southern Transects)     | Permanent transects using Braun-Blanquet to measure seagrass cover in quadrats (2006-2007) |
| National Oceanic and Atmospheric Administration – Study to evaluate the impact of overwater structure on underlying <i>H. johnsonii</i>   | NOAA Dock Study                    | Survey using Braun-Blanquet to measure seagrass cover in quadrats along transect (2007)    |
| National Oceanic and Atmospheric Administration and Florida Fish and Wildlife Conservation Commission / Fish and Wildlife Research Institute - Jupiter Inlet Survey performed to develop monitoring methodology for <i>H. johnsonii</i> .                       | NOAA-FWRI (Jupiter Inlet)          | Random quadrat sampling using Braun-Blanquet to measure seagrass cover (2003)              |
| Florida Fish and Wildlife Conservation Commission / Fish and Wildlife Research Institute - Compiled site surveys from data mining project.  | FWRI compiled site surveys         | Quadrat sampling (Compiled in 2006 with some data from previous years)                     |
| University of North Carolina Wilmington and Florida Fish and Wildlife Conservation Commission / Fish and Wildlife Research Institute – Detailed survey and assessment of the baseline distribution of <i>H. johnsonii</i> at its southern distributional limit. | UNCW-FWRI                          | Random quadrat sampling using Braun-Blanquet to measure seagrass cover (2003)              |

**Table 2. Continued.**

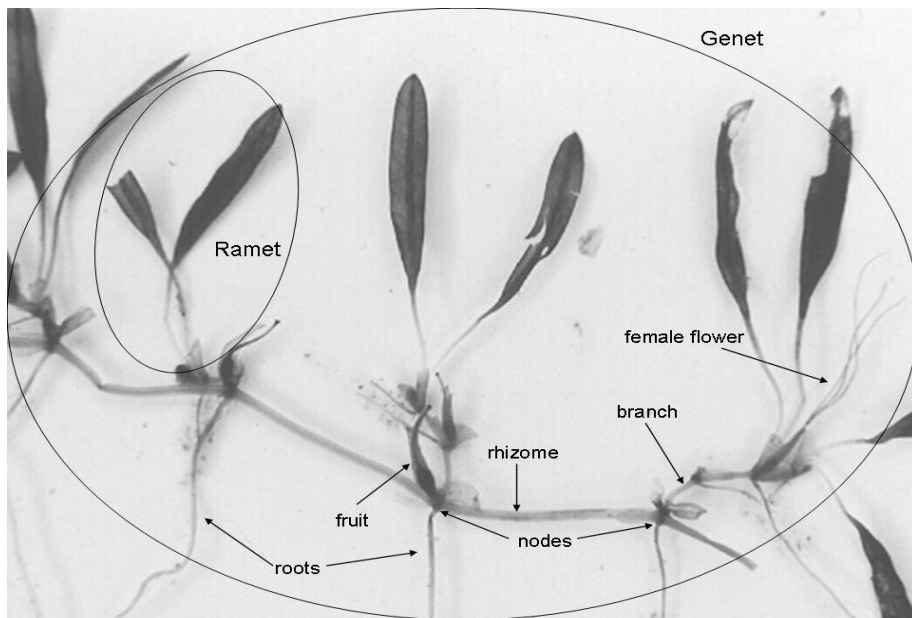
| ORGANIZATION AND PROJECT   | AS SEEN ON LEGENDS IN FIGURES 8-19            | SURVEY METHOD and YEAR  |
|--|---|---|
| St. Johns River Water Management District – Distribution and abundance of <i>H. johnsonii</i> in the Indian River Lagoon   | SJRWMD Transects                              | Permanent transect sampling using percent cover to measure seagrass (1994-2007)   |
| Post, Buckley, Schuh & Jernigan, Inc. – Survey to determine the distributional ecology of <i>H. johnsonii</i> in areas not previously inspected.   | PBS&J   | Transect surveys (2000)   |
| Post, Buckley, Schuh & Jernigan, Inc. – Distribution data compiled for the purpose of the report titled “The distributional ecology of the seagrass <i>Halophila johnsonii</i> .” Distribution data was received from St. Johns River Water Management District, South Florida Water Management District, Wildpine Ecological Laboratory, Florida Department of Environmental Protection, Florida Fish and Wildlife Conservation Commission, the U.S. Army Corps of Engineers, and the Broward County Department of Planning and Environmental Protection. | PBS&J Areas and Transect Lines                | Method and Year both vary   |
| Miller Legg and Associates, Inc. – Seagrass surveys around Spoil Island 15 near the Ft. Pierce Inlet North Causeway Bridge to establish appropriate location for placement of barges, at Ft. Pierce Bridge as a feasibility study for the Florida Department of Transportation, and in West Lake Park to document SAV changes after mitigation improvements.   | Miller Legg                                   | Site Inspection (2002, 2003, and 2001 respectively)   |
| Dial Cordy and Associates, Inc. – Seagrass surveys in areas of proposed dredging including: Palm Beach County Small Navigation Project, Port Everglades Dredge, and Miami Harbor Navigation Project.   | Dial Cordy                                    | Site Inspection (2004, 2001, and 2003, respectively)  |
| Wildpine Ecological Laboratory, Loxahatchee River District – These include an evaluation of seagrass communities in the southernmost reach of the Indian River Lagoon and a collaborative study with the SFWMD to map seagrass in the central embayment of the Loxahatchee River.  | Wildpine Ecol Lab - Loxahatchee River         | Transect survey with percent composition recorded (2000) and area perimeter survey measuring density (2004), respectively |
| Palm Beach County Department of Environmental Resource Management – Represents several studies/surveys conducted by PBD-DERM or contracted out to various laboratories or consulting agencies.   | PBC-DERM                                      | Seagrass surveys, methods vary (1999 – 2004)  |
| Palm Beach County Department of Environmental Resource Management – City of Lake Worth Lagoon Restoration Project  | City of Lake Worth Lagoon Wetland Restoration | Site Inspection (1999)  |



