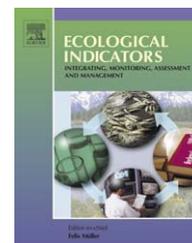


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Eastern oysters (*Crassostrea virginica*) as an indicator for restoration of Everglades Ecosystems

Aswani K. Volety^{a,*}, Michael Savarese^a, S. Gregory Tolley^a, William S. Arnold^b, Patricia Sime^c, Patricia Goodman^c, Robert H. Chamberlain^c, Peter H. Doering^c

^a Coastal Watershed Institute, Florida Gulf Coast University, 10501 FGCU Boulevard South, Fort Myers, FL 33965, United States

^b Fish and Wildlife Research Institute, 100 Eighth Avenue SE, St. Petersburg, FL 33701, United States

^c South Florida Water Management District, 3301 Gun Club Road, West Palm Beach, FL 33406, United States

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ABSTRACT

The Comprehensive Everglades Restoration Plan (CERP) attempts to restore hydrology in the Northern and Southern Estuaries of Florida. Reefs of the Eastern oyster *Crassostrea virginica* are a dominant feature of the estuaries along the Southwest Florida coast. Oysters are benthic, sessile, filter-feeding organisms that provide ecosystem services by filtering the water column and providing food, shelter and habitat for associated organisms. As such, the species is an excellent sentinel organism for examining the impacts of restoration on estuarine ecosystems. The implementation of CERP attempts to improve: the hydrology and spatial and structural characteristics of oyster reefs, the recruitment and survivorship of *C. virginica*, and the reef-associated communities of organisms.

This project links biological responses and environmental conditions relative to hydrological changes as a means of assessing positive or negative trends in oyster responses and population trends. Using oyster responses, we have developed a communication tool (i.e., Stoplight Report Card) based on CERP performance measures that can distinguish between responses to restoration and natural patterns. The Stoplight Report Card system is a communication tool that uses Monitoring and Assessment Program (MAP) performance measures to grade an estuary's response to changes brought about by anthropogenic input or restoration activities. The Stoplight Report Card consists of both a suitability index score for each organism metric as well as a trend score (– decreasing trend, +/- no change in trend, and + increasing trend). Based on these two measures, a component score (e.g., living density) is calculated by averaging the suitability index score and the trend score. The final index score is obtained by taking the geometric score of each component, which is then translated into a stoplight color for success (green), caution (yellow), or failure (red).

Based on the data available for oyster populations and the responses of oysters in the Caloosahatchee Estuary, the system is currently at stage “caution.” This communication tool instantly conveys the status of the indicator and the suitability, while trend curves provide information on progress towards reaching a target. Furthermore, the tool has the advantage of being able to be applied regionally, by species, and collectively, in concert with other species, system-wide.

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* Corresponding author. Tel.: +1 239 590 7216; fax: +1 239 590 7200.

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1. Introduction and background

The Eastern oyster (*Crassostrea virginica*) once supported a Native American subsistence fishery prior to and during early European colonization of North America (Quitmyer and Massaro, 1999) and today continues to be an important economic and ecological resource to coastal inhabitants (Ingle and Smith, 1949; Coen et al., 1999; Gutierrez et al., 2003). Although not commercially harvested in southern Florida estuaries, oysters provide habitat for other estuarine species that have significant recreational and commercial value. Oysters are also ecologically important: they improve water quality by filtering particles from the water and serve as prey and habitat for many other animals (Coen et al., 1999). For example, oyster reefs are home to gastropod mollusks, polychaete worms, decapod crustaceans, boring sponges, fishes, and birds. Over 300 macrofaunal species may associate with oyster reefs and over 40 species may inhabit a single oyster bed (Wells, 1961).

In the Caloosahatchee, Loxahatchee, Lake Worth Lagoon, and St. Lucie Estuaries (Northern Estuaries of the Everglades), oysters have been identified as a Valued Ecosystem Component (VEC; Chamberlain and Doering, 1998a, 1998b). Oysters are natural components of estuaries along the eastern seaboard of the U.S. as well as the Gulf of Mexico and were once abundant in the Northern Estuaries (Systems Status Report 2007). The Eastern oyster possesses a broad geographical distribution and wide temperature and salinity tolerances (Gunter and Geyer, 1955; Cake, 1983) and is the dominant species in these oyster-reef communities. Adult oysters normally occur at salinities between 10 and 30 ppt, but they tolerate salinities of ~2 to 40 ppt (Gunter and Geyer, 1955). Occasional, short pulses of freshwater inflow can greatly benefit oyster populations by reducing predator (e.g., oyster drill, whelk) and parasite (e.g., *Perkinsus marinus*) impacts (Owen, 1953), but excessive freshwater inflow may kill entire populations of oysters (Gunter, 1953; Schlesselman, 1955; MacKenzie, 1977; Volety et al., 2003; Volety and Tolley, 2005; Bergquist et al., 2006). Reefs located near the head of an estuary, where salinities range from 0 to 15 ppt, are sparsely populated due to frequent flooding and high mortality rates (Butler, 1954; Volety and Savarese, 2001; Savarese et al., 2003). Spat recruitment and juvenile growth rates are also low in this location. Where salinities are between 15 and 20 ppt, populations are dense, reproductive activity is high, predator numbers are low, and spat recruitment and growth rates are high (Shumway, 1996; White and Wilson, 1996). Toward the higher salinity waters near the mouth of the estuary, oyster reefs are sparse, spat recruitment and growth are low, diseases and predators are high, and suitable substrate is often lacking. Salinity also affects gametogenesis, condition index, spawning, and disease in oysters (Shumway, 1996). Salinities <5 ppt impair gametogenesis while normal gametogenesis occurs above 7.5 ppt; oysters from Texas, for example, showed suppressed gonadal activity at salinities <6 ppt (Shumway, 1996). Similar trends were observed in Caloosahatchee River oysters in 2003 in response to regulatory freshwater releases (Volety, unpublished results).

Additionally, the protozoan parasite *P. marinus* has devastated oyster populations in the Atlantic (Burreson and Ragone-Calvo, 1996) as well as in the Gulf of Mexico (Soniati, 1996), where

it is currently the primary pathogen (Dermo disease) of oysters. Andrews (1988) estimates that *P. marinus* can kill ~80% of the oysters on a reef. Temperature and salinity influence the distribution and prevalence of *P. marinus* with higher values favoring the parasite (Burreson and Ragone-Calvo, 1996; Soniat, 1996; Chu and Volety, 1997; La Peyre et al., 2003). Laboratory studies by Chu and Volety (1997) suggest that although temperature is important, salinity is the most important factor influencing the disease susceptibility and disease progression of *P. marinus* in oysters. High salinities also attract various predators such as crabs, starfish, boring sponges, and oyster drills, along with Dermo disease (Butler, 1954; Hopkins, 1962; Galtsoff, 1964; Menzel et al., 1966; Shumway, 1996; Livingston et al., 2000). Therefore, the quality (e.g., nutrients, contaminants, suspended sediments), quantity, timing, and duration of freshwater inflow have a tremendous effect on oyster health, survival, growth, and reproduction, and thus the biological responses of oysters are directly related to freshwater-influenced environmental conditions.

1.1. Indicator history

Oysters are common throughout the estuarine portions of the Northern (Caloosahatchee, St. Lucie, Loxahatchee, and Lake Worth Lagoon) and Southern (Whitewater Estuary, Shark River, Coot Bay, Oyster Bay, and areas of the Ten Thousand Islands) Estuaries (Fig. 1). Water management and dredging practices have had a major impact on the presence, density, and distribution of oysters within the mesohaline areas of the Northern and Southern Estuaries. Historically, drainage patterns were characterized by gentle, meandering surface water flows through rivers, creeks, and sloughs and overland sheet flow through contiguous marshy areas. This natural system absorbed floodwater, promoted ground water recharge, assimilated nutrients, and removed suspended materials (ACOE and SFWMD 2002). As South Florida developed, the canal network, built as part of the Central and Southern Florida Flood Control Project, worked very efficiently in preventing floods and drastically altered the quantity, quality, timing, and distribution of freshwater entering the estuaries. Freshwater flow into the estuaries and their tributaries increased both in volume and frequency (often to prevent flooding) relative to the pre-drainage era. This caused rapid, often within a few hours, changes in salinity resulting in degradation of the biological integrity of the estuaries. Furthermore, inflow is often too great in the wet season and too little in the dry season to support a healthy estuary. Additionally, flood releases and inland runoff contain numerous contaminants from urban and agricultural development including excess suspended solids, nutrients, pesticides, and other Emerging Pollutants of Concern such as hormones and pharmaceuticals. This results in poor quality water entering the estuaries.

