

Submerged Aquatic Vegetation Exposure to Deepwater Horizon Spill

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Executive Summary

On April 20, 2010, an explosion aboard the *Deepwater Horizon* (DWH), an offshore, Macondo prospect (MC252) oil rig in the northern Gulf of Mexico (GOM), resulted in the largest oil spill in US history. Oil discharged from the uncapped well and drifted shoreward, reaching the outer barrier islands of Chandeleur and Mississippi Sounds in early May where it posed a direct threat to ecologically sensitive, shallow water coastal ecosystems. Oil, the by-products of weathered petroleum, and the dispersants used to break down the oil were transported over and through shallow water subtidal habitats, potentially impacting natural resources associated with submerged aquatic vegetation (SAV). SAV in the northern GOM's coastal and oceanic ecosystems were some of first nearshore benthic ecosystems to be exposed to the oil. The SAV community consists of six species of submerged flowering plants referred to as seagrass. Seagrasses and their epiphytic communities are one of the most important coastal ecosystems in the GOM. In addition to the production of large quantities of organic matter, seagrasses form the structural habitat that supports vertebrate and invertebrate species and is the basis for a large amount of secondary productivity, a diverse food web, important biogeochemical processes, and one of the primary indicators of good water quality.

The seagrass community was especially vulnerable to MC252 oil exposure. Seagrasses are rooted vascular plants that are physically and chemically integrated with the sediments they grow in, are fixed in place, and are unable to actively avoid contact with submerged oil transported in the water column or deposited on and in the substrate. The seagrass plants in the northern GOM are generally distributed in water depths less than two meters (m). During low tide, seagrasses in shallow water can form a three dimensional canopy that occupies the entire water column, further increasing potential exposure to both surface and submerged oil. The seagrass canopy and epiphytes growing on the leaves baffle water currents and wave turbulence, acting as a filter that traps and promotes the deposition of suspended materials within the seagrass meadow. The plant's physical structure and associated metabolism are also key components of the biogeochemical cycling of materials between the water column and the substrate. In the substrate beneath the canopy, roots and rhizomes bind and stabilize sediments, effectively retaining and concentrating inorganic particulate material, organic matter, and any other materials susceptible to deposition. These bio-engineering properties are some of the most well-known and ecologically valuable services that seagrasses provide to coastal ecosystems. In an oil spill, these attributes and processes may serve as an "Achilles Heel", enhancing the potential for direct exposure to oil within a seagrass meadow by intercepting water flow, increasing deposition, and concentrating organic and inorganic material.

In response to the spill, the National Oceanic and Atmospheric Administration (NOAA) assembled a SAV technical working group (SAV TWG) of experts to assess MC252 oil exposure and determine if SAV resources were injured. The SAV TWG work plan followed the Natural



Resource Damage Assessment (NRDA) framework established by the Oil Pollution Act of 1990 (OPA). The framework included three phases:

- Phase 1: Pre-assessment,
- Phase 2: Restoration planning (injury assessment and restoration selection), and
- Phase 3: Restoration Implementation.

This report addresses Phases 1 and 2. To accomplish their objectives The SAV TWG divided the assessment activities into three tiers:

- Tier 1: Characterization of baseline and reference conditions
- Tier 2: Assessment and characterization of initial post-spill conditions, and
- Tier 3: Injury assessment and recovery.

Several large datasets were compiled and numerous studies were completed to address the transport and fate of MC252 oil and assess exposure pathways. The presence and exposure of MC252 oil in the SAV communities of the northern GOM was quantitatively evaluated by repeating shoreline oiling observations, mapping the trajectories and calculating the spatial accumulation and thickness of oil transported on surface waters, and determining the concentration of oil-related chemicals detected in 20,000 sediment, tissue, and stranded oil samples.

In Tier 1, the SAV TWG identified areas of known seagrass habitat in the northern GOM and determined the extent of sampling required to comprehensively assess the potential threat from the MC252 oil spill. Originally, the concern was that oil would be entrained in the Gulf loop current and widely transported throughout the northern and eastern GOM. Consequently, Tier 1 baseline assessments were conducted at 19 sites ranging from Louisiana to the Florida Keys. However, instead of widely dispersing the oil, prevailing oceanographic and climatological conditions transported most of the MC252 oil northward. Accordingly, the Tier 2 assessment investigated five sites with known communities of seagrass that were threatened by potential exposure to oil:

- Big Lagoon, FL
- Robinson Island in Perdido Bay, AL
- Horn Island, MS
- Petit Bois Island, MS
- Chandeleur Islands, LA.