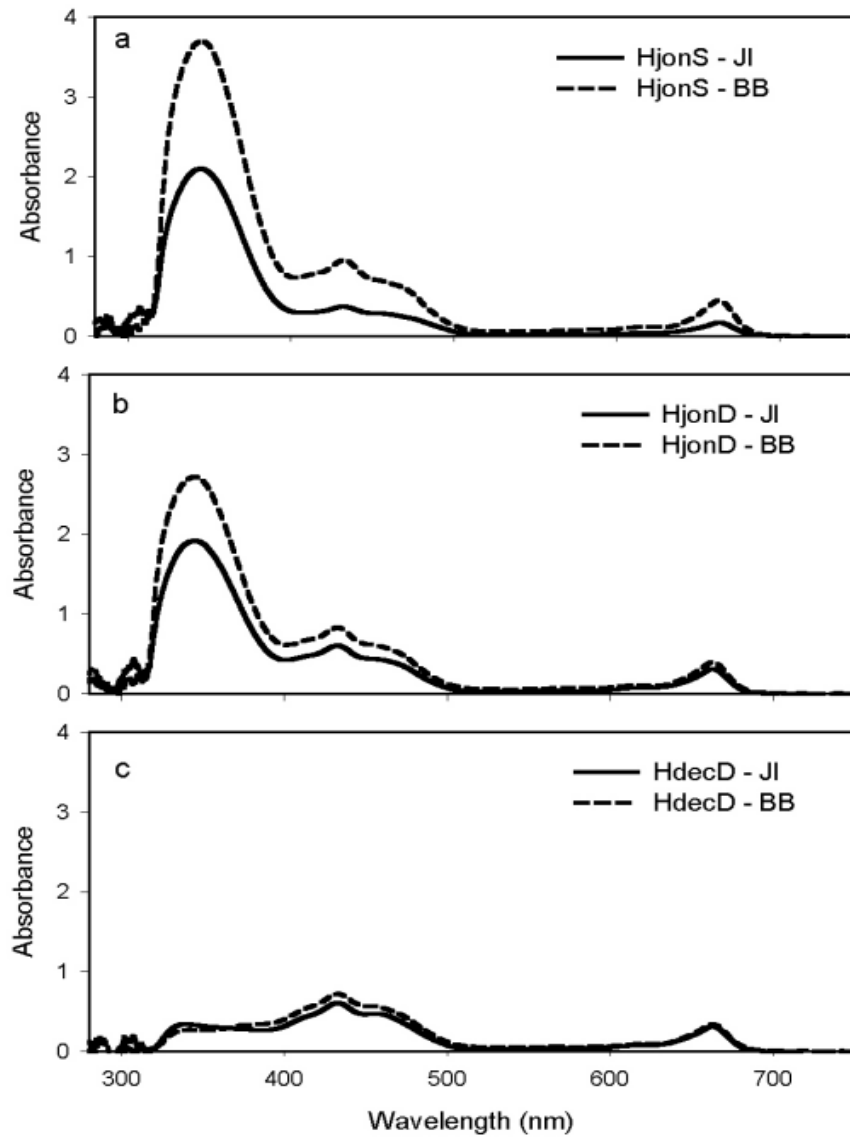
## 6. FIGURES



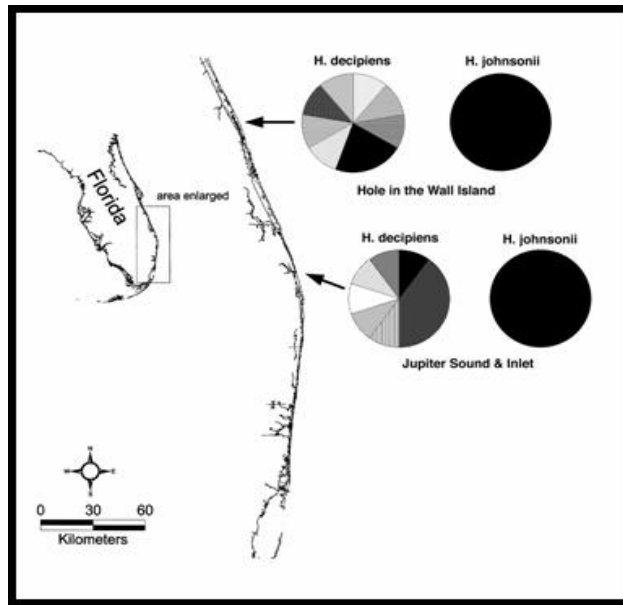
**Figure 1a.** *Halophila johnsonii*. Leaves are generally 2-5 cm long. Adopted from Eiseman and McMillan (1980).



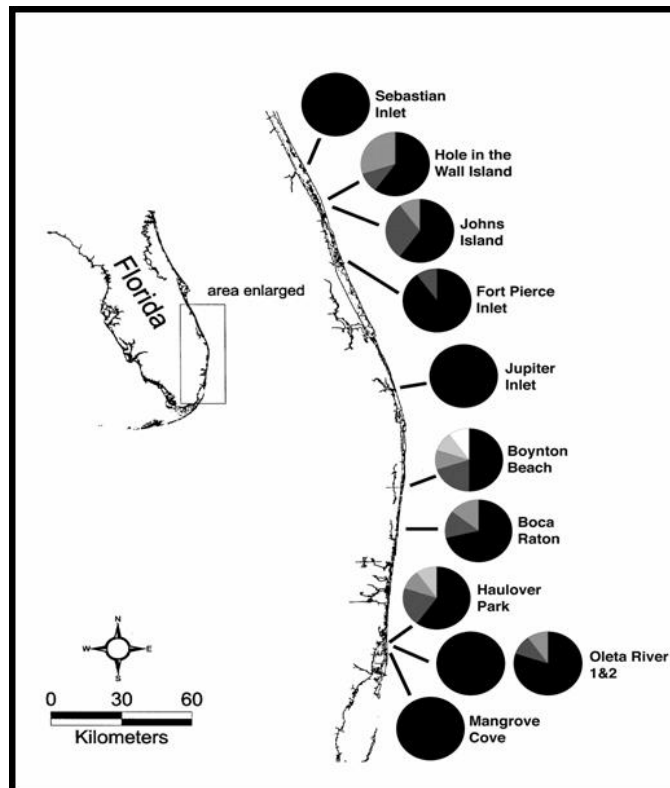
**Figure 1b.** Photograph of *Halophila johnsonii* showing the genet and individual ramets, the rhizome, a female flower, fruit, nodes, and lateral branching of rhizome.



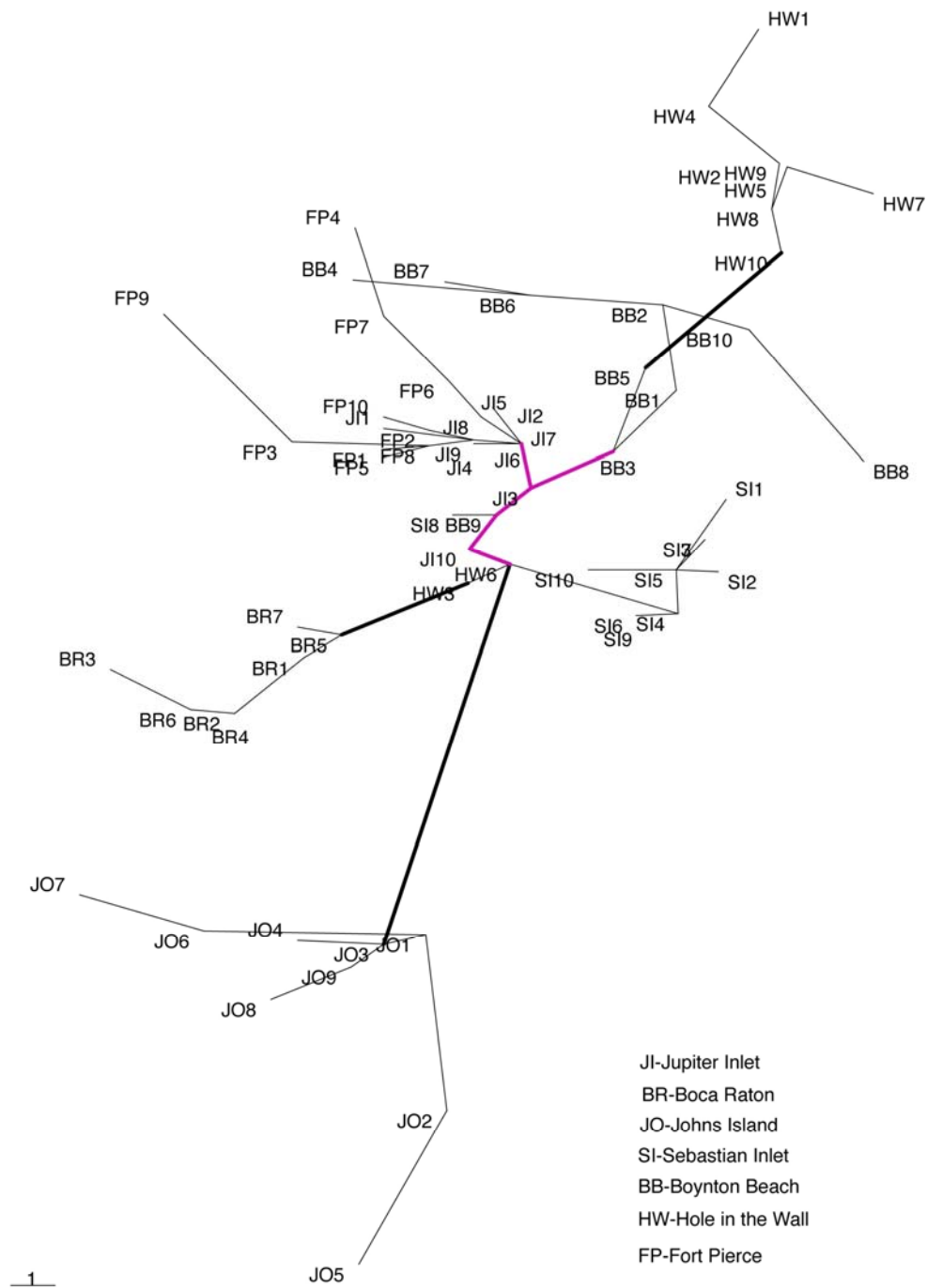
**Figure 2.** *Halophila johnsonii* (Hjon) and *Halophila decipiens* (Hdec). Mean absorption spectra of 90% acetone leaf extracts for a) intertidal Hjon, b) subtidal Hjon, and c) subtidal Hdec populations at Jupiter Inlet (JI, n=8) and Biscayne Bay (BB, n=4). S and D following name abbreviations indicate shallow or deep populations.



