

Seagrass Ecosystem Restoration Research Compound Ribbon Cutting Ceremony

SEAGRASS RESTORATION TECHNOLOGY DEVELOPMENT INITIATIVE 403.93344(3)(e) F.S. **ACCOMPLISHMENTS AND PRIORITIES REPORT** JANUARY 2026

Seagrass is critically important to the ecology and economy of coastal ecosystems, serving vital functions for various fisheries, nursery habitats, and food for a variety of keystone species. Seagrasses provide shoreline/sediment stabilization, absorb nutrients, sequester carbon, and enhance coastal resilience. Unfortunately, seagrasses have significantly declined over the past decades and continue to be rapidly lost in Florida due to both indirect and direct anthropogenic impacts as well as the broader effects of global change.

A science-based restoration effort is needed to address diminishing seagrass habitat, distribution, and diversity. This applied research and restoration initiative builds upon the ongoing partnership between seagrass nursery restoration practitioners and top marine and genetic scientists to generate

a sustainable source of genetically resilient and naturally reproducing seagrass, in both land-based nurseries and in-water seagrass field nurseries, as well as large-scale seagrass gene banks.

This report is being provided to meet the requirement of 403.93344(3)(e) Florida Statutes which states: “Beginning January 15, 2024, and each January 15 thereafter until its expiration, the initiative shall submit a report that contains an overview of its accomplishments to date and priorities for subsequent years to the Governor, the President of the Senate, the Speaker of the House of Representatives, the Secretary of Environmental Protection, and the executive director of the Fish and Wildlife Conservation Commission.”

SEAGRASS RESTORATION TECHNOLOGY DEVELOPMENT INITIATIVE

The Florida Legislature and Governor, in recognizing the need for innovative seagrass research to guide optimizing restoration, passed the Seagrass Restoration Technology Development Initiative (Initiative, 403.9334 Florida Statutes). Under this statute, the Legislature established a collaborative and coordinated effort among public and private research entities to develop restoration technologies and approaches for addressing the loss of seagrass and cascading ecological and economic impacts of that loss to communities across Florida.

The goal of the Initiative is to develop, test, and implement innovative, effective, cost-efficient, and environmentally sustainable technologies and approaches for restoring coastal seagrass ecosystems. The Initiative has been established within the Department of Environmental Protection (Department) as a partnership between the Florida Aquatic Preserve Program, Mote Marine Laboratory (Mote), and the University of Florida (UF).

SEAGRASS INITIATIVE OBJECTIVES

- Develop and operate a seagrass research nursery with various statewide seagrass populations at Mote’s Aquaculture Research Park for this Initiative.
- Develop and grow partnerships to establish additional seagrass nurseries across the state to further this Initiative’s research goals.
- Test the resilience of different statewide seagrass populations to multiple environmental stressors in controlled lab and nursery settings to determine the most genetically resilient seagrass.
- Collaborate with coastal restoration practitioners to develop sustainable sources of genetically resilient seagrass.
- Develop and maintain a genetic library of

Florida seagrass to utilize for statewide restoration efforts.

- Facilitate engagement with other pertinent marine science and technology development organizations to pursue applied research and technology for the successful restoration of seagrass ecosystems.
- Create a 10-year Florida Seagrass Restoration Plan to implement tools and technologies developed under the Initiative.

APPROPRIATED FUNDING AND REPORTING

The Initiative legislation requires the Department to award \$2-million each year for 5 years to Mote to function as the lead administrative component to achieve the goals of the Initiative. The contract was executed in February of 2024. Mote may not use more than 5 percent of its awarded funds for direct annual Initiative administration and coordination costs.

Financial and performance accountability will be managed via the contract from the Department to Mote and subawards from Mote to UF and other research and technology development collaborators. These contractual agreements will include deliverables and financial documentation and will align with this required Annual Report and updates provided at public Technology Advisory Council meetings.

LEVERAGING WITH PRIVATE AND FEDERAL FUNDS

In accordance with 403.93344(3)(c)(3) F.S., the Initiative shall leverage state-appropriated funds with additional funds from private and federal sources. Leveraged funds and collaborations are well underway with generous support from the Bernard and Norton Wolf Family Foundation and Publix Super Markets, Inc., as well as grants from the National Oceanic and Atmospheric Administration and Indian River Lagoon National Estuary Program, and support with postdoctoral fellowship funding from

SUMMARY OF INITIATIVE PROGRESS

- Administrative Structure Implemented
- Mote Leveraged Funding Secured
 - NOAA
 - Philanthropic
 - Publix Seagrass Genetics Research Hub
 - Wolf Foundation Biogeochemical Carbon Sequestration Building
- Initiative Technical and Financial Reports submitted
- Released 1st competitive RFP
 - Funded 8 projects
 - Projects wrapping up
 - Receiving Final Reports
- Released 2nd competitive RFP
 - Funded 7 projects
 - Subcontracts sent and executed
 - Projects underway
 - Updates on these at next TAC Meeting
- Mote Annual Accomplishments and Priorities Reports Delivered
- Funded 6 additional projects utilizing funds from Year 3 of the Initiative
- 10 Year Strategic Seagrass Restoration Plan in development
 - 122 management documents reviewed
 - Florida Seagrass Restoration Sites website completed
 - Draft Plan Framework
- Seagrass Restoration Center Greenhouse structure completed
 - Projects in progress and completed
- Renovating on-site housing for visiting researcher lodging
- Seagrass Genetic Library submittal website created
- Extracted DNA from 816 samples and prepared 472 for sequencing

the National Science Foundation. Please refer to the Appendix: Project Summaries section for more details about leveraged project funding.

RESEARCH PARTNERSHIPS

Another important part of the Initiative was included under 403.93344(3)(c)(1), which states that “Mote Marine Laboratory may, with the concurrence of the department, use a portion of the awarded funds to facilitate additional engagement with other pertinent marine science and technology development organizations in this state and around the world to pursue applied research and technology for the successful restoration of seagrass ecosystems.” Thus, in close partnership with the Department and UF, and in addition to funding distributed to UF for targeted Initiative research, Mote releases annual Requests For Proposals to bring together the best and brightest scientists from Florida and around the world to assist this Initiative.

The Initiative’s competitive funding opportunities are open to any and all interested parties, and Mote has received widespread local, state, national, and international interest. Mote convenes highly qualified research proposal review panels as needed to guide project selection and funding. The use of seagrass research facilities at the Mote Aquaculture Research Park are available at no charge to funded Initiative collaborators. Subcontracts are structured on 12-month timeframes to expedite the goals of the Initiative, though researchers can apply for multiple projects and across subsequent years.

Request For Proposals were launched in December 2023 and 2024 with these funding priorities:

- examining the genetic diversity of seagrass populations considering watershed stressors across Florida;
- developing a genetic library of Florida seagrass;
- testing the resilience of different statewide seagrass populations to multiple environmental

- stressors in controlled lab and nursery settings;
- examine existing seagrass restoration technologies and/or conduct planning efforts and pursue regulatory approval for testing novel seagrass restoration technologies;
- organizing and encouraging collaboration among the scientists and restoration practitioners working on Florida seagrass genetics to achieve the goals of the Initiative (this will be partially accomplished through this RFP, the Initiative Technology Advisory Council, and a forthcoming Initiative Workshop).