Of the five sites, only the seagrass beds in the Chandeleur Islands were determined to be exposed based on evidence of extensive oiling on nearby shorelines documented by shoreline oiling classifications assessments (Shoreline Cleanup and Assessment Technique, herein referred to as 'SCAT'), provisional total petroleum hydrocarbon (TPH) concentrations in sediment, and estimates of oil on surface waters using Synthetic Aperture Radar (SAR) and aerial imagery.



Tier 2 samples of sediments, seagrass tissue, and invertebrate tissue within affected seagrass beds in the Chandeleur Islands showed concentrations of total polycyclic aromatic hydrocarbons (tPAHs) orders of magnitude higher than ambient (baseline) concentrations and forensic PAH and biomarker analyses matched with the MC252 oil from the DWH spill. In fact, almost all stranded oil samples and 70% of sediment samples matched. Concentrations of sediment tPAHs were 8-12 times greater, on average, than baseline, pre-spill conditions and SAV tissue tPAH concentrations were 13 times higher than baseline. Concentrations in invertebrate tissue samples were over 400 times higher than the pre-spill baseline.

Elevated concentrations of tPAHs corresponded with shoreline SCAT data and SAR accumulation estimates of oil on surface water. Passive water samplers (PEMDs) documented chronic exposure to biologically available PAHs in the water column almost four months after the initial oiling of the Chandeleur Islands in May. SCAT and SAR data indicate that the most extensive oiling of the Chandeleur Islands occurred from mid-June through mid-July 2010. Heavy oiling of shorelines near seagrass beds were last observed by SCAT surveys in February 2011.

Tier 3 samples were collected in June 2011, thirteen months after the Chandeleur Islands were first exposed to MC252 oil. Concentrations of tPAHs in sediment and plant and animal tissue had decreased to approximately one-fifth of the levels measured in September 2010. The observation of relatively lower tPAH concentrations, a reduction in oil concentrations in external materials on seagrass, and a decline in the number of forensic matches indicated that acute surface oiling from MC252 was possibly no longer present. However, the concentrations in June 2011 continued to be higher than pre-spill, baseline levels suggesting the presence of chronic exposure for at least a year following the spill.

In Tiers 2 and 3, benthic core samples, accompanied by visual observations made both in the field and laboratory, quantitatively characterized the species composition and abundance of seagrasses, as well as assessed their health and condition. Canopy cover ranged between 70 – 100% where seagrasses were present. Species composition and abundance (shoot density) was highly variable and fell within the broad range of data reported for the northern GOM. The majority of samples collected at Chandeleur indicated the presence of just one species, but meadows of multiple species were also recorded. Common species identified in the samples were Ruppia maritima, Halodule wrightii, and Thalassia testudinum. Only a few samples had Syringodium filiforme and none had Halophila engelmannii. New leaf and shoot growth was documented and there was no visual or morphometric evidence of acute stress or seagrass mortality at the time of Tier 2 or Tier 3 sampling. Yet despite a suite of healthy diagnostic characteristics, relatively homogeneous environmental conditions, and water depths suitable for growth, the spatial distribution and areal coverage of seagrasses across the shallow shelf west of the Islands was not continuous. Seagrasses were heterogeneously distributed in various sized patches among gaps of unvegetated substrate. The heterogeneous seagrass distribution pattern was consistent with the variation in exposure documented by sediments and tissue samples,



shoreline oiling classifications, and oil on water observations. Since the Tier 2 assessments were not conducted until September 2010, three months after MC252 oil first arrived at the Chandeleur Islands, the possibility that some of the unvegetated gaps were the result of acute exposure could not be ruled out.