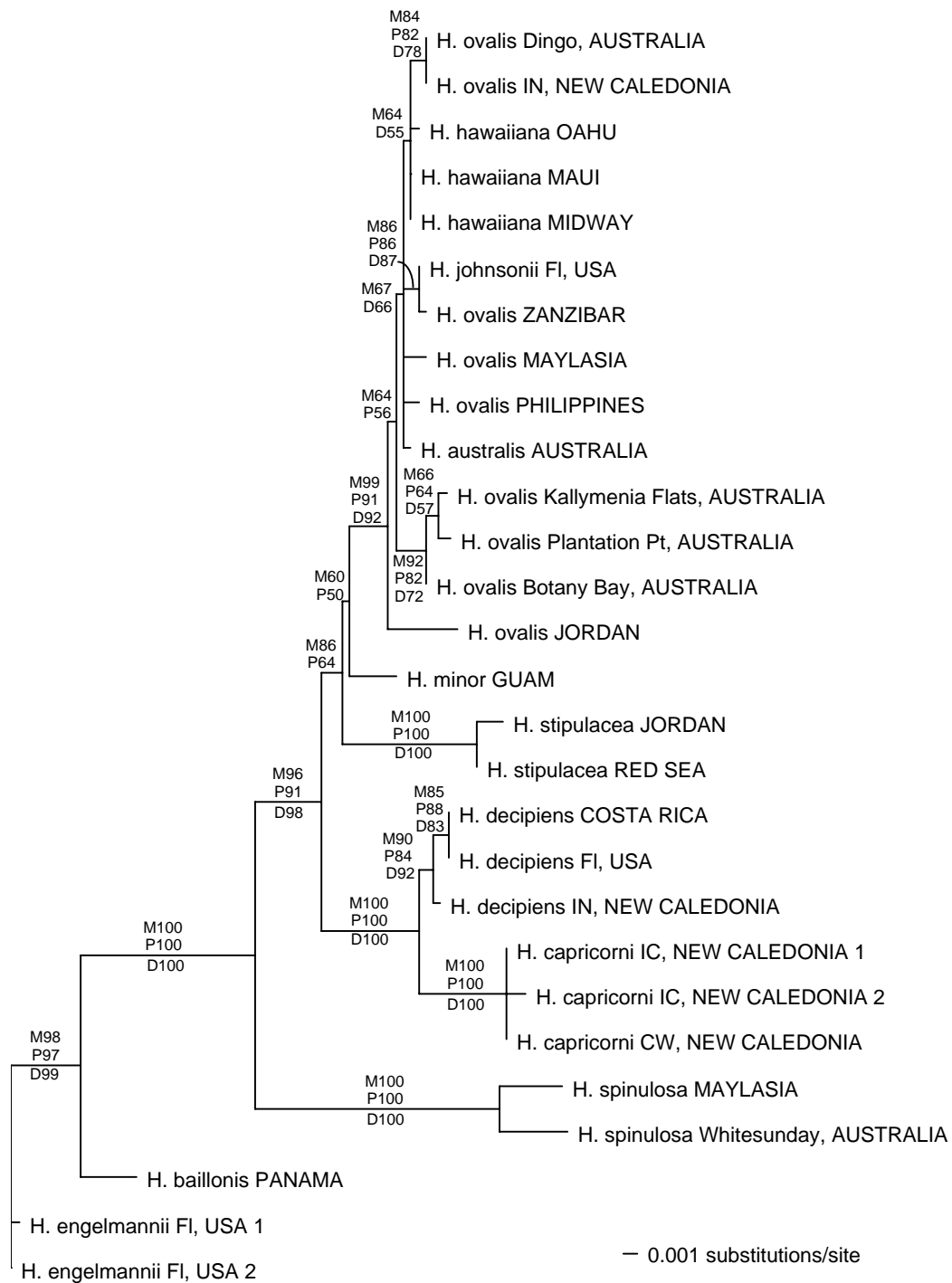
**Figure 3a.** Pie diagram showing the proportion of RAPD phenotypes found in *Halophila decipiens* and *Halophila johnsonii* samples from two sites. More genetic variation detected in *H. decipiens* than in *H. johnsonii*