A complete list of project titles, principal investigators, and summaries are attached as an Appendix to this Report. The number and diversity of private business, agency, and academic partners will continue to grow during the Initiative - below are the logos of a few key collaborators of the Initiative:



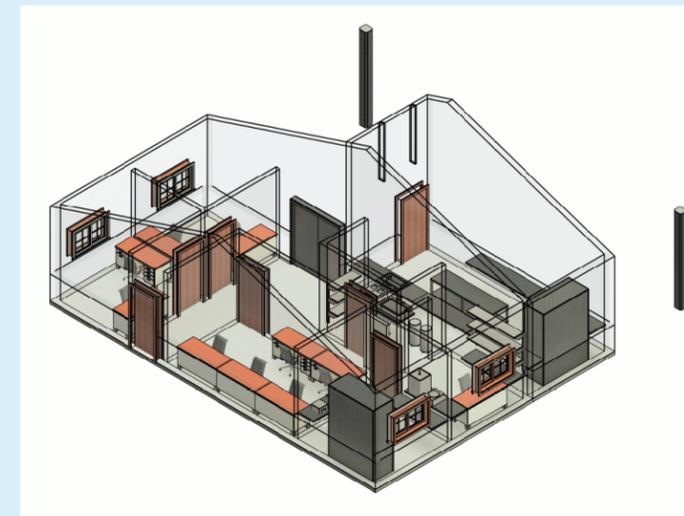
SEAGRASS TECHNOLOGY TESTING FACILITIES AND EXPERT FIELD SCIENTISTS

A key part of furthering seagrass research and restoration science is the need to safely test simulated coastal ecosystem stressors and restoration technologies. Thus, Mote created a cutting-edge seagrass greenhouse/laboratory at the Mote Aquaculture Research Park in Sarasota. This unique testing facility includes recirculating seawater systems with the ability to manipulate a full suite of environmental stressors (water clarity, temperature, pH, nutrients, etc.) to determine the most resilient seagrass to be utilized in restoration. This facility will also maintain resilient genotypes for future field propagation.

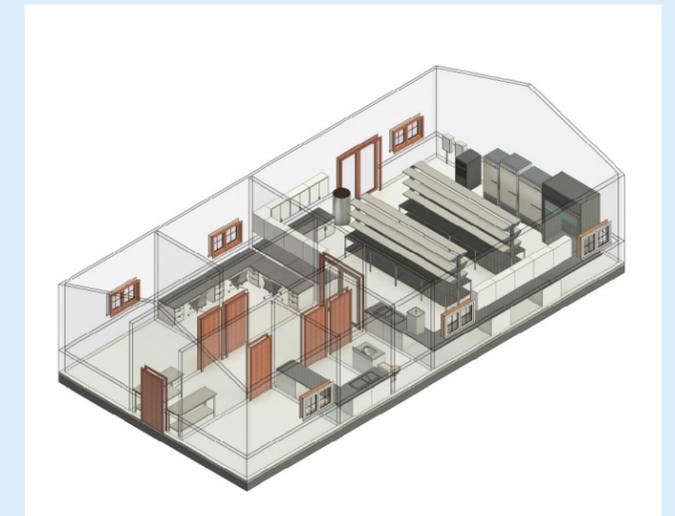
Since the start of the Initiative in 2023, Mote has completed the construction of the 60 by 60-foot Ron and Marla Wolf Seagrass Restoration Center for Ocean Sustainability Greenhouse Facility with 24 raceways that have electronic monitoring devices to measure and track dissolved oxygen (DO), temperature, and pH. This facility can be used free of charge by Initiative-funded collaborators for their seagrass research experiments. Due to increased interest and demand for the testing facility, Mote is planning to expand the Greenhouse by 30 feet to fit an additional 12 raceways. To complete Mote's Seagrass Ecosystem Restoration Research Compound (SERRC) at the Mote Aquaculture Research Park, we have also made significant progress on the construction of the Biogeochemical Carbon Sequestration Lab and Publix Seagrass Genetics Research Hub. These buildings, funded by the Bernard and Norton Wolf Family Foundation and Publix Super Markets, Inc., respectively, will work in tandem with the goals of the Initiative. Lastly, design and permitting activities are underway for the construction of a seagrass research, education, and restoration nursery, in close partnership with the Florida Department of Environmental Protection, at Bahia Honda State Park.



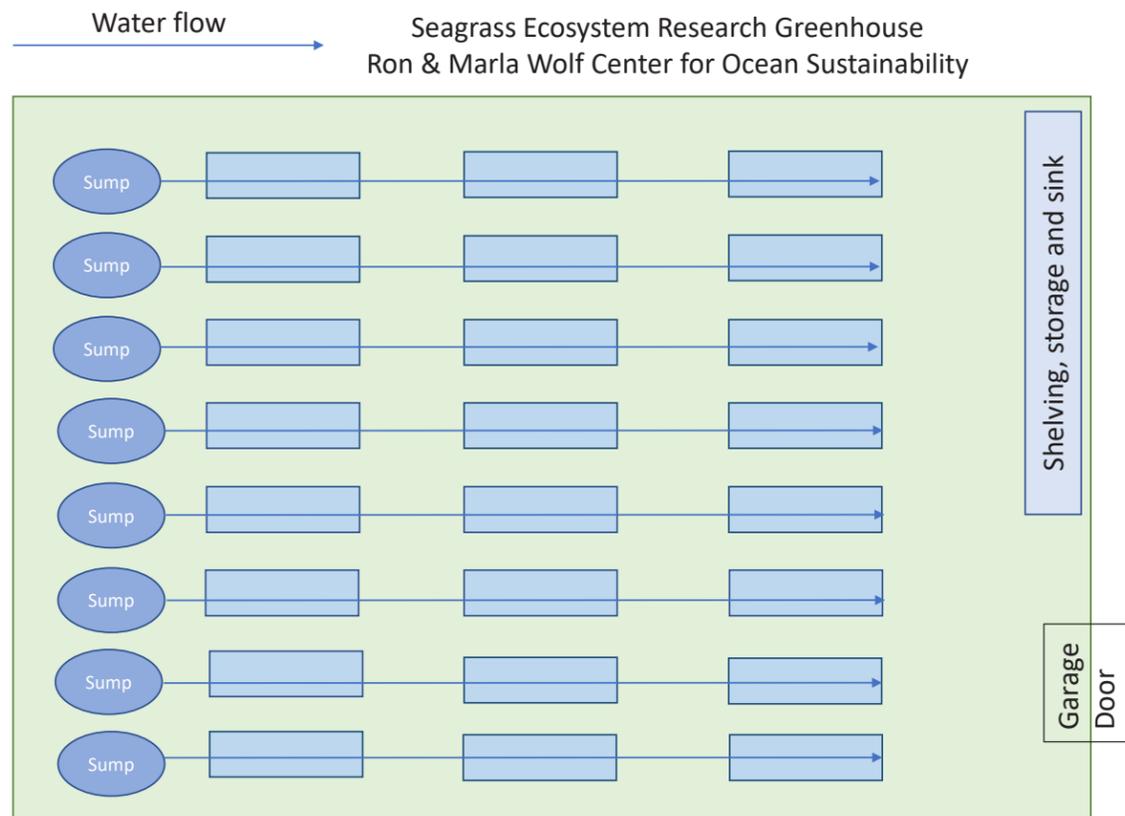
The Ron and Marla Wolf Seagrass Restoration Center for Ocean Sustainability Greenhouse Facility