Given that oiling was heterogeneous and widespread across the Island, and exposure in the seagrass beds was documented both in September 2010 (Tier 2) and again in June 2011(Tier 3), the injury assessment was expanded to include a quantitative change analysis of seagrass using aerial imagery for three consecutive years: October 2010, Fall 2011, and Fall 2012. The areal coverage of seagrass in October 2010 was considered baseline for injury assessment. The change analysis identified areas of seagrass loss that could not be attributed to natural processes or interpretation error, and likely resulted from MC252 oil exposure. Three categories were used to quantify the change in areal seagrass coverage:

Category 1: Gains in seagrass Category 2: Losses of seagrass Category 3: No change in seagrass

Using an object-based image analysis method verified by standardized seagrass mapping and interpretation protocols, changes in seagrass areal coverage in five locations in the Chandeleur Islands was documented. Injury to the seagrasses of Chandeleur Islands was quantified by acres lost. Polygonal areas were designated as "persistent loss" if seagrass was absent for two consecutive mapping intervals (2011 and 2012) following exposure to oil. A "delayed loss" classification was assigned to areas that had seagrass in 2010 and 2011, but lost seagrass in 2012. A total of 111.7 acres were identified as persistent loss and 159.5 acres were classified as delayed loss, totaling 271.2 acres.

To complete the injury assessment, the science-based approach to seagrass injury and recovery developed in NOAA's Mini 312 Program was adopted. This approach recognizes that the ecological services provided by seagrasses are lost or impaired during the recovery period and the time needed to reach full recovery from an injury is contingent upon the type and size of the injury, the species composition, and the prevailing environmental conditions. Recovery calculations were limited to areas of persistent loss exceeding 100 m² (0.0247 acres). The initial analysis identified 583 individual persistent loss polygons with a total area of 51.08 acres. Of these polygonal areas, 131 (33.85 acres) were identified as having recovery times that exceeded one year. Approximately one third of these persistent loss areas (11.24 acres) have a predicted recovery time of between one and two years. Roughly 37.4% of the persistent loss areas (12.65 acres) have a recovery time of between two and ten years. The remaining 9.96 acres have recovery times ranging from 14 to 26 years.



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Submerged Aquatic Vegetation Exposure to Deepwater Horizon Spill

1 Introduction

On April 20, 2010, an explosion aboard the *Deepwater Horizon* (DWH), a mobile, offshore Macondo prospect (Mississippi Canyon 252 or MC252) oil drilling rig, caused the largest and most prolonged offshore oil spill in United States (US) history (NOAA, 2012a). Early attempts to cap and plug the well were unsuccessful until July 15th – 87 days after the initial blowout, when a temporary cap was fitted to stop the release of oil into the Gulf of Mexico (GOM) (U.S. District Court, 2014 and 2015). In response to the spill, significant quantities of dispersants were applied both at the source of the leak and to surface waters. The resulting plume of oil and dispersant traveled upward from the wellhead through the 1.5 kilometer (km) water column to form expansive surface oil slicks, which were then transported to nearshore coastal waters by wind and currents. Travelling from wellhead to shore, the oil underwent significant chemical and physical changes (Emsbo-Mattingly and Martin, 2015). In all, more than 2,000 km of the northern GOM shoreline were reported as oiled, of which more than 600 km were moderately to heavily oiled (Nixon et al., 2015). Oil reaching the shoreline was transported over and through shallow water subtidal habitats and potentially impacted natural resources associated with submerged aquatic vegetation (SAV).

SAV resources are a vital component of their aquatic ecosystems. In the GOM there are at least 26 species of SAV growing in freshwater, brackish, and saline environments. Coastal and oceanic ecosystems with relatively higher salinity were most vulnerable to oil exposure. In these ecosystems. SAV consists of a group of submerged flowering plants referred to as seagrass. Seagrasses provide a wide range of ecological services rivaling or, in some instances, exceeding the functions of tropical rain forests and coral reefs (Rasheed et al., 2006; Larkum et al., 2006, Orth et al., 2006; Barbier et al., 2011). Seagrasses and their epiphytic communities produce large quantities of organic matter that form the structural habitat and biochemical basis of a diverse food web leading to high secondary production rates of ecologically important and commercially valuable fish, shellfish, and wildlife communities (Ogden and Zieman, 1977; Borowitzka et al., 2006). Seagrass primary production also maintains good water quality by recycling and temporarily storing nutrients, filtering the water column, dissipating wave and current energy, and stabilizing sediments (Zieman, 1982; Zieman and Zieman, 1989; Larkum et al., 2006; Romero et al., 2006). Unlike most of their algal counterparts in the marine ecosystem, seagrasses also produce organic matter below ground as roots and rhizomes (Kenworthy and Thayer, 1984). The labile fraction of below ground production fuels benthic microbial processes and secondary production, while the more retractile carbon may remain buried and sequestered for extended periods of time (Gacia et al., 2002; Duarte et al., 2005).