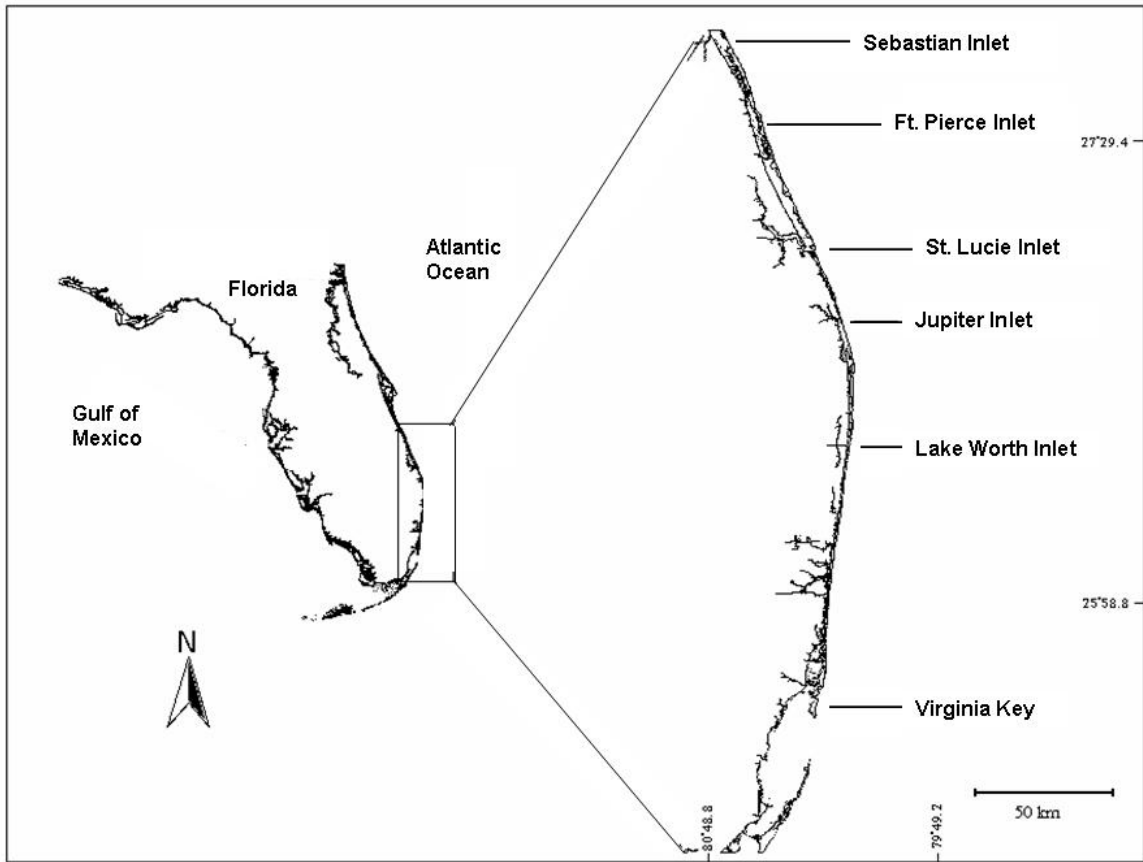
**Figure 3b.** Pie diagram showing the proportion of RAPD phenotypes for *Halophila johnsonii* samples from sites throughout its geographic range. RAPDs detect only a small amount of variation.



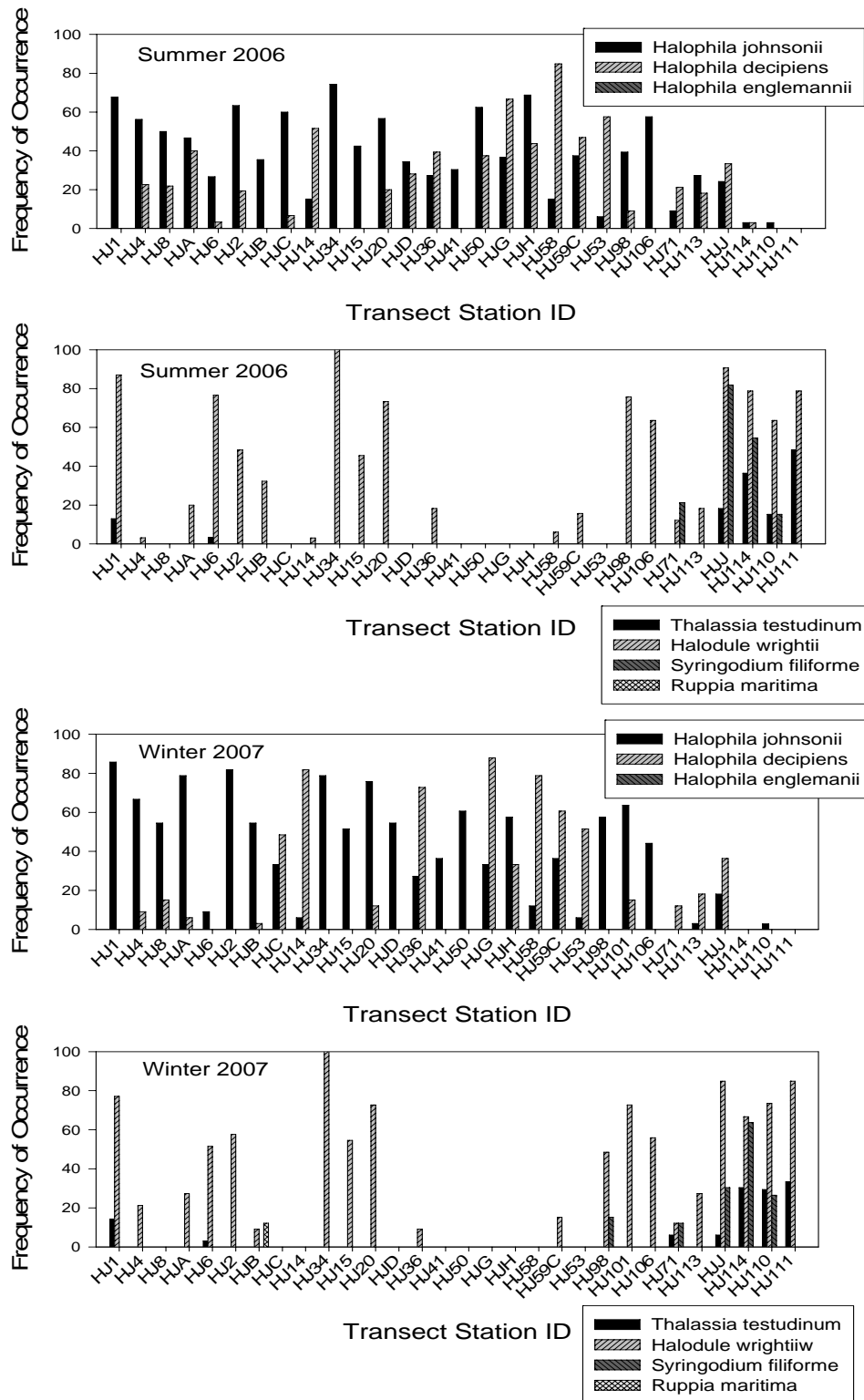
**Figure 4.** An unrooted genetic distance Neighbor Joining Tree by Michelle Waycott, James Cook University, Townsville, AU. (unpublished). The combined data set suggests that there are ‘core’ genotypes found in seven different locations representing a colonizing form of *H. johnsonii*. See pp. 12 of text for discussion.