Although the Caloosahatchee Estuary (Fig. 2) is used as a specific example below, similar water quality concerns are present in all Northern and Southern estuaries given the similarities in watershed alteration. The Caloosahatchee River is the major source of freshwater for the Caloosahatchee Estuary (CE) and southern Charlotte Harbor. The river, which has been transformed into a canal (C-43), conveys both runoff from the Caloosahatchee watershed and regulatory releases

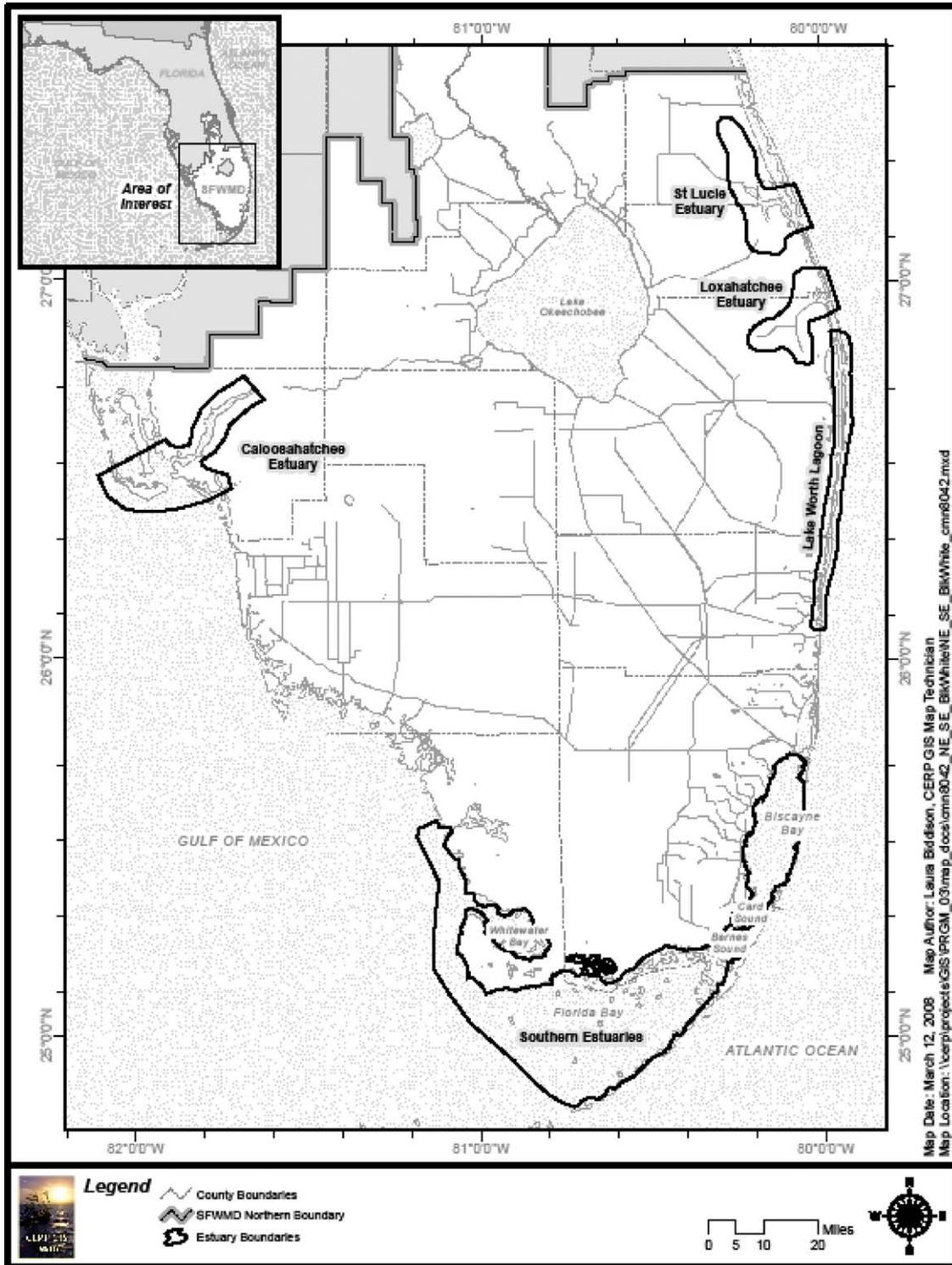


Fig. 1 – Location of Northern (Caloosahatchee, St. Lucie, Loxahatchee, and Lake Worth Lagoon) and Southern (Whitewater Estuary, Shark River, Coot Bay, Oyster Bay, and areas of the Ten Thousand Islands) Estuaries in Florida.

from Lake Okeechobee. The C-43 canal has undergone a number of alterations to facilitate this increased freshwater discharge and improve flood protection: channel straightening and enlargement, bank stabilization, the development of an intricate network of ancillary canals within the watershed,

and the addition of three locks and dams. The final downstream structure, the W.P. Franklin Lock and Dam (S-79), demarcates the beginning of the estuary and acts as a barrier to salinity and tidal action, which historically extended much farther upstream. All of these alterations to the Caloosa-

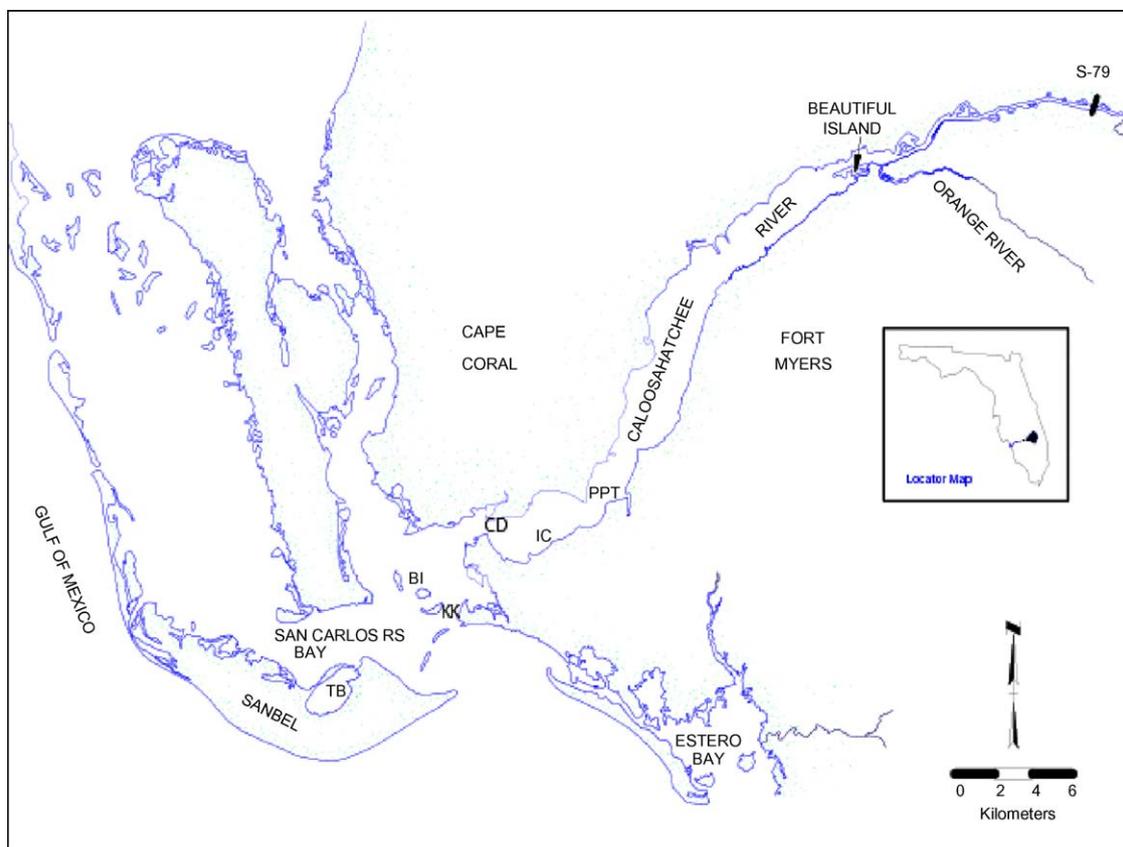


Fig. 2 – Sampling locations within the Caloosahatchee Estuary. Locations (PPT = Pepper Tree Point, IC = Iona Cove/Shell Point, CD = Cattle Dock, BI = Bird Island, KK = Kitchel Key, and TB = Tarpon Bay) are from upstream to downstream along a salinity gradient.

hatchee River and its watershed have resulted in a drastic change in freshwater inflow to the ecosystem downstream resulting in large fluctuations of salinity and water quality that adversely impact estuarine biota (Chamberlain and Doering, 1998a; Sklar and Browder, 1998).