The seagrass beds consisted primarily of six species: *Thalassia testudinum*, *Halodule wrightii*, *Syringodium filiforme*, *Ruppia maritima*, *Halophila decipiens*, and *Halophila engelmannii*. These species are widely distributed in the GOM in water depths generally < 20 m (Iverson and Bittaker, 1986; Zieman and Zieman, 1989; Dawes et al., 2004; Handley et al., 2007). It is estimated that the GOM has greater than 50% of the total US distribution of seagrasses and at least or greater than 5% of the known global occurrences (Green and Short, 2003). The Florida Keys, Florida Bay, and the adjacent Continental Shelf in the eastern GOM make up one of the largest documented semi-continuous seagrass meadows in the western hemisphere encompassing a significant component of the world's seagrass ecosystem biodiversity (Continental Shelf Associates, Inc. 1985 and 1987; Iverson and Bittaker, 1986; Zieman and Zieman, 1989; Fourqurean et al., 2001; Fourqurean et al., 2002; Short et al., 2007; Handley et al., 2007).

During the late spring and summer of 2010 oil from the DWH spill, by-products of weathered petroleum, and the chemicals used to break down the petroleum all posed a potential threat to GOM SAV resources (McRoy and Williams, 1977; Nadeau and Bergquist, 1977; Zieman et al., 1984; Thorhaug et al., 1986). Types of MC252-specific contaminants that could have occurred during the life of the spill include:

- MC252 crude oil,
- Weathered oil oil that has been exposed to the environment and is in a "degraded" state,
- Dispersed oil oil that has been treated with dispersants and now is in an altered state, and
- The dispersants themselves.

Potential direct impacts of oil and dispersants on seagrass range from complete mortality (Jackson et al., 1989; Sandulli, 1998; Thorhaug and Marcus, 1987; Scarlett et al., 2005) to sublethal stress and chronic impairment of seagrass and sediment metabolism and function (Hatcher and Larkum, 1982; Ralph and Burchett, 1998; Peirano et al., 2005). Response and cleanup efforts in seagrass beds and adjacent shorelines can cause physical degradation of the environment and seagrass loss or impairment, as seen in many Gulf areas where response and clean-up equipment were placed within seagrass beds (NOAA, 2011a). Secondary impacts can also include biophysical and chemical disturbance to sediments, microbes, microfauna, and microflora (Short et al.,1995), and the impairment and mortality of secondary producers residing in the seagrass canopy and sediments (e.g., invertebrates, crustaceans, fishes, and waterfowl) (Carls and Meador, 2009). The conceptual diagram provided as Appendix A illustrates the various lethal and sub-lethal effects of oil in the SAV ecosystem.

In response to the spill, the National Oceanic and Atmospheric Administration (NOAA) assembled a SAV technical working group (TWG) to assess the spill's potential threat to GOM SAV resources, as well as to determine whether GOM SAV resources were injured by MC252 oil exposure and its associated clean-up activities. The SAV TWG consisted of technical experts



and staff from NOAA, other Federal Agencies (Department of the Interior (DOI)) and State Agency trustees from four states: Louisiana, Mississippi, Alabama and Florida. Government contractors (Industrial Economics, Incorporated (IEc) and NewFields), and representatives of the responsible party (RP), British Petroleum (BP) and their environmental contractors and scientists were also part of the SAV TWG. The TWG followed the Natural Resource Damage Assessment (NRDA) framework established by the Oil Pollution Act of 1990 (OPA) in assessing the spill's potential effects on SAV. The framework included three phases: 1) Pre-assessment, 2) Restoration planning (injury assessment and restoration selection), and 3) Restoration Implementation. This report covers two elements of the approach: 1) Pre-assessment and 2) Injury assessment. The SAV TWG divided up the assessment activities into three tiers, 1) Characterization of baseline and reference conditions (Tier 1), 2) assessment and characterization of initial post-spill condition (Tier 2), and 3) injury assessment and recovery (Tier 3). Table 1 lists the timing of each Tier and the associated locations and activities. Work plans for each tier were written by the SAV TWG and BP representatives and signed off by Department of Commerce, DOI, the four state Trustees and BP (see NOAA 2010, 2011a, and 2011b, respectively).