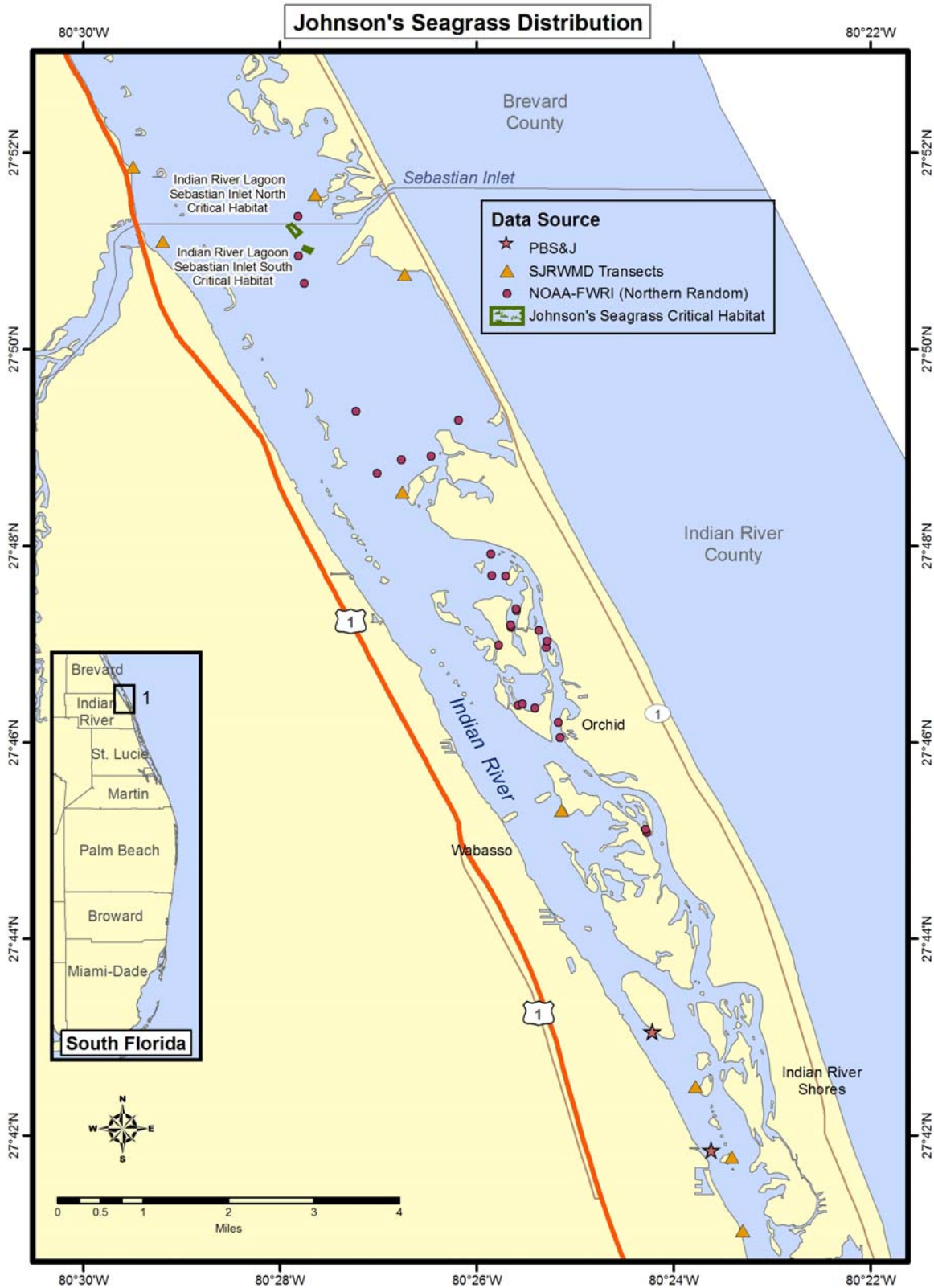
**Figure 5.** Maximum likelihood tree resulting from analyses of chloroplast-encoded *trnL* region sequences. The level of bootstrap support in maximum likelihood (M), parsimony (P), and distance (D) analyses are shown for resolved branches. See pp. 22 of text for discussion.



**Figure 6.** Geographic range of *Halophila johnsonii*: Sebastian Inlet to northern Virginia Key (Kenworthy 1997).

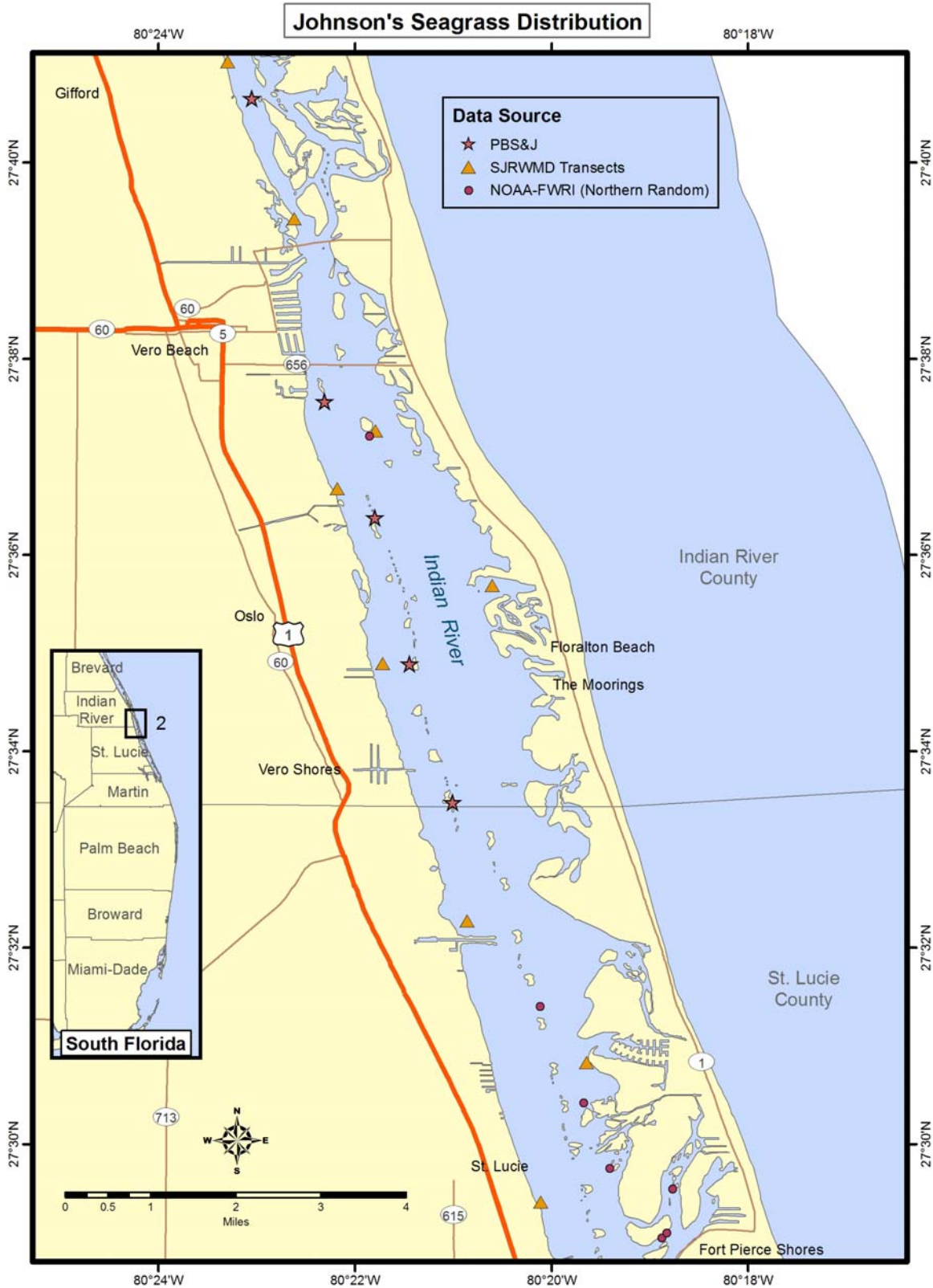


**Figure 7.** Frequency of occurrence for each of the seven seagrass species by transect station from northernmost (HJ1) to southernmost (HJ111) in Summer 2006 and winter 2007. Note that station HJ101 was added in the winter 2007 sampling.