Rendering of the Biogeochemical Carbon Sequestration Lab



Rendering of the Publix Seagrass Genetics Research Hub



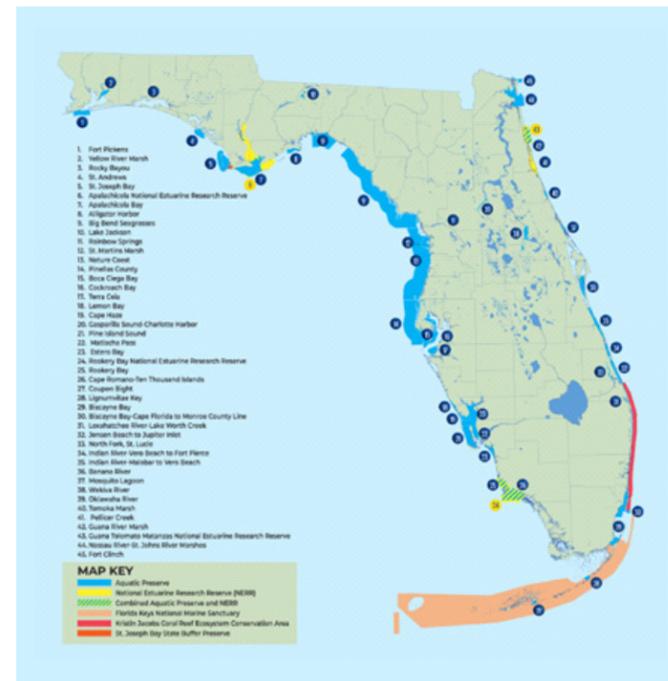
Mote staff working with *Thalassia testudinum*



Halodule wrightii growing in the Ron and Marla Wolf Seagrass Restoration Center for Ocean Sustainability Greenhouse Facility

Additionally, through this strategic Initiative partnership with the UF Department of Soil, Water, and Ecosystem Sciences, the Initiative also includes top seagrass genetics scientists and laboratory resources on the main UF campus in Gainesville. Finally, a critical part of the Initiative is having statewide place-based knowledge of the seagrass populations, field stressors, ecological trends,

research/restoration underway, regulations, potential local collaborators, and stakeholders. Thus, an important Initiative partner is the Department's Aquatic Preserve Program. This team of scientists manages 43 preserves under Florida's Aquatic Preserve Act (258.35-258.46 F.S.) covering over 2.6 million acres of Florida's best aquatic resources to ensure the continuation of natural conditions so



Location map of Florida's Aquatic Preserves

"their aesthetic, biological and scientific values may endure for the enjoyment of future generations."

TECHNOLOGY ADVISORY COUNCIL

As stated in 403.93344(4) F.S.: "The Initiative Technology Advisory Council (TAC), an advisory council as defined in s. 20.03, is established as part of the initiative. The advisory council's membership must include marine science, technology development, and natural resource management representatives from this state's aquatic preserves, private organizations, and public or private research institutions. The council shall meet at least twice annually."

These public meetings provide overviews and updates of the administrative structure developed to run the Initiative, public record laws, projects underway, proposals being considered, and planned next steps. Short biographies of the Council members, presentations, and meeting minutes can be found on the Mote Seagrass Research and Restoration website.



Mote staff and TAC Meeting attendees touring Mote's Key Largo Coral Nursery

The first TAC Meeting of 2025 was held at Apalachicola National Estuarine Research Reserve in February. The second TAC Meeting of 2025 was held at Reefhouse Resort & Marina in October, which is home to Mote's Key Largo Coral Nursery. Present at both of the meetings were TAC Members, scientists from FDEP and FWC, and PIs/Co-PIs of Initiative-funded research projects.

SEAGRASS GENETIC LIBRARY

Significant progress has been made in the Seagrass Genetic Library. Mote Marine Laboratory has received over 300 samples from four partners. Additionally, Mote scientists from the Seagrass Ecosystem Restoration Research (SERR) program have collected over 1,000 samples of shoal grass, *Halodule wrightii*, from three watersheds along Florida's Gulf Coast. These samples have been added to the Seagrass Genetic Library.

A website for partners to upload the associated metadata of their samples is now live on Mote's website [<https://mote.org/seagrass-genetic-sampling-intake>]. This data streams directly into a database that tracks the progress of the samples from DNA extraction to library preparation, and from sequencing to data analysis utilizing BreedBase.



Mote's Dr. Gallery cryopreserving seagrass samples



Publix Seagrass Genetics Research Hub



Mote staff and interns collecting seagrass samples with staff from Estero Bay Aquatic Preserve



Thalassia testudinum in Charlotte Harbor

BreedBase, developed at the Boyce Thompson Institute, tracks individual seagrass genotypes and the data collected regarding each individual's resiliency to various stressors.

The SERR program at Mote has extracted DNA from 816 samples and prepared 472 for sequencing. Mote will be shipping these samples to the Interdisciplinary Center for Biotechnology Research (ICBR) at the University of Florida for sequencing. Once samples are sequenced, their BreedBase Accession will be updated with their genetic

information and combined with their resiliency to stressors if applicable. The metadata also captures water quality measurements at the collection site when provided by partners.

FLORIDA SEAGRASS RESTORATION PLAN

To ensure socialization and implementation of Initiative research findings and encourage collaborations among Florida coastal seagrass ecosystem scientists, the Initiative includes section 403.93344(3)(d) that states: "In collaboration with the

Aquatic Preserve Program, Mote Marine Laboratory and the University of Florida shall create a 10-year Florida Seagrass Restoration Plan to implement tools and technologies developed under the initiative."

Beginning in June 2024, Moffatt & Nichol (M&N) began work with Mote Marine Laboratory to develop a Florida Resilient Seagrass Restoration Plan (Plan) in collaboration with the Florida Aquatic Preserve Program and University of Florida. The Plan will support implementation of tools and technologies developed under the Seagrass Restoration Technology Development Initiative and align with the Initiative objectives.

Plan development began with identifying relevant genetic studies, understanding current watershed conditions and existing seagrass management and restoration strategies, and applying information collected by watershed around seagrass stressors and associated ranges to inform resilient genetic research and technology development.