The dominant biological features in the downstream portion of the Caloosahatchee River estuary are its numerous mangrove islands and many kilometers of mangrove shoreline, which are often closely associated with oysters. Because of its biotic richness and aesthetic appeal, the Caloosahatchee Estuary supports a wide variety of recreational and fishery activities with significant economic value. The natural resources of this area are also impacted by large freshwater releases and are threatened by long-term shifts in water quantity and quality (Chamberlain and Doering, 1998b; Doering et al., 2002; Volety et al., 2003). Under current water management practices, the estuarine portion of the river is essentially fresh during the wet season due to freshwater releases and runoff from summer rains (Volety et al., 2003).

During the dry winter season freshwater releases are halted and the estuary becomes hypersaline because of its high ratio of surface area to volume and resultant evaporative loss. Freshwater releases during summer months flush oyster larvae downstream to locations that have unsuitable substrate, or create salinity conditions that are unfavorable

(<5 ppt) for larval survival in the estuarine portions of the river. Oysters in Southwest Florida spawn continuously, with peak recruitment (spat settlement) occurring May to November. Recruitment near Shell Point and possibly upstream begins to peak in March, a full 3 months earlier than in San Carlos Bay, located farther downstream, rendering these newly settled juveniles vulnerable to large releases from S-79, which are often made during this period to regulate Lake Okeechobee water level for flood protection. Large freshwater flows at this time and during the summer also expose oyster larvae to lethally low salinities or flush the larvae downstream to locations where there may not be suitable substrate for settlement (Volety et al., 2003).

Recent investigations in the CE have estimated the optimum quantity of freshwater needed to protect key biota. These VEC species (oysters and sea grasses), help sustain the ecological structure and function of the estuary by providing food, living space, and foraging sites for other estuarine species. Oysters and submerged aquatic vegetation (SAV) represent VECs in the CE. Proper management (e.g., frequency, timing, quantity, and quality) of water releases to the estuary will protect these species and should lead to the restoration of healthy and diverse estuarine ecosystems.

Work by Tolley and Volety (2005) and Volety et al. (2003) documents the importance of *C. virginica* as a VEC. They found that a greater abundance of decapods and fishes was

associated with clusters of live oysters compared to clusters of dead oysters, and that the structure provided by both living and dead oyster shells supported a greater abundance than no shells. Species richness and biomass were also higher for samples with oyster clusters (dead or live) compared to controls with no oyster shell (Tolley and Volety, 2005). This study suggests that the real significance of living oysters to habitat value lies not only in creating a three-dimensional structure, but also in maintaining this structure of clusters through time. In the absence of oyster-reef growth, individual oysters within a cluster or bed may die, leaving empty compartments for reef residents, but mass mortality of a cluster results in the disarticulation and eventual loss of the oyster shells (Volety et al., 2003; Tolley and Volety, 2005). Therefore freshwater or habitat alteration unfavorable to oysters will not only result in decreases in the extent of oyster reefs and filtration-improved water quality, but also the species residing on the oyster reefs.

Volety et al. (2003) evaluated adult and juvenile oyster survival, the prevalence and intensity of disease, and oyster recruitment success. These responses were compared to environmental factors including salinity and freshwater flow from S-79. Oysters grew best at a salinity of 14–28 ppt. Infection by the oyster pathogen *P. marinus* increased during periods of higher salinity and temperature. Field studies determined that the prevalence of infection was high, but disease intensity was low, likely resulting from an antagonistic interaction between temperature and salinity (i.e., high summer temperature occurs during the wet season when salinity is low). Therefore, freshwater releases to diminish *P. marinus* are generally not necessary during warm summer months and could potentially threaten oyster populations through further reductions in salinity.

In the CE, the greatest oyster growth and recruitment occurs during the wet season, but slower growth and poor spat production occur at salinities below 14 ppt. Volety et al. (2003) found that salinity conditions were best suited for oyster growth just upstream of Shell Point in the CE; however, this area is also the most vulnerable to high mortality when large freshwater releases cause salinity to fall below threshold tolerance (<5 ppt), sometimes for prolonged periods (>1–2 weeks). These authors further report that although adult oysters are tolerant of variable salinity, salinities ≤ 5 ppt result in >95% mortality of juvenile oysters. High mortality can occur when juveniles are exposed to this salinity for just a week (Volety et al., 2003).

Experimental results indicate that adults are able to tolerate salinities as low as 5 ppt for up to 8 weeks, but cannot tolerate salinities lower than 3 ppt, which can occur at upstream portions of the CE when S-79 discharges exceed 4000 cubic feet per second (cfs) (Volety et al., 2003). Therefore, high discharges can limit survival and abundance in this region where oysters were historically present. Volety et al. (2003) indicated that since high spat recruitment, fast growth, and low disease incidence and intensity are found at intermediate salinities (10–20 ppt), it is feasible to reestablish oyster reefs within the CE by strategically deploying oyster cultch in suitable areas, as long as freshwater releases are managed to maintain salinity within the range of normal oyster tolerances (5–35 ppt).

1.2. Comprehensive Everglades Restoration Plan

The implementation of the Comprehensive Everglades Restoration Plan's (CERP) Monitoring Assessment Plan (MAP) will help determine how well CERP is meeting its restoration goals and objectives. The premise of CERP is that restoring hydrology in the Northern and Southern Estuaries will improve the spatial and structural characteristics of oyster reefs and improve recruitment and survivorship of *C. virginica* and associated fauna. The hypotheses below are the result of a conceptual model of stressors that impact oysters, oyster reefs, and secondary habitat (Fig. 3; SSR, 2007). The conceptual model is a product of the known cause and effect relationships between stressors and *C. virginica* ecological and physiological responses from published studies for this region (cited previously) and elsewhere throughout the species' range. Consequently, the hypotheses have withstood rigorous testing and therefore serve as guideposts for restoration. Correlations between biological responses and environmental conditions relative to hydrological changes contribute to the assessment of positive or negative trends in restoration. Restoration success or failure related to the oyster indicator can be evaluated by comparing recent monitoring efforts and future trends and health status of oyster reefs in the Northern Estuaries, unaltered or control estuaries, and model predictions (e.g., Habitat Suitability Index), as stated in the CERP hypotheses related to the oysters (CERP Monitoring and Assessment Plan sections 3.3.3.6; RECOVER 2004, SSR, 2007).

1.3. CERP MAP hypotheses related to the Eastern oyster indicator

The following hypotheses developed for the Eastern oyster and adopted for CERP-MAP serve as defining criteria for the evaluation of Everglades Restoration.

- Sudden and drastic changes in the estuarine salinity envelope can result in decreased survival, reproduction, spat recruitment, and growth, and increased susceptibility to diseases by *P. marinus* and MSX.
Rationale. Large rainfall events or large volume releases from Lake Okeechobee (e.g., Caloosahatchee Estuary) cause large volumes of freshwater over a short period of time to enter the estuaries resulting in a sudden drop in salinity. This abrupt reduction can lead to significant mortality in the oyster population as well as decreased growth, reproduction, and spat recruitment. Extreme droughts can also negatively impact oysters by making them more prone to disease and predation.
- Accumulation of muck (i.e., sediment with high organic content) will render available substrate unsuitable for oyster larval settlement and thus for recruitment and growth of larval oysters. In addition, accumulation of muck may also impact dissolved oxygen content making the substrate unsuitable for larval settlement and growth.

Rationale. Oysters recruit successfully to immobile sandy or shell-rich substrates, while recruitment is negatively affected by the accumulation of mucky sediments. Freshwater releases from the Lake and inland canals

deposition of phytoplankton and suspended detritus in the form of mucus and uneaten food (Newell, 1988; Newell and Langdon, 1996; Coen et al., 1999). Oyster reefs also provide habitat, shelter, and food for over 300 species (Wells, 1961; Coen et al., 1999). Many of the crustaceans and fishes that are members of oyster-reef communities are important prey for secondary and tertiary carnivores such as fishes and birds (Tolley and Volety, 2005). Furthermore, biomass and community structure of these oyster-reef communities are directly linked to hydrology and oyster-reef survival and morphology (Tolley et al., 2005, 2006).