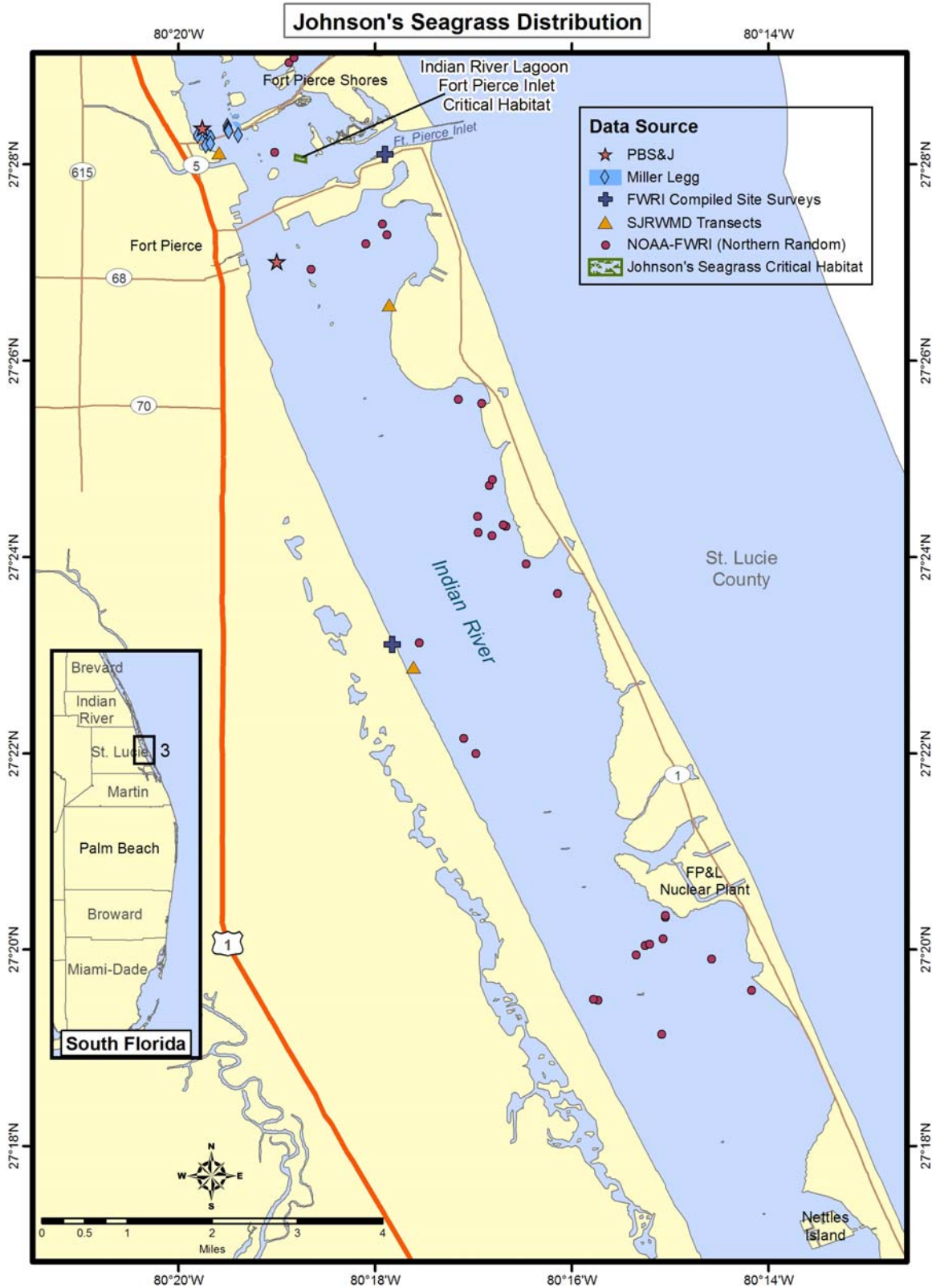


**Figure 8.** *Halophila johnsonii* distribution map 1. Symbols indicate confirmed presence of *H. johnsonii*. See Table 2 for further information about surveys.





**Figure 9.** *Halophila johnsonii* distribution map 2. Symbols indicate confirmed presence of *H. johnsonii*. See Table 2 for further information about surveys.



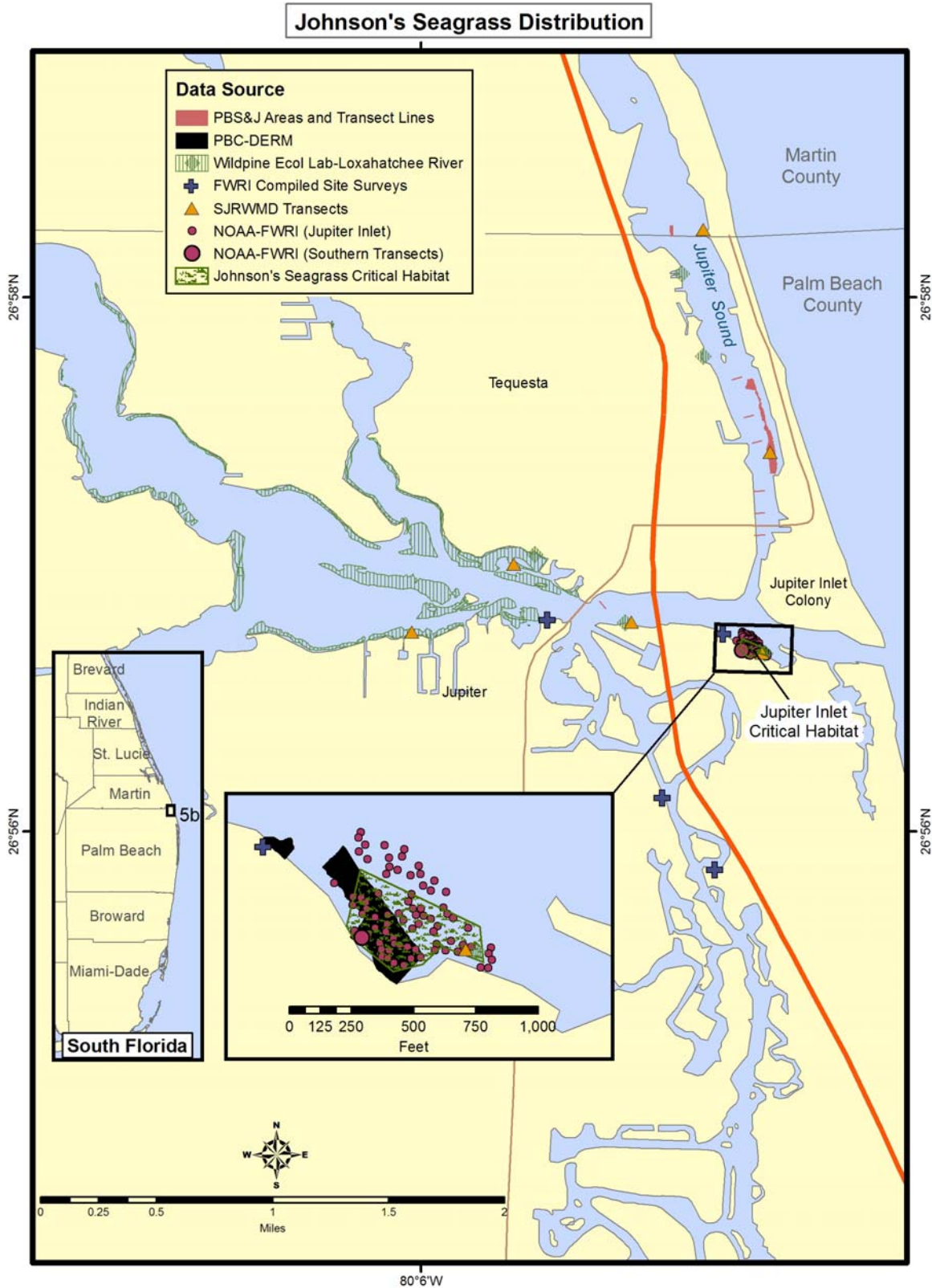
**Figure 10.** *Halophila johnsonii* distribution map 3. Symbols indicate confirmed presence of *H. johnsonii*. See Table 2 for further information about surveys.