2 Methods

2.1 General Description of Study Plan

The SAV TWG general study plan was executed in three tiers. Tier 1 was designed to acquire as much information as possible on baseline pre-oil conditions of SAV communities potentially threatened by oiling. The following was initially considered when selecting Tier 1 sites: 1) the known distribution of SAV beds in the GOM, 2) the proximity of the SAV beds to the location of the DWH well head (Figure 1) and, 3) the anticipated oil dispersal trajectories. The SAV TWG also evaluated published scientific peer reviewed papers and reports, and consulted with Trustee experts with knowledge on seagrass ecology and distribution in the GOM. The SAV TWG also took into account the probable transport of oil by the GOM Loop Current as far south as Dry Tortugas National Park in the Florida Keys. Initially, it was expected that some of the oil released from the DWH well would be entrained and transported in the Loop Current, suggesting a potential threat to seagrasses throughout the northern, eastern, and southeastern GOM. Collectively, this information was used to identify the spatial scope of sampling needed to comprehensively assess the potential threat. In May and June 2010 the SAV TWG identified all potentially threatened areas and initiated baseline assessments at 19 locations before MC252 oil could reach their SAV beds. The initial areas of interest extended east from Vermillion Bay, LA across the northern GOM, and as far south as the Florida Keys National Marine Sanctuary and Dry Tortugas National Park (Table 1 and Figure 1).

The climatological and oceanographic conditions expected to facilitate the transport mechanisms directing the oil south along the west coast of Florida to Florida Bay and the Florida Keys did not



materialize. Most of the MC252 oil and related weathered products were transported north and northeast (Graettinger et al., 2015). Based on preliminary evidence from a wide range of sources including SCAT data (Michel et al., 2013), observations of oil transported on surface waters (Graettinger et al., 2015)¹, and the known distribution of seagrasses (Handley et al., 2007), the SAV TWG identified nine primary locations in the northern GOM where SAV exposure to MC252 oil was deemed most probable (Table 1 and Figure 2a). In July, August, and September of 2010 the SAV TWG conducted Tier 2 post-spill exposure assessments at these locations. The assessments included the confirmation of oil exposure within SAV beds and animal communities and SAV health and condition assessments at a subset of five of the nine locations: 1) Chandeleur Islands, LA, 2) Horn Island, MS, 3) Petit Bois Island, MS, 4) Perdido Pass, AL (Robinson Island), and 5) Pensacola Bay (Big Lagoon), FL (Figure 2a). Tier 2 activities also included deploying submerged oil sentinels at eight locations (Figure 2b) (Zelo and Benson, 2011) as well as passive water samplers (low-density polyethylene membrane devices referred to as PEMDs). The PEMDs were designed to detect mobile and biologically active petroleum in the water column (Figure 2b) (Carls et al., 2011).

The Chandeleur Islands were initially considered the most impacted of the above-listed Tier 2 locations due to its proximity to the DWH wellhead and available SCAT data. Using the SCAT data, the Chandeleur Islands were stratified into locations with indication of heavy oiling (southern region), moderate oiling (middle region) and no oiling (northern region) (Figure 3a). These qualitative oiling descriptions within the Chandeleur Islands were used throughout Tier 2 planning.

During Tier 2, the SAV TWG worked closely with the aerial imagery technical working group (AITWG) to plan and acquire high resolution aerial imagery of SAV beds in the northern GOM from Galveston Bay, Texas to Alligator Point, Florida (Handley et al., 2010).