1.4.2. *The indicator is feasible to implement and is scientifically defensible*

Relatively long-term data sets exist for oyster reefs at numerous sites in Southwest Florida, spanning a course of 8 years in some cases (Volety, 2007), and there are existing funded cooperative research and monitoring programs supported by RECOVER with Florida Gulf Coast University and the Florida Fish and Wildlife Conservation Commission to continue this type of work. In addition, reliable models and scientific studies exist to predict the impacts of water management on these populations (Cake, 1983; Soniat and Brody, 1988; Wilber and Bass, 1998; Livingston et al., 2000; Barnes et al., 2007), and oysters have been included as part of the CERP Habitat Suitability Index model (Volety et al., 2005). Pattern metrics (e.g., abundance, density, survival, spat recruitment, disease prevalence, and condition index) are also statistically correlated to ecosystem drivers (Volety et al., 2003; Volety, 2007).

1.4.3. *The indicator is sensitive to system drivers (stressors)*

The peer-reviewed literature detailing the responses of oysters to environmental stressors is somewhat extensive (for reviews see Shumway, 1996; Kennedy, 1996). System drivers (e.g., rainfall, water quantity, water quality, sediment loads) have been statistically correlated to species abundance and indicators of oyster health such as density, survival, spat recruitment, disease prevalence, and condition index (Volety et al., 2003; Wilber and Bass, 1998; Livingston et al., 2000; SSR, 2007). Oyster abundance, density, survival, and health indices have been causally linked to hydrological factors such as water salinity, the frequency of killing floods, sedimentation, and contaminants in the water (Wilber and Bass, 1998; Livingston et al., 2000; Volety et al., 2003; Bergquist et al., 2006). As a result, high and low salinity estuaries have been demonstrated to possess distinct oyster abundance, distribution, and health responses (Volety and Savarese, 2001; Volety et al., 2003).

1.4.4. *The indicator is integrative*

Oyster survival, abundance, and distribution are linked to water quality, phytoplankton production, sedimentation, and, in turn, crustacean, fish, and bird success are linked to oyster-reef health and abundance (Cake, 1983; Reinhardt and Mann, 1990; Coen et al., 1999; Bergquist et al., 2006). Furthermore, oyster-reef community responses are correlated with changes in hydrology (e.g., salinity and freshwater inflow) and can therefore be linked to water management (Tolley et al., 2005, 2006).

1.4.5. *Goals and performance measures are established in the RECOVER MAP for the indicator and the following metrics are being monitored*

- Number of live oysters per square meter.
- Number of acres of oyster reefs.
- Condition index of live oysters.
- Disease prevalence and intensity of *P. marinus* in oysters.
- Larval/spat recruitment and reproductive potential.
- Temperature and salinity of water near the reefs.

2. Communicating the oyster indicator

2.1. Methods and development of the indicator

The CERP MAP hypotheses related to the Eastern oyster and how stressors impact them are detailed in Section 1. These were derived through the development of the conceptual model of the stressors influencing oysters (Fig. 3). As previously mentioned, the oyster responses listed below are being measured in all of the Northern Estuaries, but the Caloosahatchee River will be used as an exemplar. The results presented below are from six sampling locations along the estuarine axis (salinity gradient) in the Caloosahatchee Estuary collected over 5–7 years (Fig. 2). Sample temporal trends are presented below; however, for the sake of brevity detailed statistical analyses of the data along with the temporal and spatial variation of oyster responses are not presented. Details of the spatial and temporal trends can be found in Volety et al. (2003), Volety (2007), and SSR (2007). Since the objective of this study is to use the available oyster performance data from all five sampling locations in the CE to derive an indicator score for the estuary, this study does not examine the causes of spatial and temporal variation of oyster responses.

2.2. The metrics and performance measures

2.2.1. Metrics

The oyster indicator uses:

- density of living oysters (per square meter);
- condition index;
- reproductive activity (gonadal condition);
- larval recruitment;
- disease prevalence and intensity of *P. marinus* (and MSX in east coast estuaries);
- growth and survival;
- coverage of oysters in the estuary (# acres).

2.2.2. Performance measures

- density of living oysters (per square meter);
- condition index;
- reproductive activity (gonadal condition);
- larval recruitment;
- disease prevalence and intensity of *P. marinus* (and MSX in east coast estuaries);
- growth and survival.

All of these parameters are correlated with hydrological conditions including depth, flow, salinity, temperature, dissolved oxygen, season, spatial extent, and water quality. Salinity is a critical parameter in estuarine habitats. CERP RECOVER targets for oyster performance measures are based on patterns that are considered natural for the Northern Estuaries along the east and west coasts of Florida.

3. Methods

3.1. The Stoplight Report Card system applied to oysters—determination of thresholds for success (green), caution (yellow) or failure (red)

The Stoplight Report Card system is a communication tool that uses MAP performance measures to grade an estuary's response to anthropogenic or restoration inputs. Questions or

decision rules are developed for each performance measure and translated as suitability curves. Two questions are addressed using suitability curves: (1) Have we reached the restoration target? And (2) are we making progress toward targets? Finally, results are translated into a stoplight display (see below for example).

The system-wide indicator communication tool is based on RECOVER MAP ecological attributes and performance measures. This communication tool has been designed to distinguish between the effects of restoration projects and natural phenomena, assuming that the available data cover periods of high, normal, and low rainfall (and inflow) years. Targets for performance measures are established from historical data or reference sites. By using spatially referenced suitability indices the indicator communication tool can be linked directly to both the RECOVER evaluation and assessment processes and the Task Force indicator assessments. The communication tool instantly conveys the status of the

Table 1 – Decision rule questions for forming performance measure/suitability relationships for the oyster indicator communication tool

1. What is the current living density, in individuals per square meter, of oysters in the Caloosahatchee Estuary. Use yearly average of twice a year (wet and dry season) sampling.			
a.	0–200	Score: 0	Red
b.	>200–800	Score: 0.5	Yellow
c.	>800–4000	Score: 1.0	Green
2. What is the current condition index of oysters in the Caloosahatchee Estuary? Use the yearly average of monthly sampling.			
a.	0–1.5	Score: 0	Red
b.	>1.5–3.0	Score: 0.5	Yellow
c.	>3.0–6.0	Score: 1.0	Green
3. What is the current gonadal condition of oysters in the Caloosahatchee Estuary? Use the yearly average of monthly sampling.			
a.	0–1	Score: 0	Red
b.	>1–2	Score: 0.5	Yellow
c.	>2–4	Score: 1	Green
4. What is the current spat recruitment of oysters (spat/shell) in the Caloosahatchee Estuary? Use mean spat/shell/month for the estuary of monthly sampling.			
a.	0–5	Score: 0	Red
b.	>5–20	Score: 0.5	Yellow
c.	>20–200	Score: 1.0	Green
5. What is the current growth of juvenile oysters in mm/month? Use yearly average of monthly sampling.			
a.	0–1	Score: 0	Red
b.	>1.0–2.5	Score: 0.5	Yellow
c.	>2.5–5	Score: 1.0	Green
6. What is the prevalence of <i>Perkinsus marinus</i> (% of infected oysters) in oysters from the Caloosahatchee Estuary? Use the yearly average of monthly sampling.			
a.	0–20	Score: 1	Green
b.	>20–50	Score: 0.5	Yellow
c.	>50–100	Score: 0	Red
7. What is the intensity of <i>Perkinsus marinus</i> (scale 0–5) in oysters from the Caloosahatchee Estuary? Use the yearly average of monthly sampling.			
a.	0–1	Score: 1	Green
b.	>1–3	Score: 0.5	Yellow
c.	>3–5	Score: 0	Red
Trend question			
a.	– slope	Score: 0	Red
b.	No slope	Score: 0.5	Yellow
c.	+ slope	Score: 1.0	Green

A score of 1.0 for a performance measure is the restoration target. All performance measures are averages of 2–5 years. The trend is based on the data availability at various estuaries (5–8 years) and is presented separately.

Table 2 – Translation table for converting the suitability or trend index for a performance measure or indicator into an index score and stoplight color

Index range	Index score	Stoplight color
0.0–0.3	0	Red ●
>0.3–0.6	0.5	Yellow ●
>0.6–1.0	1.0	Green ●

indicator, and the suitability and trend curves provide information on progress towards reaching a target. This indicator communication tool has the advantage of being able to be applied regionally, by species, and collectively system-wide. Importantly, both targets and suitability curves can be adapted as science improves our understanding of Greater Everglades Ecosystems.