**Figure 12.** *Halophila johnsonii* distribution map 5. Symbols indicate confirmed presence of *H. johnsonii*. See Table 2 for further information about surveys.

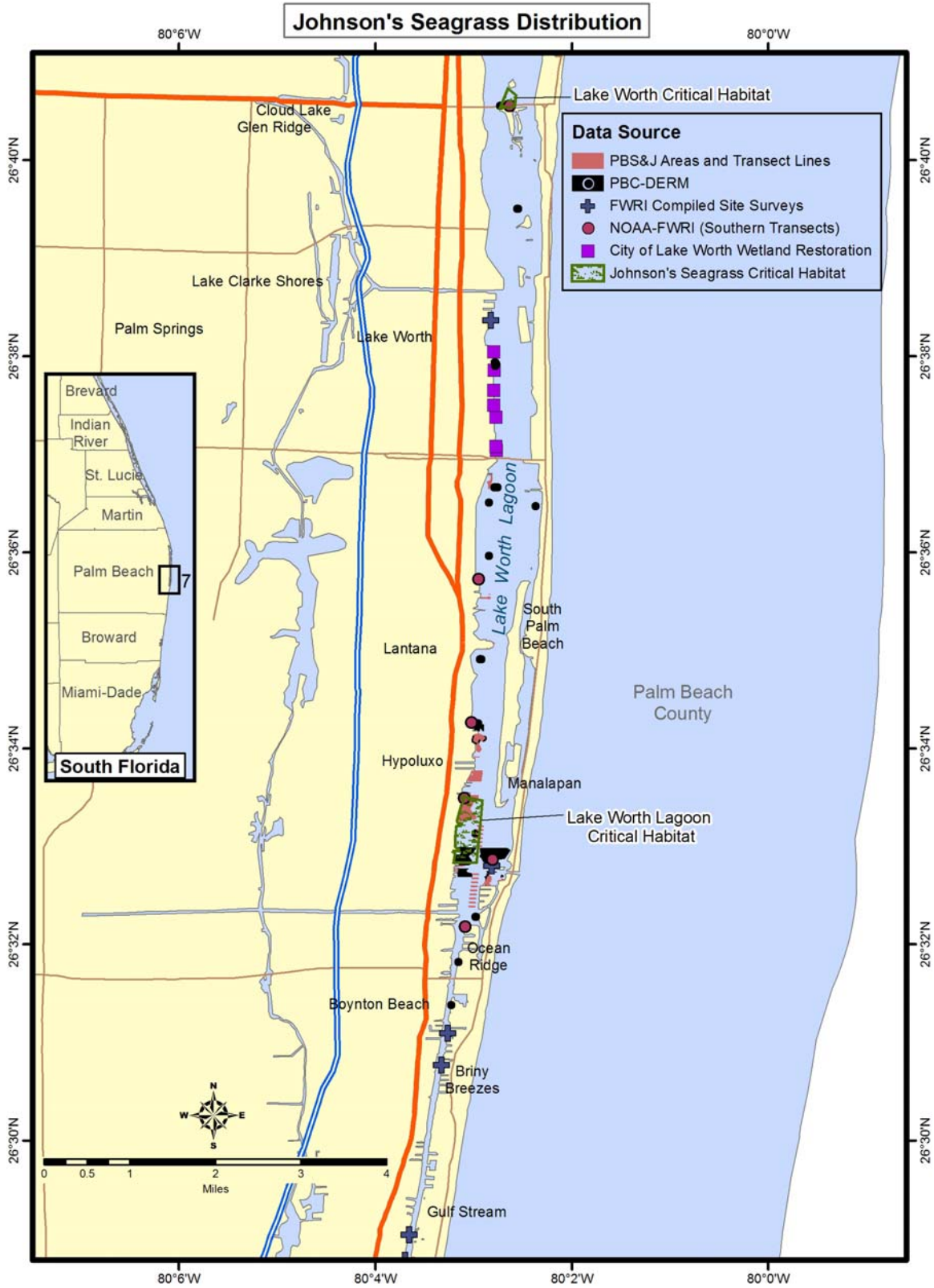


**Figure 13.** *Halophila johnsonii* distribution map 5b. Symbols indicate confirmed presence of *H. johnsonii*. See Table 2 for further information about surveys.



**Figure 14.** *Halophila johnsonii* distribution map 6. Symbols indicate confirmed presence of *H. johnsonii*. See Table 2 for further information about surveys.