Using the provisional Total Petroleum Hydrocarbon (TPH) sediment sample results of Tier 2 and the preliminary fingerprint identification of MC252 oil (Emsbo-Mattingly and Martin, 2015) within seagrass beds, the SAV TWG determined that the Chandeleur Islands, LA had the most probable and potentially most severe exposure to MC252 oil. According to Graettinger et al. (2015), the Chandeleur Islands were the first shallow water habitats in the northern Gulf to experience exposure, estimating that the Gulf side of the Islands were oiled as early as May 4, 2010 and demonstrated between 6 to 10 days of oil on water in the middle and southern regions of the western side of the Islands where seagrasses occur (Figure 3b). SCAT data collection was not as frequent at Chandeleur Islands due to its remote location. However, oiling of the Islands was confirmed from SCAT surveys as early as May 8, 2010. Widespread light to heavy oiling throughout the Islands was identified during SCAT surveys on June 21st through the 28th and

¹ The arrival times of oil-on-water at various locations were determined by analysis of daily satellite information (Graettinger et al., 2015).



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July 11th through the 13th. Finally, visible oil on water was evident in May 10, 2010 imagery from the National Agriculture Imagery Program (NAIP), which was coincidentally collected at the Chandeleur Islands during the oiling event.

No other investigated SAV area was exposed to the same level of oiling as the Chandeleur Islands (see Figure 2a for cumulative oiling estimate near all Tier 2 sites and Section 4.1 for more information on individual oiling events). Therefore, Tier 3 sampling efforts were focused in the southern and middle regions of the Chandeleur Islands (Figure 3a). Tier 3 assessments were performed in June 2011.

The SAV TWG conducted a quantitative change analysis of seagrass areal coverage based on high resolution aerial imagery acquired by the AITWG in the fall of 2010, 2011, and 2012 (Figure 4a, b and c, respectively) as part of the Tier 3 injury assessment. The imagery analysis focused on documenting the changes in seagrass coverage in five core areas of the Chandeleur Islands following exposure to MC252 oil. For the change analysis, the areal coverage of SAV (seagrasses) was quantitatively documented for each time interval as three categories: 1) gains in SAV, 2) losses of SAV, and 3) no change in SAV. Areas were designated as "persistent loss" if seagrass was absent (no SAV) from an area (polygon) for two consecutive mapping intervals (2011 and 2012) following exposure to oil and the initial areal mapping in 2010. Fall 2010 imagery was considered baseline in the injury assessment and the persistent loss of SAV was defined as injury.

2.2 Exposure Data Sources and Data Management

Due to the spill's geographic extent, three-dimensional nature, and ecological complexity, numerous response and NRDA efforts were undertaken to characterize the effects of the spill (USCG, 2011; NOAA, 2012a). These efforts have generated a large number of compiled chemistry datasets associated with various media. The SAV TWG used the available polycyclic aromatic hydrocarbon (PAH) data collected near and within SAV areas. Data discussed herein were acquired from two web-based sources maintained by NOAA:

- Data Integration Visualization Exploration and Reporting (DIVER), which is a collection of tools and processes to standardize and make available a vast range of data associated with DWH spill (https://dwhdiver.orr.noaa.gov/); and
- Environmental Response Management Application (ERMA®), which is an online mapping tool that integrates both static and real-time data, such as Environmental Sensitivity Index (ESI) maps, ship locations, weather, and ocean currents, in a centralized format (http://response.restoration.noaa.gov/erma/).

The primary exposure data summarized in this paper are total PAH (tPAHs) concentrations, which are calculated using the toxPAH50 formula available in DIVER. This formula calculates the arithmetic sum of 54 individual and co-eluted PAH compounds, as listed in Table 2. For the



purposes of this analysis, if the concentration of a given compound in a sample is below its reported detection limit, the compound concentration is treated as a 0 value in the summation.²

2.3 Shoreline Oiling Classification

Repeated shoreline oiling observations along the northern GOM, observed by SCAT and Louisiana Rapid Assessment (RA^3) teams were used to develop temporal oiling classifications (NOAA, 2013b). Based on these observations, shoreline segments were assigned to specific oil exposure categories (Nixon et al., 2015). For vegetated shorelines, shoreline oil exposure categories included heavier persistent oiling (where heavy or moderate oiling was repeatedly observed over a period of 12 weeks or longer), heavier oiling (where moderate or heavy oiling persisted for less than 12 weeks), lighter oiling (where only trace to light oiling was observed), no oil observed, and shoreline not surveyed. Beaches were classified using a similar framework with additional categories to account for significant subsurface oiling and persistence over time.