Oyster response data for various metrics are available for 5–7 years for the Caloosahatchee Estuary and were thus chosen as an illustrative example. Tables 1 and 2 present the decision rules used to create the suitability scores. Data in excess of 5 years are not available for all of the responses being measured; therefore, in addition to available data (Table 4), a hypothetical data set using the ranges of oyster responses in the Caloosahatchee Estuary was also used to generate an index score (Fig. 4) and a trend score (Fig. 5) (Table 3). For most of the oyster performance measures empirical data were used from study sites (Volety, 2007; Systems Status Report, 2007). Both targets and curves can be modified as more is learned about this indicator. Trends for each performance measure are determined from a five-year plot of the performance measure (Fig. 5). All metrics were measured at each station monthly while the living density was measured twice a year (wet and dry seasons). The component score (e.g., living density) is the average of the suitability index score and the trend score (Table 1). Tables 3 and 4 provide a hypothetical example of how component scores can be combined into a system-wide indicator score.

3.2. Oyster metrics

3.2.1. Water quality determination and relationship between freshwater inflows and salinity

Water quality measurements were taken in association with sample collection (oysters) using a YSI data sonde. Tempera-

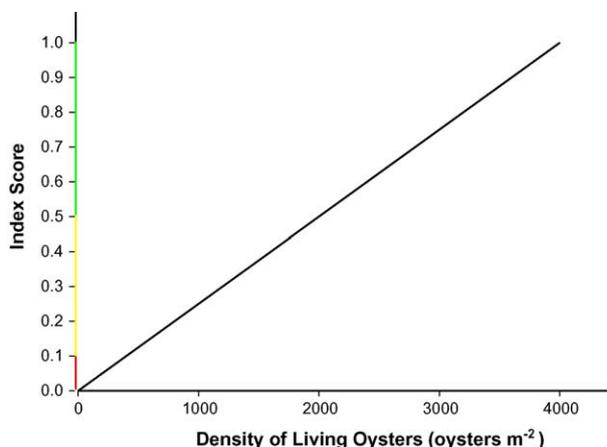


Fig. 4 – Example of linear suitability curve for an oyster indicator performance measure (living density). The x-axis varies for each performance measure as described in Table 1.

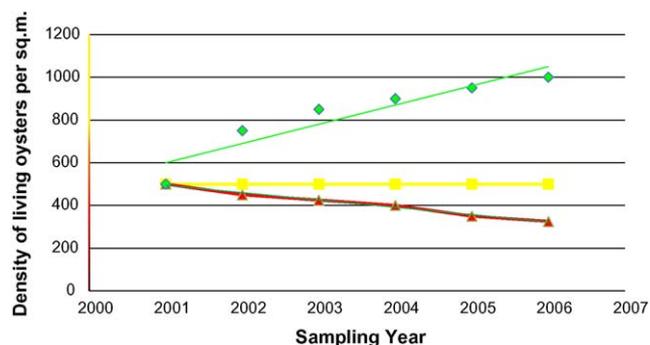


Fig. 5 – Example of trend curves for a oyster performance measure (living density). The green line is a positive trend and gets a score of 1.0; the yellow line indicates no change and is scored 0.0; and the red line is a negative trend (away from the target) and is scored 0.0.

ture, salinity, and dissolved oxygen were measured at the surface (given the shallowness of the environment). Freshwater inflows (cubic feet per second; cfs) into the Caloosahatchee Estuary from S-79 lock and dam were obtained from

Table 3 – Hypothetical example of translating performance measures into a stoplight display

Component	Parameter value	Parameter value stoplight	Index score	Trend	Trend stoplight	Trend score	Average component score	Component stoplight
Oysters								
Living density (per m ²)	50	●	0	+	●	1	(0 + 1)/2 = 0.5	●
Condition index	2.3	●	0.5	–	●	0	(0.5 + 0)/2 = 0.25	●
Spat recruitment per shell	2	●	0	±	●	0.5	(0 + 0.5)/2 = 0.25	●
Juvenile growth (mm)	2	●	0.5	±	●	0.5	(0.5 + 0.5)/2 = 0.5	●
<i>Perkinsus marinus</i> prevalence	15	●	1	+	●	1	(1 + 1)/2 = 1	●
<i>Perkinsus marinus</i> intensity	1.5	●	0.5	+	●	1	(0.5 + 1)/2 = 0.75	●
Geometric mean of oyster component scores								
$(0.5 \times 0.25 \times 0.25 \times 0.5 \times 1 \times 0.75)^{1/6} = 0.477$								
Final Eastern oyster index score = 0.5								

Table 4 – Component score for oysters in the Caloosahatchee Estuary for translating performance measures into a spotlight display

Component	Parameter value	Parameter value spotlight	Index score	Trend	Trend spotlight	Trend score	Average component score	Component spotlight
Oysters								
Living density (per m ²)	1029	●	1	±	●	0.5	(1 + 0.5)/2 = 0.75	●
Condition index	2.96	●	0.5	±	●	0.5	(0.5 + 0.5)/2 = 0.5	●
Gonadal Index	2.61	●	1	±	●	0.5	(1 + 0.5)/2 = 0.75	●
Spat recruitment per shell	6.43	●	0.5	±	●	0.5	(0 + 0.5)/2 = 0.5	●
Juvenile growth (mm/month)	2	●	0.5	±	●	0.5	(0.5 + 0.5)/2 = 0.5	●
<i>Perkinsus marinus</i> prevalence	49.5	●	0.5	–	●	0	(0.5 + 0)/2 = 0.25	●
<i>Perkinsus marinus</i> intensity	0.83	●	1	–	●	0	(1 + 0)/2 = 0.5	●
Geometric mean of oyster component scores (0.75 × 0.5 × 0.75 × 0.5 × 0.5 × 0.25 × 0.5) ^{1/7} = 0.508								
Final Eastern oyster index score = 0.5								

the South Florida Water Management District (courtesy of Dr. P. Doering and K. Haurert). The relationship between freshwater inflow and salinity at various sampling locations was determined using power regressions (SPSS). Salinity at various sampling locations obtained during the sampling period was regressed against the 30-day moving average flows for the sampling month.

3.2.2. Estimating oyster density

Density of living oysters, an indirect measure of reef productivity, also varies considerably along an estuarine salinity gradient. Patterns attributable to human alterations in freshwater flow were detected previously in the Blackwater and Faka Union estuaries of the Ten Thousand Islands (Volety and Savarese, 2001). Oyster living density at CE sampling stations was measured during the late fall and early spring every year. This period is ideal for density measurement because oysters have reproduced for the year and spat have settled from the water column. Four 0.25-m² quadrats were randomly located at the mean-low-tide height at each reef. The number of living oysters within each quadrat were counted and compared among reefs at various locations.

3.2.3. Determining oyster condition index

The physiological condition of an oyster can be measured by its condition index—the ratio of meat weight to shell weight (Lucas and Beninger, 1985). Although oysters tolerate salinities between 0 and 42 ppt, growth is maximized at salinities of 14–28 ppt; slower growth, poor spat production, and excessive valve closure occur at salinities below 14 ppt (Shumway, 1996). Because the metabolic energy remaining after daily maintenance is converted into biomass, an oyster stressed either by poor water quality or by disease has less energy for growth or reproduction. Consequently, a comparison of oyster condition index among oyster reefs located along the salinity gradient should be indicative of oyster health and the influence of salinity and disease. Oysters from an altered estuary having extreme salinities have significantly lower condition index compared to oysters from an unaltered estuary (Volety and Savarese, 2001). Oysters were collected for condition index determination monthly at the same time disease prevalence was surveyed.

3.2.4. Gonadal Index

Histological analysis was used to examine gonadal state and reproductive potential of oysters from different sites (see above) during the study period. Gametogenic stage was identified under a microscope according to Fisher et al. (1996) and the International Mussel Watch Program (1980). Samples of 10–15 oysters were collected monthly from each sampling location. For histological sectioning, a 3–5 mm thick band of tissue was cut transversely with a razor blade in such a manner as to contain portions of mantle, gill, digestive tubule, and gonad. Dissected tissue was fixed for 48–72 h in Davidson’s fixative and stored in 70% ethanol before paraffin embedding. After embedding, sections were made with a microtome, and slides were stained with hematoxylin and eosin. Gonadal portions of the sections were observed by light microscopy to determine sex and gonadal condition (Fisher et al., 1996).