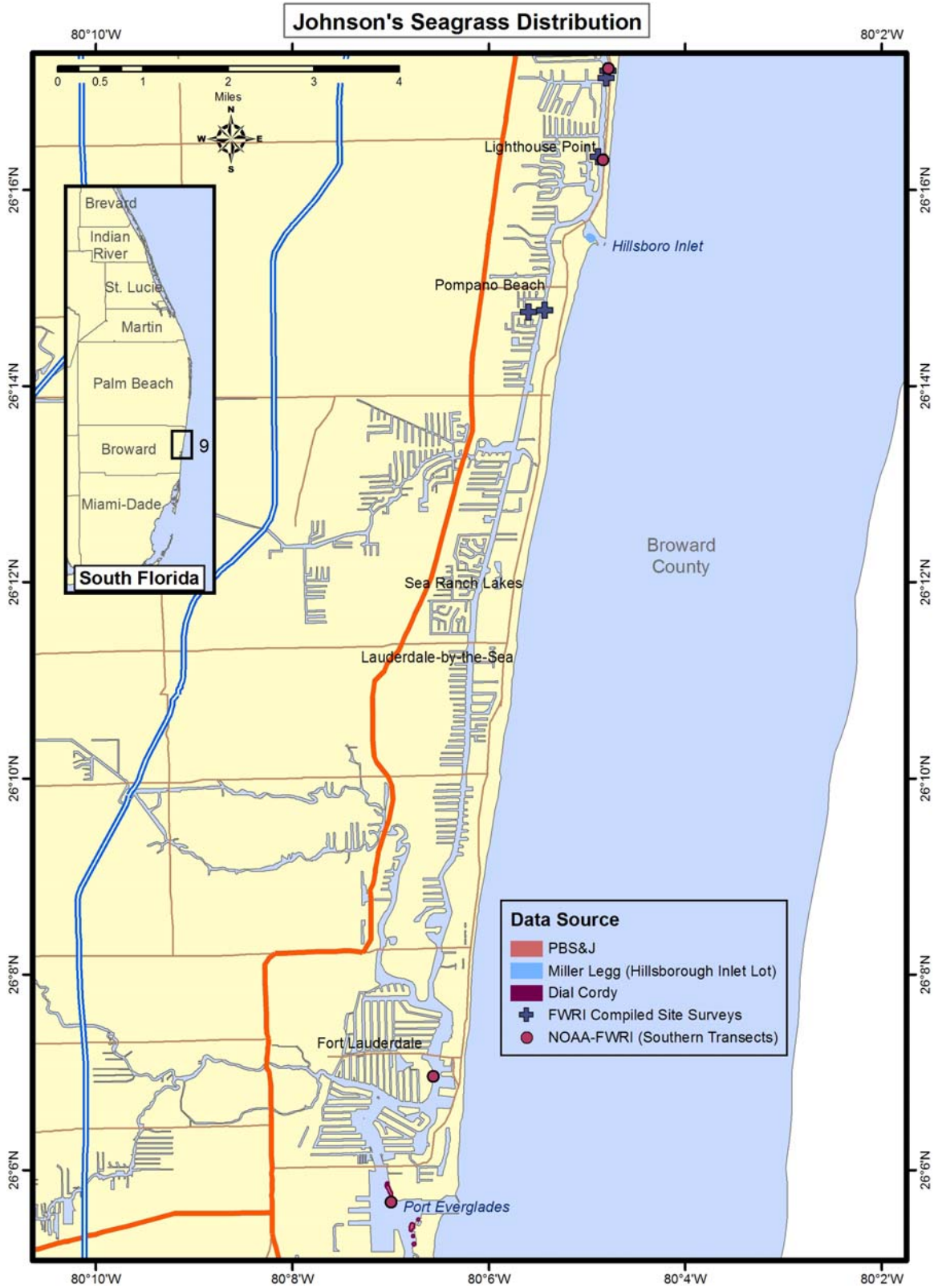


**Figure 15.** *Halophila johnsonii* distribution map 7. Symbols indicate confirmed presence of *H. johnsonii*. See Table 2 for further information about surveys.



**Figure 16.** *Halophila johnsonii* distribution map 8. Symbols indicate confirmed presence of *H. johnsonii*. See Table 2 for further information about surveys.

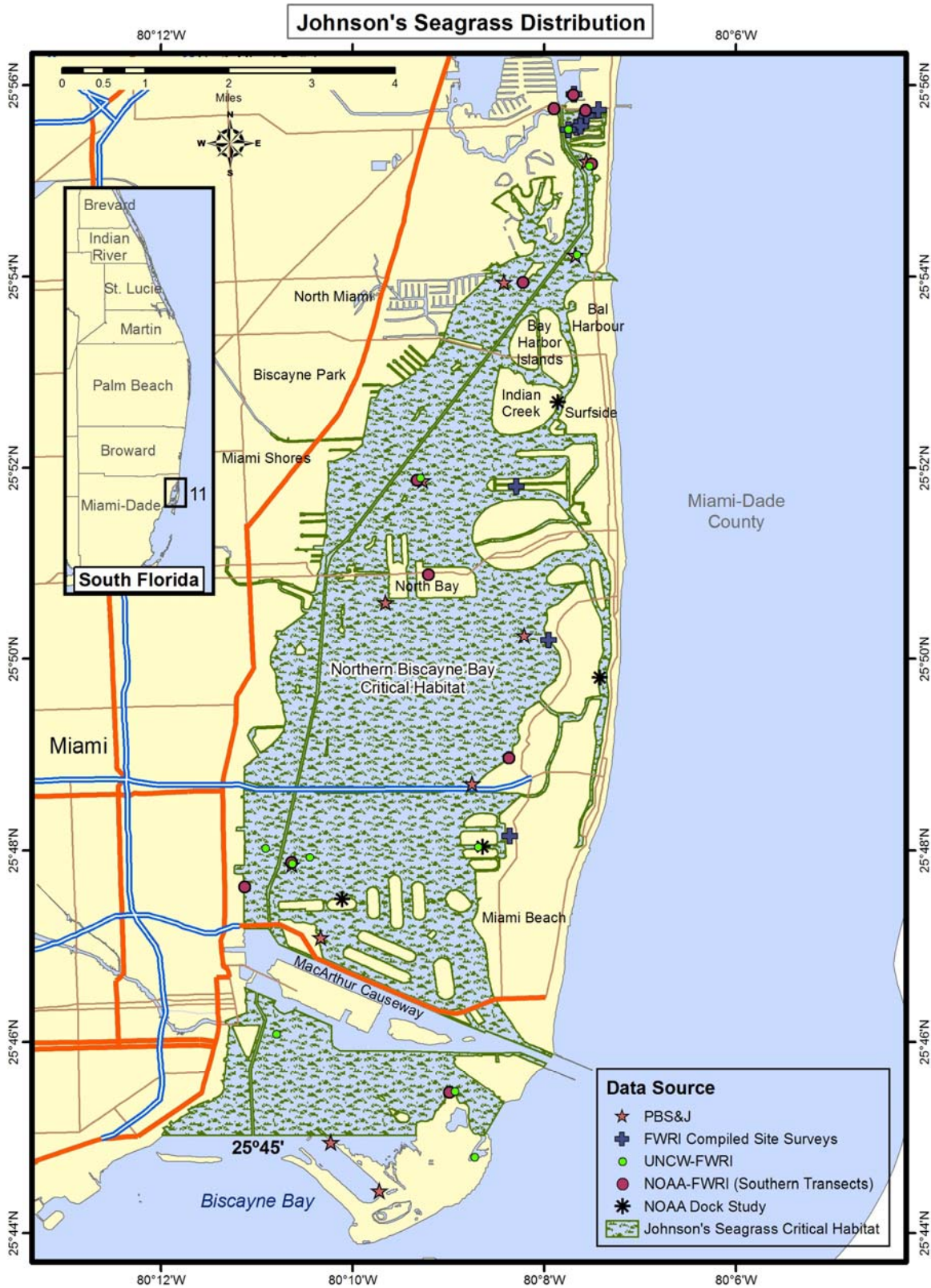




**Figure 17.** *Halophila johnsonii* distribution map 9. Symbols indicate confirmed presence of *H. johnsonii*. See Table 2 for further information about surveys.



**Figure 18.** *Halophila johnsonii* distribution map 10. Symbols indicate confirmed presence of *H. johnsonii*. See Table 2 for further information about surveys.



**Figure 19.** *Halophila johnsonii* distribution map 11. Symbols indicate confirmed presence of *H. johnsonii*. See Table 2 for further information about surveys.



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NATIONAL MARINE FISHERIES SERVICE  
5-YEAR REVIEW  
*Halophila johnsonii* Eisman

**Current Classification:** Threatened

**Recommendation resulting from the 5-Year Review**

- Downlist to Threatened  
 Uplist to Endangered  
 Delist  
 No change is needed

**Review Conducted By:**

W. Judson Kenworthy, National Oceanic and Atmospheric Administration, Center for Coastal Fisheries and Habitat Research, Beaufort, NC.

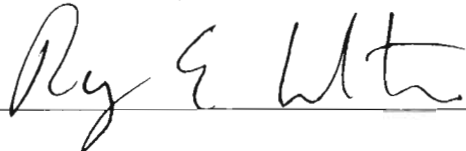
Shelley Norton, National Oceanic and Atmospheric Administration, National Marine Fisheries Service, Protected Resources Division, St. Petersburg, FL.

Stacey Harter, National Oceanic and Atmospheric Administration, National Marine Fisheries Service, Southeast Fisheries Science Center, Panama City Laboratory, FL

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**REGIONAL OFFICE APPROVAL:**

**Regional Administrator, NOAA Fisheries Service**

Approve:  Date: 11/5/07

**HEADQUARTERS APPROVAL:**

**Assistant Administrator for Fisheries**

Concur  Do Not Concur

Signature  Date 11/27/07