Approximately sixty genetic studies and one hundred and twenty-two management and restoration plan documents have been reviewed. Identified seagrass stressors, seagrass restoration and/or recovery targets and strategies for genetically resilient seagrass were compiled for each plan. Management plan review included Aquatic Preserve, Surface Water Improvement, National Estuary Program, and federal park, preserve, monument and seashore management plans. Additionally, the Florida Seagrass Integrated Mapping and Monitoring Program Report (Florida Fish and Wildlife Conservation Commission, 2016) and region-specific peer reviewed publications have been reviewed. While all reviewed management plans include objectives and activities to support healthy seagrass communities, only seven plans included restoration targets and three included restoration projects – all of which were under two acres. A total of two plans included strategies

around genetic management, demonstrating the gap in Florida's approach to recovery. Table 1 below summarizes which management plans included seagrass restoration. Following this review, seagrass restoration and mitigation projects were collected from various sources to create a comprehensive database. Sources include the National Estuary Programs Projects Dashboard, the Florida Fish and Wildlife Conservation Commission's database, Florida Department of Environmental Protection, as well as through conversations with restoration practitioners. This database was created to compile seagrass planting projects and summarize project components. One hundred and twenty-five projects and seagrass nurseries are currently featured. To the extent practicable, the coordinates of project locations were gathered and integrated into an interactive GIS Experience map (Figure 1). This map includes project details gathered as part of this project such as project stage, size, species planted, material source and site manipulation. The map allows new sites to be submitted into the database by practitioners in real time. Screen captures from the map experience can be seen in Figures 1 and 2 below. The online Plan is under development and focuses on increasing the pace and success of seagrass restoration and recovery through genetic resilience. A three-tiered site selection process has been integrated for site selection (Figure 3).

The Initiative is building a GIS based habitat suitability assessment model, which will ultimately be applicable to all regions of the state. Using publicly available high-resolution environmental data to establish a qualitative assessment of potentially suitable project locations, this habitat suitability model (HSM) is piloting the tier-one phase of resilient seagrass site selection. The pilot, focused in Florida Bay, will assess the efficacy of incorporating the model in identifying locations where data indicate suitable conditions for seagrass

| Management Document | Seagrass Restoration Targets | Seagrass Restoration Projects (<2 ac.) | Seagrass Restoration Planning Activities | Genetically Restoration Seagrass Mgmt. Activities | Genetically Resilient Seagrass Needs |
|--|------------------------------|--|--|---|--------------------------------------|
| Aquatic Preserve Management Plans ¹ | 1 | 2 | 9 | - | - |
| Estuary Program and Comprehensive Conservation and Management Plans ² | 2 | - | 5 | - | 1 |
| Surface Water Improvement and Management (SWIM) Watershed Plans ³ | 4 | - | 1 | - | - |
| Basin Management Action Plans ⁴ | - | - | 3 | - | - |
| National Park, Seashore, and Marine Sanctuary Management Plans | - | - | 2 | - | 1 |
| County Comprehensive Plans | - | - | 1 | - | - |
| State Park Unit Management Plans ⁵ | - | 1 | 5 | - | - |
| Comprehensive Everglades Restoration Plan | - | - | - | - | - |
| Seagrass Integrated Mapping and Monitoring Program Report ⁶ | - | - | - | - | - |
| TOTAL | 7 | 3 | 26 | 0 | 2 |

Table 1. Overview of management plans including seagrass restoration and planning activities in Florida.

1 Managed by the Florida Department of Environmental Protection

2 Managed by the National Estuary Programs under the Environment Protection Agency

3 Managed by the state water management districts

4 Managed by the Florida Department of Environmental Protection

5 Managed by the Florida Department of Environmental Protection

6 Managed by the Florida Fish and Wildlife Conservation Commission

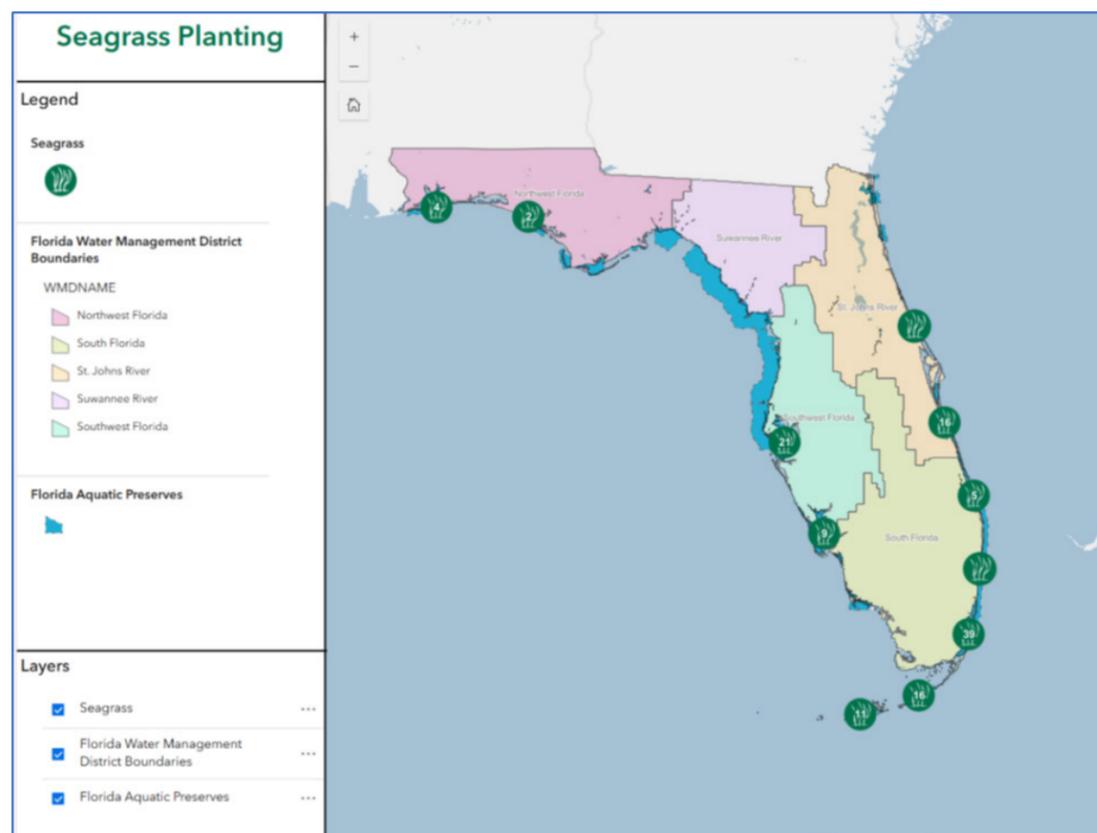


Figure 1. Screen capture of the interactive GIS Experience map featuring planted seagrass restoration projects.