A description of gametogenic characteristics (reproductive staging) observed in histological sections of oyster gonads and the values assigned for the determination of gonadal condition are presented in Appendix 1 (International Mussel Watch Program 1980; Fisher et al., 1996). Values of 6–10 were converted to 5–1 to reflect true reproductive activity (similar % of mature gametes in the follicles) for statistical purposes. For example, in a random sample, if five oysters are at stage 1 and 5 oysters at stage 10, stages that have no mature gametes, when averaged the value is 5.5, which erroneously suggests that oysters are actively spawning. Changing the values from 6 to 10 would yield values that accurately reflect the reproductive stage of the oysters (Volety and Savarese, 2001; Volety et al., 2003).

3.2.5. Spat recruitment

Oyster spat recruitment experiments were conducted using old adult oyster shells strung together by a weighted galvanized wire and deployed at sampling locations. A shell string consisting of 12 oyster shells, each 5.0–7.5 cm long with a hole drilled in the center and oriented inner surface down, was suspended off the bottom at various sites (Haven and Fritz, 1985). Oyster spat settlement was monitored monthly by counting the number of spat settled on the underside of strung shells. Spat settlement is expressed as the number of spat settled per oyster shell per month.

3.2.6. Juvenile oyster growth and survival

One to two hundred juvenile oysters (10–20 mm) were deployed at all sampling locations in 0.5 mm closed wire-mesh bags in the fall months (a period that coincides with natural spawning period). Fifty randomly selected oysters were measured to the nearest 0.1 mm every month from each location. Juvenile oysters were placed at the sampling locations in wire-mesh bags to exclude predation and indicate growth and/or mortality due to water quality.

3.2.7. Estimating oyster disease prevalence and distribution

P. marinus disease susceptibility in oysters along the salinity gradient within the Caloosahatchee Estuary was determined at six locations. A total of 10–15 oysters per location were collected monthly throughout the year. Oysters were assayed for the presence of *P. marinus* using Ray's fluid thioglycollate medium technique (Ray, 1954; Volety et al., 2000, 2003). Samples of gill and digestive diverticulum were incubated in the medium for 4–5 days. *P. marinus* meronts enlarge in the medium and stain blue-black with Lugol's iodine allowing for visual identification under a microscope. Prevalence of infection was calculated as % infected oysters. The intensity of infection was recorded using a modified Mackin scale (Mackin, 1962) in which 0 = no infection, 1 = light, 2 = light-moderate, 3 = moderate, 4 = moderate-heavy, 5 = heavy.

4. Results

4.1. Water quality determination and relationship between freshwater inflows and salinity

As expected, temperatures at the sampling locations in the CE were higher during the warmer summer—early fall months (April–October) and were lower during the cooler drier months (November–March). In contrast, salinities at sampling locations were lower during the summer—early fall months (June–October) and higher during the cooler months (November–May; results not shown). There was a significant relationship between flows and salinity at the five sampled locations ($P < 0.001$; $R^2 = 53\text{--}75\%$ depending on the sampling location; Fig. 6). The influence of freshwater inflow on the system is more pronounced at the upstream locations compared to the downstream locations.

4.2. Oyster density

Inter-annual variation of oyster density ranged between 102 and 2345 oysters m^{-2} at various sampling locations. Because density data were only available for two years, a trend analysis was not computed. Mean density for all the sampling locations in the CE in 2006 and 2007 ranged from a low of 710 ± 663 (2006 wet season) to a high of 1296 ± 997 oysters m^{-2} (dry season 2006).

4.3. Oyster condition index

Condition index of oysters varied significantly among sampling locations and sampling months ($P < 0.001$). Condition index in oysters was higher from December to May and lowest

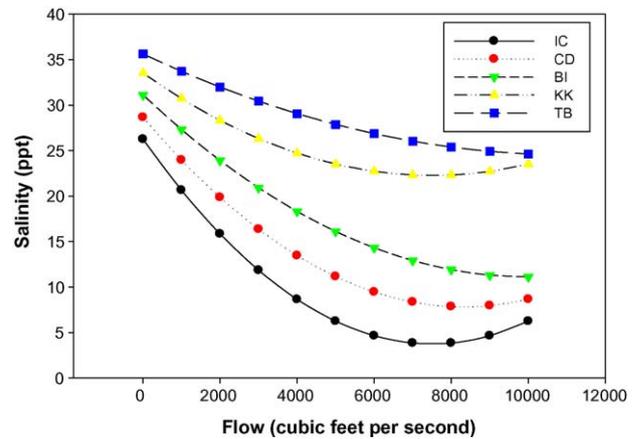


Fig. 6 – Relationship between freshwater inflows into the Caloosahatchee Estuary and salinities at various points in the estuary. Monthly means of the 30-day moving average salinity were regressed against salinity from the sampling locations. Results suggest that increasing freshwater inflow decreases the salinities at various locations in the estuary.

in October. Condition index decreased through March–October (Fig. 7), a period that coincided with oyster spawning.

4.4. Gonadal stage

Gonadal Index, a measure of the reproductive stage of spawning oysters, was very cyclical and varied significantly among sampling locations and sampling months ($P < 0.001$). Gonadal Index was higher during April–October, suggesting active spawning of oysters, and lower during November–March (Fig. 8).

4.5. Spat recruitment

Spat recruitment of oysters varied significantly among sampling locations and sampling months ($P < 0.001$). Recruitment

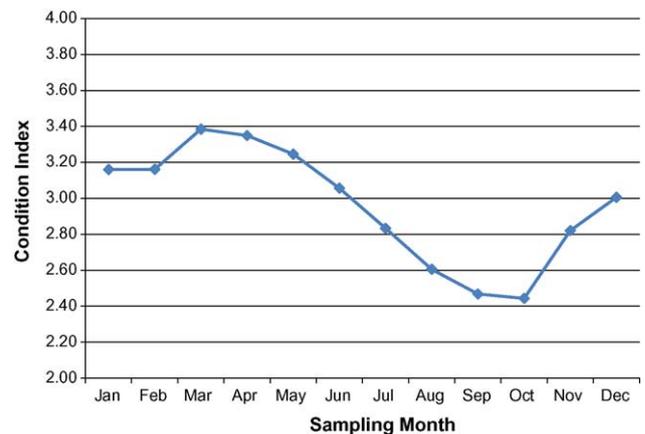


Fig. 7 – Mean condition index of oysters from all the sampling locations in the Caloosahatchee Estuary. 10–15 oysters were collected monthly from each of the sampling locations between August 1999 and January 2008.

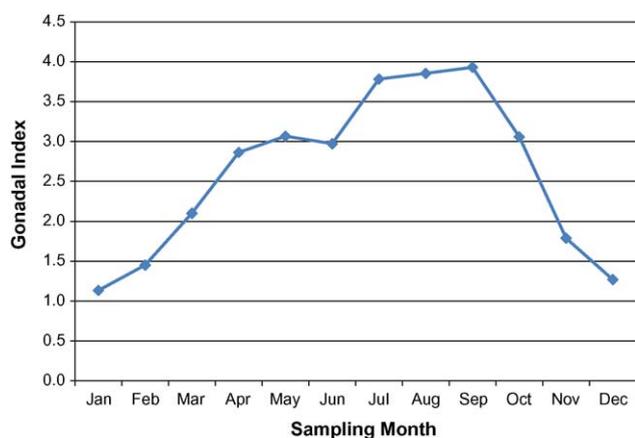


Fig. 8 – Mean gonadal stage of oysters from all the sampling locations in the Caloosahatchee Estuary. 10–15 oysters were collected monthly from each of the sampling locations between August 1999 and September 2007.

of spat was higher from April to October, with peak recruitment occurring in August. Little or no spat recruitment was observed between November and March (Fig. 9).

4.6. Juvenile oyster growth

Juvenile oyster growth and mortality varied widely among sampling locations and sampling months. Significant juvenile mortality was observed when oysters were deployed during the summer months when salinities were typically low (results not shown). When oysters were deployed in late fall months (October to December) when salinities were higher, faster growth was observed at the upstream locations, which tended to have more estuarine salinities, compared to downstream locations where salinities were marine to hypersaline. Mortality rates were typically 60–100% depending on salinity (results not shown).