2.4 Submerged Oil Sediment Chemistry

Over 34,000 sediment samples associated with the DWH spill have been collected throughout the GOM. The SAV TWG collected approximately 520 of these sediment samples following protocols outlined in the SAV TWG Tier 1 Pre-assessment Plan (NOAA, 2010).

Prior to PAH analyses, most SAV TWG-collected sediment samples were subjected to laboratory screening for presence of oil-related compounds by analyzing for TPH.⁴ Samples indicating likely presence of oil and nearby unoiled samples were subjected to PAH analysis. Of the 520 sediment samples collected by the SAV TWG, 154 were subjected to PAH analysis. Preliminary determination or "fingerprinting" of MC252 oil related compounds was performed by both Alpha and Columbia Analytical Services (CAS) laboratories (see Section 2.6). This report uses validated sediment data which was downloaded from DIVER on January 7, 2015.⁵

2.5 Biota Tissue Chemistry

The SAV TWG, as well as other TWGs, collected biota tissue samples for the purpose of evaluating exposure to PAHs. In addition to SAV tissue samples, SAV-associated benthic and epibenthic invertebrates, and mobile macroinvertebrates were collected from May to June as part of the Tier 1 effort (NOAA, 2010). Invertebrate samples were collected from the blades of SAV or within the beds and, in general, were a minimum of 30 g (wet weight). SAV-associated, mobile macroinvertebrates in deeper beds were sampled using a 16-foot otter trawl with ¾-inch

⁵ Forty-one validated, Tier 3 PAH sediment results were provided by EcoChem on June 1, 2015.



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² If the concentration of a given compound in a sample is not reported, the compound concentration is treated as a null value

³ RA refers to shoreline assessments often conducted by small teams of State and USCG representatives, as described in NOAA (2000, p. 2).

⁴ PAH and screening analyses were conducted in accordance to AQAP developed by NOAA (NOAA, 2012b).

mesh wings and ¼-inch liner. Trawls were towed for 2 minutes each. Invertebrates in shallower, inshore beds were collected with mesh nets until the minimum amount needed for chemical analysis was attained. Tier 2 and Tier 3 sampling efforts replicated many of the Tier 1 tasks and procedures.

The SAV TWG-collected SAV plant tissue samples were identified in the NRDA database DIVER as "Leaves and/or Stems". Fish and mobile macroinvertebrates were identified as "Whole Body". If possible, species information was recorded in the field and in the laboratory before analysis.

All plant tissue samples were sub-sampled twice in the laboratory. One sub-sample was rinsed with dichloromethane (DCM) and the resulting rinsate was dried and measured for PAHs. These results are identified in DIVER as "External materials." The other sub-sample was rinsed with deionized water and tissuemized before analysis. Invertebrate samples were not split and only rinsed with deionized water and tissuemized before analysis (Emsbo-Mattingly, 2015).

All tissue samples were collected according to protocols established in the SAV TWG work plans. This report uses only validated tissue data, which was last updated on DIVER on January 9, 2015.

2.6 Forensic Chemistry

To quantitatively confirm the exposure of the nearshore environment to MC252 oil, more than 1,300 stranded oil samples (usually in the form of tar balls or oil sheen) along with more than 5,500 soil, sediment, and tissue samples were subjected to forensic PAH and biomarker⁶ analyses (Emsbo-Mattingly and Martin, 2015). The PAHs present in fresh MC252 oil underwent significant weathering as it traveled to the nearshore environment and entered soil, sediment, and tissues. In contrast, biomarkers present in MC252 oil proved far more resilient to weathering than PAHs.

The biomarker pattern of each stranded oil sample was compared to the pattern found in fresh MC252 oil samples. Results of each biomarker fingerprint comparison were categorized as either Match A (fingerprints consistent with MC252 oil), Match B (fingerprints probably MC252 oil with some background material present), Match D or "Indeterminate" (fingerprints inconclusive), or Match E or "Non-Match" (fingerprints consistent with non-MC252 oil) (Emsbo-Mattingly and Martin, 2015).