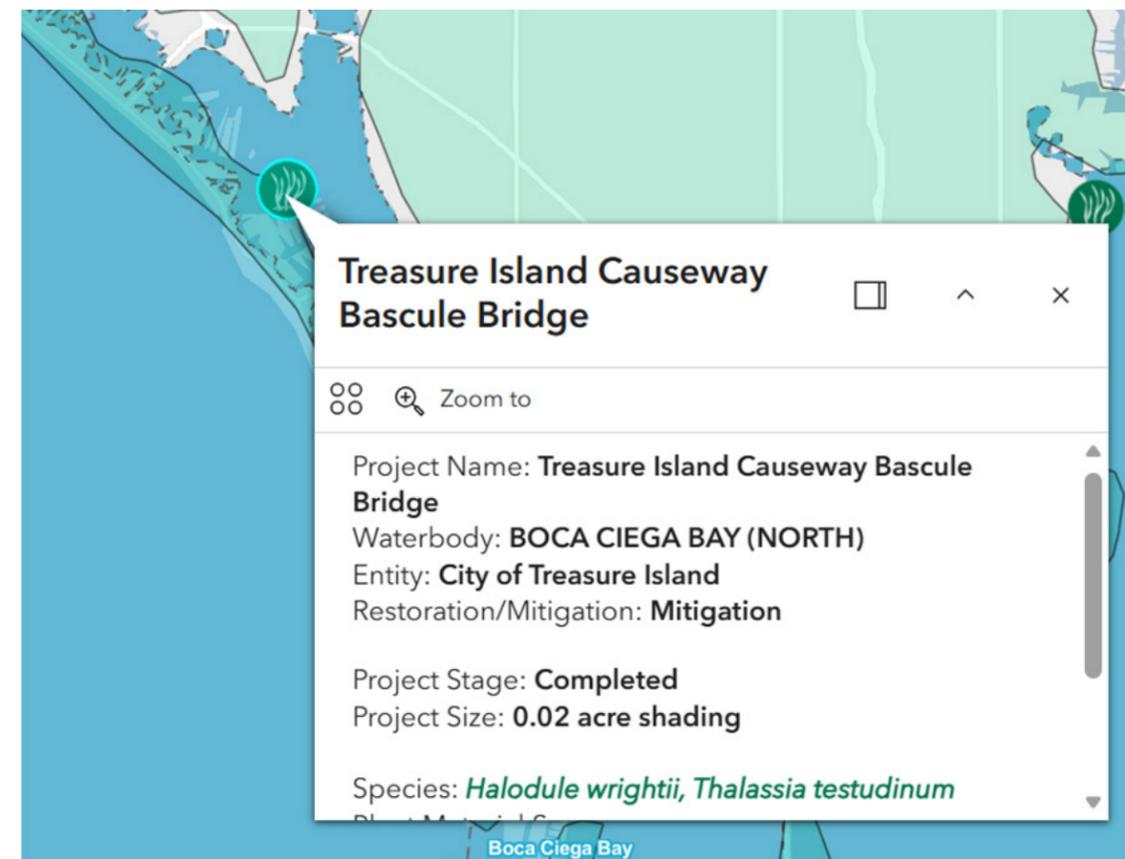


Figure 2. Screen capture of seagrass site characteristics included in the interactive GIS Experience map.

and expand the traditional considerations for the genetically resilient ranges identified by this initiative. This assessment will be the foundation of the proposed three-tiered process for selecting project locations for resilient seagrass restoration.

Tier two will then align initiative outcomes on genetic suitability to the proposed site location and provide recommendations on genotypes and/or genetic markers for known location stressors. The final tier will then rely on place-based manager knowledge for final site selection and approval.

The Plan framework provides the analytics needed for efficient review, approval and implementation of genetically resilient seagrass restoration in Florida.

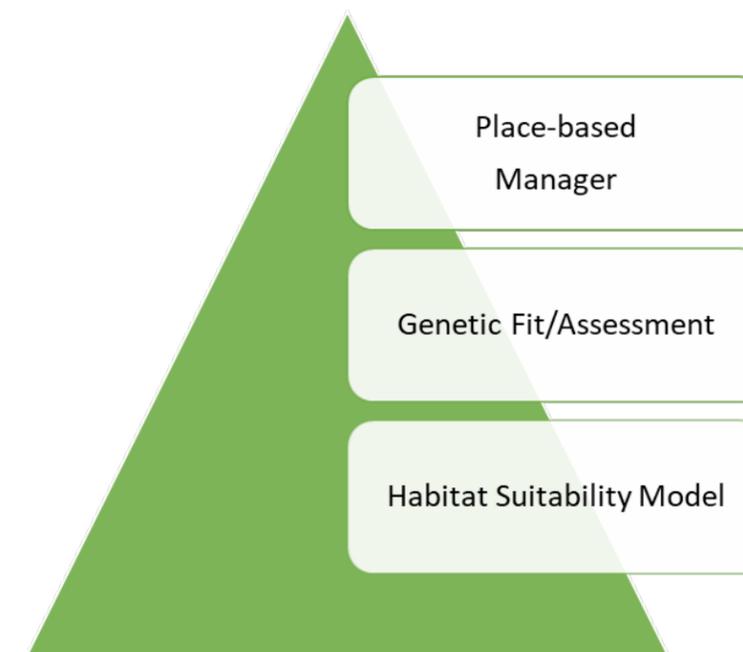


Figure 3: Three-tier approach for selecting resilient seagrass restoration sites.

APPENDIX

SEAGRASS RESTORATION TECHNOLOGY DEVELOPMENT INITIATIVE PROJECT EXECUTIVE SUMMARIES

Title: Ron and Marla Wolf Seagrass Restoration Center for Ocean Sustainability

Principal Investigator: Dr. Michael P. Crosby (Mote)

Project Date: December 2022 – Present

Summary: Science-based large-scale strategic mitigation and restoration initiatives are needed to address diminishing seagrass habitat, distribution and diversity. The creation of the Ron and Marla Wolf Seagrass Restoration Center for Ocean Sustainability targets many immediate seagrass ecosystem research needs in Florida, while also developing scientific technologies and administrative processes for increasing long-term blue carbon sequestration that will have transferability to other regions. This Center will assist the growth of land-based seagrass nurseries and seagrass gene bank as part of continued expansion of the Mote Aquaculture Research Park, and satellite seagrass operations associated with Mote coral nurseries and restoration activities along the Florida's Coral Reef. This project is supported by a gracious donation from the Bernard & Norton Wolf Family Foundation.

Title: Using genetic tools to boost seagrass restoration success in the Indian River Lagoon

Principal Investigator: Dr. Jamila Roth (NSF Postdoctoral Fellow)

Project Date: October 2023 - March 2024

Summary: As seagrasses decline in Indian River Lagoon, scientists and partners are analyzing the genetic diversity of seagrass donor material from nurseries and the genetic diversity of natural seagrass meadows with the hope of increasing the resiliency of restored seagrasses to environmental stressors. This project is a partnership among Mote, UF, Florida Oceanographic Society and Florida Atlantic University's Harbor Branch Oceanographic Institute. This project is supported by a competitive grant from the Indian River Lagoon National Estuary Program.

Title: Using Land-Based Seagrass Nurseries and Genetic Approaches to Increase Seagrass Restoration Success

Principal Investigator: Dr. Michael P. Crosby (Mote)

Project Date: September 2023 – August 2027

Summary: This National Oceanic and Atmospheric Administration funded project is working in sync with the Initiative. Funds from this grant will support Mote's Seagrass Research and Restoration Program which strives to (a) develop cutting-edge propagation and out-planting technologies, (b) establish the research structure for ensuring that there are multiple genetically distinct, locally adapted strains that are resistant to common stressors available for restoration projects, and (c) serve as a hub for innovative applied restoration research. This funding will support infrastructure and staffing needs across multiple research locations.