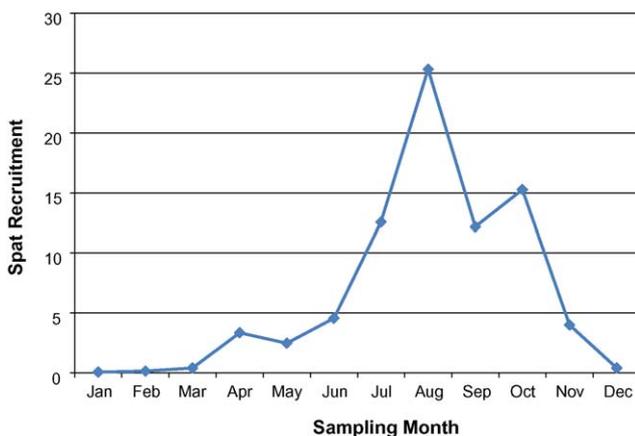


Fig. 9 – Mean spat recruitment (spat/shell) of oysters from all the sampling locations in the Caloosahatchee Estuary. Data were collected monthly from each of the sampling locations between August 1999 and January 2008.

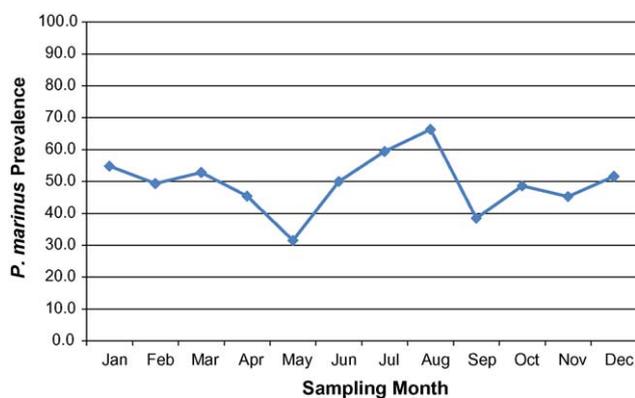


Fig. 10 – Mean prevalence of Perkinsus marinus (% of infected oysters) from all the sampling locations in the Caloosahatchee Estuary. 10–15 oysters were collected monthly from each of the sampling locations between August 1999 and January 2008.

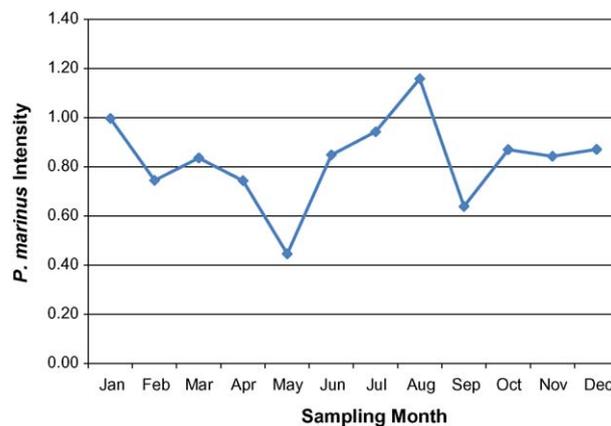


Fig. 11 – Mean intensity of Perkinsus marinus in oysters from all the sampling locations in the Caloosahatchee Estuary. 10–15 oysters were collected monthly from each of the sampling locations between August 1999 and January 2008.

4.7. Oyster disease prevalence and intensity

Disease prevalence (Fig. 10) and intensity (Fig. 11) of *P. marinus* varied significantly ($P < 0.001$) among sampling locations and sampling months. Disease prevalence and intensity increased with increasing salinity and distance downstream (results not shown). On average, disease prevalence and intensity were higher in January (when salinities tend to be higher) and August (when temperatures tend to be the highest).

5. Discussion

The RECOVER Conceptual Ecological Models identify three major stressors that affect the success of Eastern oysters and associated invertebrate and vertebrate species: altered hydrology, altered habitat (affecting habitat loss, hydrology, water

quality), and sedimentation (affecting habitat loss) (CERP MAP 2004). Land development around the watersheds of the Northern (and Southern) Estuaries represents a large loss of habitat given the watershed runoff and resulting low salinities, as well as poor water quality (e.g., contaminants, bacteria, sedimentation). This loss has had a major negative impact on oyster reefs and has thus indirectly and directly impacted macroinvertebrate and fish species.

Water management activities within these watersheds have resulted in significant alterations in the timing (excess wet season and insufficient dry season water flows), distribution (water now flows through canals instead of overland), volume, and quality of water delivered to these estuaries. Channelization and water control structures have reduced the ability of these systems to filter nutrients and have led to further degradation of water quality. These impacts reduce water storage in the watershed, dry season flows, water-quality treatment, and fish and wildlife habitat. Pre-drainage estuarine systems received freshwater inflow primarily from direct rainfall and slow basin runoff that resulted in low nutrient inputs. These natural patterns of freshwater inflow sustained an ecologically appropriate range of salinity conditions with fewer salinity extremes. Water management and dredging practices have had major impacts on the presence of oysters within these estuaries. CERP projects that will restore more natural freshwater inflows into the estuaries will provide beneficial salinity conditions, a reduction in nutrient concentrations and loads, and improved water clarity that will promote the reestablishment of healthy oyster bars and associated communities. These stressors and attributes are described in the conceptual ecological models in the RECOVER MAP.

In the present study, the Caloosahatchee Estuary was chosen as a model estuary to examine the impact of watershed alteration on oysters and to develop a Stoplight Report Card for oyster physiological and ecological response. The CE encounters high freshwater inflows due to local rainfall events and large regulatory freshwater releases from Lake Okeechobee, depressing salinities for extended periods. However, large extended regulatory releases are not a natural event and can result in harmful reductions in salinities for extended periods. A significant relationship between freshwater inflows and salinities at all the sampling sites was noted in areas where oyster reefs naturally exist. Results suggest that flows between 500 and 3500 cfs will result in salinities between 10 and 32 ppt (Fig. 6), a range that is favorable to oysters (Shumway, 1996; Volety et al., 2003). Adult oyster density, substrate availability and suitability for larval oyster spat recruitment, disease intensity and prevalence of *P. marinus*, condition of oysters, reproduction, and susceptibility to predation are all influenced by the timing, duration, and frequency of freshwater flows into the estuaries. Average oyster condition index ranged between 2.4 and 3.4 in the CE (Fig. 7), with changes coinciding with the reproductive phase of oysters. As oysters reproduce, gametes are shed resulting in a decrease in body mass and thus a reduced condition index. This trend is reinforced by seasonal patterns in oyster Gonadal Index and spat recruitment. The Gonadal Index of oysters was higher during peak spawning months (April–October; Fig. 8). Larval recruitment was observed at various sampling locations between April and October (Fig. 9). Spat recruitment per

shell ranged between 2.5 and 25, suggesting that the CE is not limited by larval availability. Juvenile oysters grow faster than adult oysters, thus enabling the determination of growth rates at various locations subjected to various salinities.

Given the variation of freshwater inflow into the CE (0–15,000 cfs), growth and survival of oysters was significantly impacted at the extreme end of the salinity range, and varied with time of deployment. Given the variability in the time of deployment, growth rates and survival of juvenile oysters due to ambient salinity, data could not be statistically analyzed for this study. Mean *P. marinus* prevalence from all the sampling locations and sampling months ranged from 31 to 66% (Fig. 10) between sampling months (when data are combined from all the sampling stations) and between 35 and 56% among sampling locations (when data are combined from all the sampling locations; results not shown) to represent the current data and trends. Similarly, *P. marinus* intensity ranged between 0.64 and 1.16 during various sampling months (scale 0–5; Fig. 11) and between 0.41 and 1.1 at various sampling locations (results not shown). These results were used to develop an easy to understand Stoplight Report Card system to characterize the current state of oysters in the Caloosahatchee Estuary, and not to examine the relationship between various water management practices and interrelationships between oyster responses and other factors that influence them.

Based on the available data of oyster responses in the Caloosahatchee Estuary, the system appears to be at stage “caution” (yellow). This level may change as modifications to the system occur (e.g., change in the quality, quantity, timing and distribution of freshwater inflows) as CERP projects are implemented. In the current study, all the metrics monitored were weighted equally in determining the Component Score; however, in other systems, various responses may be dropped or weighted more or less, as appropriate.

Suitability indices can bridge the gap between the Task Force’s System-wide Indicators (Barnes et al., 2007) and the RECOVER MAP process. When combined with spatially referenced maps of an indicator (ecological attribute in the MAP Conceptual Ecological Models), indicator performance measure suitability curves can provide a basis for restoration project evaluation or assessment. Combined with a scoring system and stoplight display (as part of a more detailed scientific report) the suitability curves form the basis for the communication tool (Table 2). Translating index scores into a stoplight display can be done for each performance measure (component) and then combined into a species score (e.g., Eastern oyster) by MAP module, and then each species individually system-wide to provide a system-wide oyster indicator score.

The Northern Estuaries CERP oyster indicator targets are based on optimization model outputs, natural variation that did occur during the period 1965–2000, and desirable salinity conditions for existing and potential aquatic resources (CERP-MAP, 2004). Targets for the CE are based on freshwater discharges from the C-43 canal at the S79 structure where the mean monthly inflow should be maintained between 450 and 2800 cfs. Targets were developed to better manage minimum and maximum flow events to the estuary to improve estuarine water quality, and to protect and enhance estuarine habitat and biota.

5.1. Low flow

The low flow target is no months from October to July when the mean monthly inflow from the Caloosahatchee watershed, as measured at S79, falls below a limit of 450 cfs (C-43 basin runoff and Lake Okeechobee regulatory releases).

5.2. High flow

The high flow target is no months with mean monthly flows greater than 2800 cfs, as measured at the S79, from Lake Okeechobee regulatory releases in combination with flows from the Caloosahatchee River (C-43) basin.

5.3. Frequency and rates of flows

The frequency distribution of monthly average freshwater inflows through S-79 for the entire period of record has been found to be important for protecting and restoring estuarine resources. Approximately 75% of the flows from S79 should be in the 450–800 cfs range and most of the remaining inflow should be in 800–2800 cfs range.

5.4. Lake Okeechobee regulatory releases

The alternative with the least daily discharge volume, the fewest number of total days of discharge, and the fewest number of consecutive days of discharge is preferred. Special considerations are provided for pulsed releases that benefit the estuary.

5.5. Optimal flows in the Caloosahatchee Estuary

Volety et al. (2003) recommended freshwater inflows for the protection and enhancement of oyster recruitment and survival around Shell Point and San Carlos Bay that are consistent with the flows outlined above and for SAV. Flows between 500 and 2000 cfs would result in salinities of 12–30 ppt at all stations, conditions that are favorable to sustain and enhance oyster populations in the Caloosahatchee Estuary. Under current water management practices, oysters in the Caloosahatchee are not stressed by low flows of <300 cfs from S79; however, complete cessation of discharges during the winter will increase salinities in areas normally associated with lower salinity and result in the immigration of marine predators and pests. Volety et al. (2003) further speculate that oyster spat in downstream areas also will be exposed to higher salinity and heavy predation pressure resulting in very little survival.

Nonetheless, the greatest threat to oysters under current water management practices is due to high flows exceeding 3000 cfs for extended periods (2–4 weeks). This is especially true for summer months during peak oyster spawning, juvenile recruitment, and growth. Volety et al. (2003) recommended that although freshwater releases may still be necessary prior to CERP, repeated pulses of <1 week duration during winter months should be made instead of sustained releases of freshwater during summer or winter months. Interpretation of these results also indicates that such pulses would be least damaging during December through February,

before increased spawning and recruitment begins at upstream locations. Salinities preferred by oysters will be maintained if the target flow frequency distribution is achieved, especially if 75% of the flows are between 450 and 800 cfs. In addition to the freshwater inflow resulting from regulatory lake releases, natural events such as tropical storms and hurricanes contribute to significant amount of rainfall and watershed runoff resulting in depressed salinities. However, the residence time of freshwater resulting from these events is relatively short (1–3 days), compared to the regulatory releases which could last for several weeks, and thus have a minimal impact on oyster responses.

The CERP goal for the Northern Estuaries is to enhance habitat conditions while providing for economic and recreational opportunities. CERP projects are expected to moderate the stressors (i.e., freshwater discharges, diminished water quality, and habitat loss) and enhance the natural attributes (i.e., oysters) of the Northern Estuaries. This will be accomplished through habitat enhancement, as well as through water storage and treatment projects. As various CERP projects are implemented, changes in the hydrology, and thus the biology of oysters will take place. Provided that sufficient data covering normal variability in rainfall and temperature events (normal, dry and wet years; hot and cool years) exist, a Stoplight Report Card system that integrates various responses that are currently being measured as part of a monitoring plan can provide a powerful way to distinguish between restoration changes and natural patterns. For example, the existing data (that can be enhanced by continuous monitoring) will provide ranges of oyster responses. Comparing oyster responses after restoration-related alterations have taken place with normal ranges (prior to restoration), one can distinguish whether restoration activities are enhancing or diminishing oyster responses.

6. Longer term science needs

The oyster monitoring protocol metrics currently include changes in oyster distribution and abundance at a variety of estuarine sites on both the east and west coasts of Florida, including the St. Lucie Estuary, Caloosahatchee Estuary, Loxahatchee River, and Lake Worth Lagoon. This long-term monitoring program for the Eastern oyster focuses on four aspects of oyster ecology: spatial and size distribution patterns of adults, distribution and frequency patterns of diseases, reproduction and recruitment, and juvenile growth and survival. The current monitoring protocol for oysters in the Northern Estuaries provides a good starting point to assess the current state of oysters, evaluating the baselines responses (pre-CERP alterations). Data will be analyzed to determine if the health and spatial extent of oysters is improving with time as CERP projects are implemented and if the predictions for the oyster Interim Goal are being met.

The responses measured are based on the rationale previously provided; however, several improvements can be made to the existing monitoring protocol. For example, water quality (i.e., temperature, salinity, and DO) is being measured at all the sampling sites during each sampling event (once a month). However, a more frequent sampling is

required to capture episodic events. In addition, predation pressure may be a significant factor in the survival of oysters, and is currently not being studied. Given that predation pressure is anticipated to be high in some locations, such information is necessary and will enhance the Habitat Suitability Index (HSI) model by strengthening the predictability of potential suitable habitat.

A Habitat Suitability Index model (HSI) is currently under development for oysters in the Northern Estuaries. This GIS-linked HSI will enable resource managers to make comparisons between different scenarios enhancing the decision-making process. Basic information about salinity changes and their impacts on oysters and associated communities is relatively well understood. However, these data are mostly from areas outside the Northern Estuaries. Oysters are adapted to local conditions, and therefore their responses to ambient environmental conditions in the Northern Estuaries may be different. Continued work is needed to standardize the measurements between estuaries and continue to capitalize on developing time-series information on oysters in various estuaries.

The sample size for each of the performance metrics is partly dictated by sample collection and processing time as well as expense. For example, analyses of reproduction are currently performed using histological techniques. Such techniques are time-consuming, expensive and require specific expertise limiting the number of samples that can be analyzed. Newer techniques such as enzyme-linked immunosorbant assay that use antibodies against egg protein of Eastern oysters are now being developed. These techniques will greatly enhance the sample processing and increase the sample size and thus the power of the analyses.

Although some factors that influence oyster responses are not being measured in the current study, their influence in the success of oyster reefs in these estuaries will be examined in the future, based on need. In addition, use of newer tools such as Habitat Suitability Index models (Barnes et al., 2007) will shed light on some of the factors influencing oyster growth and survival, which can then be included in the monitoring plan. Factors that are not currently examined but may have potential influences on oysters and those that may be examined at a later time are indicated by dashed boxes in the conceptual model. Predictions of oyster-reef development following implementation of CERP can be made by using a HSI model (Barnes et al., 2007). Although the existing sampling design and sampling frequency can adequately assess the direction and magnitude of change in the performance measures of oysters, the sampling protocol may be adjusted to better capture the spatial variation of responses. The Stoplight Report Card system, in concert with HSIs such as those developed for oysters by Cake (1983), Soniat and Brody (1988), and Barnes et al. (2007) will enable resource managers to enhance decision making by providing them with a powerful tool that is based on real scientific data rather than relying solely on informal judgments or professional opinion. These models can then provide input and direction for monitoring efforts. These approaches are also easily exportable for use in other estuaries in Florida and other Gulf states with minor modifications.

Acknowledgments

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Appendix 1

Gonadal stage	Observations
0	Neuter or resting stage with no visible signs of gametes
1	Gametogenesis has begun with no mature gametes
2	First appearance of mature gametes to approximately one-third mature gametes in follicles
3	Follicles have approximately equal proportions of mature and developing gametes
4	Gametogenesis progressing, but follicles dominated by mature gametes
5	Follicles distended and filled with ripe gametes, limited gametogenesis, ova compacted into polygonal configurations, and sperm have visible tails
6 = 5	Active emission (spawning) occurring; general reduction in sperm density or morphological rounding of ova
7 = 4	Follicles one-half depleted of mature gametes
8 = 3	Gonadal area is reduced, follicles two-thirds depleted of mature gametes
9 = 2	Only residual gametes remain, some cytolysis evident
10 = 1	Gonads completely devoid of gametes, and cytolysis is ongoing

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