Title: Impact of Environment-Seagrass-Microbe Interactions on Seagrass Stress Response and Ecosystem Functions

Principal Investigators: Dr. Jamila Roth (NSF Postdoctoral Fellow), Drs. Julie Meyer (University of Florida), Laura Reynolds (University of Florida), Ashley Smyth (University of Florida), and Loraé Simpson (St. Johns River Water Management District)

Project Date: March 2024 – February 2027

Summary: Many drivers of seagrass loss, including heat waves, nutrient over-enrichment, increased turbidity, hypersalinity, hypoxia, and invasion by macroalgae, co-occur with sulfide toxicity, often producing synergistic effects. Sulfide is a phytotoxin that negatively impacts plant growth, aerobic metabolism, and nutrient uptake, often leading to seagrass mortality and die-back events. Since seagrasses grow in sediments with high sulfide concentrations, their survival requires adaptations that support sulfide detoxification. Results from this project will provide information on the abundance of beneficial sulfide-oxidizing bacteria in seagrass meadows throughout Florida, exploring how nutrient availability and stress levels may impact the abundance of the sulfide-oxidizing bacteria. Additionally, information on how sulfide-oxidizing bacteria influence seagrass resilience and response to stress can be applied to seagrass management and restoration efforts.

INITIATIVE-FUNDED PROJECTS

Title: "University of Florida Seagrass Initiative Research"

Principal Investigators: Dr. Laura K. Reynolds (University of Florida)

Project Date: May 2024 – October 2026

Summary: Dr. Reynolds is creating maps of seagrass diversity around the state (~1000 samples from 10 locations), and beginning restoration work (sampling donor material from restoration sites, microsatellite analysis to balance ideas of planting a diverse source vs planting resilient material). So far, her and her collaborators have collected over 800 seagrass samples and have extracted DNA from almost 600, with ongoing analysis. She has also started piloting microbial analyses, which will later be applied to this project. Dr. Reynolds is also working with Dr. Dominique Gallery to understand genome size for *Halodule wrightii*, in addition to *Syringodium filiforme*, and beginning efforts to annotate those genomes.

Title: "Assessment of Population Genomic Variability Associated to Stress Resistance in Florida Seagrasses"

Principal Investigators: Dr. Iris Segura-Garcia (Florida Atlantic University Harbor Branch Oceanographic Institute)

Co-Principal Investigators: Dr. Krista McCoy and Dr. Michael McCoy (Florida Atlantic University Harbor Branch Oceanographic Institute)

Project Date: July 2024 – June 2026

Summary: The aim of this proposal is to identify genetic variability associated with the ongoing stressors using a genome-wide approach that will capture the functional genetic variability of a species. To achieve this goal, fieldwork was conducted throughout 2024 at multiple sites along the Indian River Lagoon (IRL), Lake Worth Lagoon, Biscayne Bay and Upper Florida Keys. In addition, scientists sampled and labelled 18 plants growing at the Florida Oceanographic Society Seagrass Nursery and six at the Harbor Branch Seagrass Nursery by applying the same genome-wide genotyping approach used for wild populations to nursery-grown seagrasses. Using Restriction-site Associated DNA sequencing (RADseq), 3,220 single nucleotide polymorphisms (SNPs) were identified. The results revealed significant genetic differentiation between populations from the Florida Keys and Biscayne Bay, as well as between the Lake Worth Lagoon (LW) and IRL seagrass populations. Dr. Segura-Garcia was selected to be funded again and proposed to expand the geographic scope of her current research to comprehensively characterize *S. filiforme* across the state of Florida. This research will allow them to quantify

levels of clonal and genetic diversity, patterns of connectivity among populations, and to identify genetically distinct populations whose genetic architecture may need to be protected to avoid disrupting local adaptations.

Title: “Testing variation in stress tolerance and restoration potential of Florida seagrass subpopulations”

Principal Investigators: Dr. Althea Moore (Texas A & M University – Corpus Christi), Dr. Jimmy Nelson (University of Georgia Skidaway Institute), Dr. Cliff Ross (University of North Florida), and Dr. Justin Campbell (Florida International University)

Project Date: July 2024 – September 2026

Summary: This project is testing stress tolerance of different statewide seagrass subpopulations to multiple environmental stressors in a controlled setting, looking at proteins to see which serve as a biomarker of stress tolerance. Scientists set up treatments in experimental aquaria and tested the effects of two stressors in combination (high temperature and high salinity) on seedlings from multiple regions of Florida in monoculture and in diversity combinations, then determined how many leaves the seedlings retained during the experiment, measured seedling growth, measured photosynthetic activity (YII) as a metric of stress response using PAM fluorometry, and analyzed the plant responses using mixed effects mode. Overall, the seedlings tested were surprisingly tolerant to the two-week exposure to a stressor combination of high salinity and high temperature, designed to partially simulate field conditions associated with die-offs in south Florida. As this project moves forward, data on the genetic diversity and variation across field sites and regions can help guide seedling group selection. Results suggest preliminarily that combining seedlings from disparate regions may be less successful for stress tolerance than within-region or within-site seedling groups. Dr. Moore was selected for an additional year of funding to analyze previously collected genetic data to determine subpopulation structure and test whether more genetically diverse groups perform better, test stress tolerance and resilience in terms of recovery over a period of months, and characterize the cellular mechanisms underpinning the stress response of *T. testudinum* using quantitative proteomics.

Title: “Addressing uncertainties to facilitate restoration success of *Halodule wrightii* beds: Does seagrass genetic variation and genotypic identity enhance primary productivity and confer resilience to stressors?”

Principal Investigators: Dr. T. Erin Cox (University of New Orleans)

Co-Principal Investigators: Nicola Anthony (University of New Orleans)

Project Date: June 2024 - December 2025

Summary: This project examined the role of SGV (seagrass bed genetic variation) and genotypic identity in enhancing primary productivity and conferring resilience to low light and high heat stressors. Seagrass was collected from Apalachicola, Florida and planted into containers to produce replicate beds of increasing genotypic levels. Replicates were assigned to one of twelve raceways. Then half of these replicated beds were exposed to a heat stress for two weeks followed by seven weeks at a lower temperature for recovery. After seven weeks heated, replicates had higher shoot density and biomass than controls, confirming the robust physiology of this opportunistic species. Microsatellite genotyping is currently being done to verify the genotypic identity and variation planted and relate to resilience. Results will be used to inform on genetic variation needed to improve restoration success.

Title: “Genetic Diversity of Targeted Seagrass Assemblages in Florida”

Principal Investigators: Thomas F. Ries (Ecosphere Restoration Institute)

Co-Principal Investigators: Dr. Susan Bell and Dr. Christina Richards (University of South Florida)

Project Date: June 2024 – September 2026

Summary: This project is focused on the field sampling of a suite of *Halodule wrightii* seagrass populations representing naturally persistent assemblages to collect baseline information on the genetic diversity of *H. wrightii*. Scientists took advantage of previously conducted seagrass restoration projects in Florida to examine genetic diversity of *H. wrightii* in restoration sites of varying ages, utilizing information and approaches from their earlier surveys of restoration. After collecting samples from the field sites, they completed DNA extractions and ddRadSeq library prep and completed the sequencing provided by Novogene. From the 10 collection sites, they have processed and analyzed 50 sediment samples. Furthermore, they have conducted field sampling at 16 additional sites, where they collected 323 *H. wrightii* samples and transferred them to Mote Marine Laboratory for their long-term storage in a genetic library. The analyses show that overall, the seagrass populations appear to be highly clonal, with limited but significant differences between sites. These differences seem to be geographically structured within the Tampa Bay area. Across comparisons of the four restoration sites, they found restored sites are comparable to natural sites in terms of clonal and genetic composition, suggesting that restoration has not altered genetic structure. Thomas was selected for an additional year of funding to expand upon their ongoing research and will use RNA sequencing to identify candidate stress tolerance genes in seagrass collected across their range of sites within Tampa Bay.

Title: “Investigating Potential Effects of *Caulerpa prolifera* on Shoal Grass Restoration in Florida”

Principal Investigators: Dr. Jenny Hansen (Brevard Zoo)

Co-Principal Investigators: Dr. Linda Walters (University of Central Florida), Dr. Austin Fox (Florida Institute of Technology), and Dr. Margaret Vogel (University of Lausanne)

Project Date: June 2024 - May 2026

Summary: *Caulerpa prolifera* is a macroalgae that can increase sediment accumulation and siltation which leads to the accumulation of phytotoxins in the water column. This project looked at the interactions between seagrass and *C. prolifera* in the sediments and water column. In the laboratory study, both *H. wrightii* and *C. prolifera* were found to alter sediment porewater chemistry and nutrient availability, with effects that persisted after transplantation between sediment types. Findings indicated a negative effect of *C. prolifera* on *H. wrightii*, even in the absence of direct competition for light or nutrients. In the mesocosm study, results indicate an apparent threshold *C. prolifera* biomass between 5-25% cover, above which *H. wrightii* growth becomes negatively impacted. Finally, the field study illustrated highly variable interactions between *C. prolifera* and *H. wrightii* based on site-specific constraints. Consistent to all three components of this study, competition between *H. wrightii* and *C. prolifera* was demonstrated in the laboratory, mesocosms, and in situ. Effects of competition were influenced by the abundance of *C. prolifera* and by external pressures such as depth and the presence of grazers (i.e., *Elysia subornata*). Evidence of sediment modification by both species was apparent in the laboratory study. *C. prolifera* altered sediments lowering ORP, leading to increased sulfide and greater phosphorous availability. Even with increased nutrient concentration in treatments where seagrass was planted into previous *C. prolifera* sediments than those that were previously bare or seagrass-inhabited, new growth rates of seagrass declined, indicating sediments were likely more toxic to *H. wrightii*. Modification of the sediment may support growth of *C. prolifera*, and toxicity can negatively impact, but

is not likely to eliminate, *H. wrightii*, as shoal grass persisted in each component of this study, though often with reduced growth. Overall, *C. prolifera* has a negative effect on *H. wrightii* growth rates that scales with its abundance and can persist even in the absence of direct competition. Dr. Hansen was selected for an additional year of funding to continue her research and further investigate how sediment characteristics, porewater chemistry and nutrients, the rhizosphere microbiome, and competition stressors influence shoal grass establishment through experimental field plantings and controlled laboratory studies.

Title: “Developing Technology for Kilometer Scale Seagrass Restoration in Florida”

Principal Investigators: Akhil Voorakkara (Ulysses Ecosystem Engineering)

Co-Principal Investigators: Dr. James Fourqurean (Florida International University)

Project Date: May 2024 – June 2026

Summary: Ulysses research objective is to determine ways in which to enable effective, low-cost seagrass restoration at the kilometer scale (rather than the hectare scale) through the use of a scalable subsea robotic planting system. So far, they have successfully developed and validated a mechanical seed injection system specifically designed for *Syringodium filiforme*. The device was developed and tested in aquaria using 3D printed seed analogues created based on real measurements and images supplied by our collaborators at the FIU Seagrass Ecosystems Research Laboratory. They have also worked with FIU to perform some initial field testing in July 2024 in Miami, validating the mechanics of the seed injector with real seed and real sediment. It was validated that 3D-printed seed analogues accurately replicate real seed behavior. Following successful testing in Miami, Ulysses has designed and tested a robotic seed injection system in laboratory conditions using 3D-printed seed analogues. The team is now working on a second iteration of the robotic seed injection system based on data gathered during the testing of the first iteration. During testing, the seed loading mechanism jammed several times, so a system redesign is underway. The full-scale ROV platform has the potential to plant 5 hectares a day, given seed supply and the right amount of injectors. Akhil was selected for an additional year of funding to further develop and test their robotic planting system.

Title: “Assisting seagrass recovery in Southwest Florida: Evaluation of hard clam (*Mercenaria campechiensis*) facilitation and identification of resilient *Halodule wrightii* phenotypes”

Principal Investigators: Dr. Stephen Hesterberg (Gulf Shellfish Institute)

Co-Principal Investigators: Dr. Nicole Rhody (Mote)

Project Date: July 2024 – October 2026

Summary: This project utilized raceways at Mote’s Aquaculture Research Park to evaluate two low-cost approaches to speed recovery of seagrass habitats: (1) co-planting of native southern hard clams and (2) identifying resilient shoal grass (*Halodule wrightii*) phenotypes that can be bred in nurseries on-land, testing the resilience of different seagrass sources to low light conditions and evaluate how hard clams further mediate seagrass responses. Progress so far includes identifying four regions for seagrass collections within Collier, Hillsborough, Lee, and Manatee counties and refining experimental design for the mesocosm experiment to include greater replication along the light intensity gradient. Dr. Hesterburg was selected for an additional year of funding to further evaluate the resiliency of shoal grass to low light conditions while observing transcriptome-wide gene expression of *H. wrightii* individuals pre- and post-stress exposure. Resilient populations and/or genotypes identified through this study will be propagated for future use in state-wide restoration efforts.

Title: “A new approach to seagrass restoration in Florida: exploring the potential for seed-based restoration”

Principal Investigators: Dr. James Fourqurean (Florida International University)

Co-Principal Investigators: Dr. Johannes Krause (Florida International University)

Project Date: June 2024 – June 2026

Summary: This project is: assessing the suitability of fast-growing, early successional seagrass *Syringodium* for seed-based restoration; investigating sexual reproduction, seed set, and seed banks in the field; and experimentally determining cues for promoting seed germination in mesocosms. Progress so far includes the collection of field observations of *Syringodium* sexual reproduction and morphology phenology data during the winter survey period, and observations of flowering and seed production phenology across the south Florida seagrass landscape. Scientists have observed that flowering densities have shown a notable decline from May to August with evidence of any reproductive shoots being absent from most sites by July. By the end of the summer survey period, 35 of 46 sites showed evidence of flowering, with the lowest observed densities of 3.2 flowers m⁻² and the highest of 638.4 flowers m⁻². Further, they found that all the summer sites with flowers still present were female-biased with one having a balanced proportion of male and female shoots. This is a direct comparison to the observations during the winter sampling period, where the vast majority of locations presented as being male-biased. Seed bank surveys have also occurred. Core samples were successfully taken from 38 sites, of which 17 showed evidence of a *Syringodium filiforme* seed bank. Mesocosm work has also begun, testing the effects of seed sterilization on germination, where seedlings appeared above the sediment surface 17 days after planting. Dr. Fourqurean was selected for an additional year of funding to continue to assess the suitability of *Syringodium filiforme* for seed-based restoration by investigating sexual reproduction, seed set, and seed banks in extensive seagrass meadows in the Florida Keys; and then experimentally determining the cues for promoting germination of seeds using the Mote Marine Lab seagrass raceway facilities.

Title: “The Resiliency of *Halodule wrightii* to Increased Temperature, Freshwater Discharge, and Light Limitation”

Principal Investigators: Dr. Megan Conkling (Florida Atlantic University Harbor Branch Oceanographic Institute)

Co-Principal Investigators: Dr. Rachel Brewton (Florida Atlantic University Harbor Branch Oceanographic Institute), Dr. Iris Segura-Garcia (Florida Atlantic University Harbor Branch Oceanographic Institute)

Project Date: July 2025 – June 2026

Summary: The goal of this one-year study is to provide an initial assessment of how molecular processes and genotypes relate to the resiliency of *Halodule wrightii* from the Indian River Lagoon in response to increased temperature, freshwater discharge, and light limitation. This goal will be achieved in a series of experiments at the Harbor Branch Seagrass Nursery. Experiments will be conducted to determine the impact of environmental stressors on *H. wrightii* health and growth, analyze the molecular response, and identify specific genes that confer resilience. These data will be related to the resilience of *H. wrightii* plants to both short- and long-term exposure of environmental stressors. By identifying if there are desirable genotypes of *H. wrightii* with molecular characteristics that relate to resiliency, the fitness of it and other nursery-grown seagrass species can be improved, leading to more successful restoration efforts. This study is crucial for the integration of assisted evolution strategies into restoration to reinforce seagrass ecosystems, avoid rapid deterioration, and promote their adaptation to local and global pressures.

Title: “Differential gene expression and productivity in response to ocean warming for two *Halodule wrightii* populations across a latitudinal cline”

Principal Investigators: Dr. T. Erin Cox (University of New Orleans)

Project Date: January 2026 – December 2026

Summary: Dr. Cox will look at *H. wrightii* located in waters on Florida’s gulf coast to test (1) whether northern and southern FL populations contrast in adaptive response to heat stress and (2) examine for potential gene regulation involved in resilience of seagrass productivity to a warming event. She will use transcriptome analysis (TagSeq) to examine differential gene expression (DGE) coupled to measures of productivity in a common garden experiment. This experiment will be done at Mote in collaboration with Dr. Dominique Gallery and Liz Longstreet. She will collect twelve cores of *H. wrightii* beds, at a similar depth and salinity, from Apalachicola Bay in the northern panhandle and from FL Keys National Marine Sanctuary (FKNMS) in the southern peninsula. Cores will be brought to Mote submerged into twelve raceways with one replicate per population in each raceway. Plants will acclimate for four weeks, a heat stress will be applied for two weeks, followed by a seven-week recovery period. To gauge whether populations differ in physiology, seagrass productivity will be monitored and leaf samples collected for DGE before, during, and after heat stress is applied in both heat and control replicates. DGE with gene ontology and productivity will be compared for each period for both populations and treatments. Outcomes will inform managers on adaptation potential of two *H. wrightii* populations while identifying genes that may regulate for greater resilience to warming.

Title: “Seagrass Strikes Back: A new hope for fighting Marine heatwaves (MHWs) with Thermo-Priming”

Principal Investigators: Dr. Linda Walters (University of Central Florida)

Co-Principal Investigators: Dr. Robert Fitak (University of Central Florida)

Project Date: July 2025 – June 2026

Summary: This project outlines a groundbreaking approach to restoring coastal seagrass ecosystems by enhancing thermal tolerance of seagrass through thermo-priming. Focusing on *Halodule wrightii*, our multi-disciplinary team with partners from the University of Central Florida, Bethune-Cookman University, Florida Fish and Wildlife Research Institute, St. John’s River Water Management District and Mote Marine Laboratory aim to identify molecular mechanisms critical for heat resilience for this seagrass species.

Title: “Influence of biochar on seagrass growth, health, and ecological interactions”

Principal Investigators: Dr. Toufiq Reza (Florida Institute of Technology)

Co-Principal Investigators: Dr. Ralph Turingan (Florida Institute of Technology)

Project Date: August 2025 – February 2026

Summary: In this one-year project, Florida Tech will develop a biochar technology for stabilization of Indian River Lagoon (IRL) sediments and evaluate the growth and health of shoal grass (*Halodule wrightii*), manatee grass (*Syringodium filiforme*), and turtle grass (*Thalassia testudinum*). Young seagrass for all three types will be collected from Florida Oceanographic Society and genomics will be performed at Mote and uploaded to the genetic library of Florida seagrass. Meanwhile, commercial biochar, procured from Green Carbon Solutions LLC., will be mixed with IRL sediments at various ratios (0-5 %v/v) and a 30-day grow out will be performed at a set of small tubs at Florida Tech using IRL water. Rhizome and biomass growth will be measured after the grow-out phase and sediment and water quality will be chemically characterized to further understand the biochar interaction with the sediments.

Title: “Investigating the Influence of Ocean Acidification on Seagrass Resilience to Nutrient Loading”

Principal Investigators: Dr. Robert Johnson (University of Wisconsin-Madison)

Project Date: July 2025 – July 2026

Summary: Growth and survivorship responses in turtle grass (*T. testudinum*) and shoal grass (*H. wrightii*) to co-occurring environmental stressors will be investigated using laboratory mesocosm experiments. *T. testudinum* and *H. wrightii* will be used because they are widely distributed across Florida, and are both important foundation species in coastal habitats. Seagrass plants will be collected from nearshore meadows in either Pine Island Sound AP or Estero Bay AP in Lee County to be grown in flow-through seawater raceways at Mote’s IC2R3 facility in Summerland Key. Mesocosm experiments will be coupled with in situ characterization of the seagrass and environmental conditions in the source meadows at the time of collection. Scientists will manipulate nutrient availability and pH in experimental mesocosms to evaluate seagrass responses and identify thresholds.

Title: “Using Long-Term Passive Acoustics to Evaluate Seagrass Restoration Efforts”

Principal Investigators: Dr. Jim Locascio (Mote)

Project Date: July 2025 – June 2026

Summary: This project will use passive acoustic recordings to compare the soundscapes of restored and unrestored seagrass beds/failed restoration sites. Soundscapes reflect habitat use by a wide range of taxa, including invertebrates, spawning fishes (e.g., drums), and socializing or foraging marine mammals. By capturing these acoustic signatures, passive recordings can document when and where these behaviors occur. This data serves as a proxy for habitat condition and ecosystem function while also providing a baseline to assess the effectiveness of seagrass restoration efforts. Six monitoring sites—three control and three experimental—will be selected in collaboration with the DEP Aquatic Preserve Program staff and seagrass restoration practitioners.



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