Upon completion of analyses of stranded oil samples, biomarker patterns of each stranded oil sample categorized as Match A were used to evaluate the source of PAHs detected in their nearby soil, sediment, and tissue samples. The approach used to determine sample-match

⁶ Biomarkers refer to complex organic compounds found in crude oils, bitumens, petroleum source rock, which are derived from formerly living organisms (Wang et al., 2006)



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category for these latter samples included examination of the chromatogram for biomarker patterns, unresolved complex mixtures, naturally occurring organic matter, and petrogenic PAHs in comparison to their nearby Match A stranded oil samples. Based on the observed similarities, soil, sediment, and tissue samples were then categorized as Match A, Match B, Indeterminate, or Non-Match. Indeterminate samples were then evaluated for their proximity to other manifestations of MC252 oil, such as Match A or B stranded oil, other Match A or B sediment samples, or shorelines characterized by Trustees as oiled. Indeterminate sediment, soil or tissue samples situated within 100 m of MC252 oil manifestations were categorized as Match C. Stranded oil samples are demonstrated in Figures 5 through 11.

2.7 Tier 1 Baseline Sampling

2.7.1 Sample Site Selection

The primary objective of the Tier 1 investigation was to develop a quantitative basis in order to compare pre-versus post-oiling conditions at a SAV site. Statistical sampling design procedures coupled with geostatistical analyses of data collected within SAV beds in the GOM were used to determine sampling density. Typical statistical decision parameters are:

- Type I error or α (erroneously declaring a site not-impacted) = 5% (significance, or a confidence = 95%);
- Type II error or β (erroneously declaring a site impacted) = 10%, or a Test Power of 90% to correctly rejecting the null hypothesis; and
- Test resolution or Δ which is the percent change in mean value detected at 95% confidence.

Post-Hurricane Katrina seagrass survey data along Ship, Horn and Petit Bois Islands off the Mississippi coast were used to help determine Tier 1 sampling density for small, discontinuous SAV beds. A statistical analysis of this data determined that a 400 meter separation distance between sample stations was required to meet the above decision parameters. Therefore, a target distance of approximately 500 meters between stations was determined sufficient in detecting the desired percent change in observed SAV conditions at a site relative to those observed at a comparable reference site with 95% confidence (Type I error = 5%) and 90% statistical test power (Type II error =10%). A similar analysis was performed using seagrass density data from Ecofina River and Fenholloway River, Florida which have large, continuous SAV beds. Using the same decision parameters, the target distance between stations was increased to 1 to 2 km for areas with more continuous and extensive SAV beds.

Tier 1 samples were collected from May 3 to July 9, 2010. As shown by Figure 1, 285 Tier 1 sediment samples were collected as far west as Vermillion Bay, LA and as far east as the Florida Keys. In addition, 226 tissue and 12 water samples were collected as part of the Tier 1 effort. However, PAH laboratory analysis of Tier 1 samples was limited to areas that were eventually



impacted by oil. PAH analysis of Tier 1 tissue samples was limited to 26 SAV and invertebrate samples collected at the Chandeleur Islands, LA.

2.8 Tier 2 Post-Spill Sampling, Exposure Documentation, and SAV Health and Condition

2.8.1 Sample Site Selection

As mentioned earlier, Tier 2 assessments focused on the SAV resources in the northern GOM from the Chandeleur Islands, LA west to Pensacola Bay, FL. Some Tier 1 locations were revisited during Tier 2 sampling in order to obtain chemical and biological temporal trends, but most locations were targeted to evaluate potential exposure of SAV beds and their associated faunal communities to MC252 oil based on SCAT data and other oiling observations. A maximum 500 m sampling target distance (determined in Tier 1 planning) was also used for Tier 2 sample site selection.

2.8.2 Oil Exposure Documentation

Tier 2 sampling took place between August 2 and September 16, 2010 and 83 sediment, 156 tissue, and 8 water samples were collected following procedures outlined in the Tier 2 SAV TWG work plan (NOAA, 2011b). In almost all cases, tissue samples were collected with sediment samples. As mentioned earlier, sediment samples were screened for the presence of TPH before PAH analysis was performed. Approximately 20% of the Tier 2 sediment samples were not tested for PAHs due to both low TPH concentrations and presence within areas perceived at the time to have minimal impact from MC252 oiling. Most of these sediment samples were located along Petit Bois and Horn Islands, MS and Big Lagoon, FL and Robinson Island, AL. Almost all of the tissue samples collected as part of Tier 2 were analyzed for PAHs. In some cases, split samples were collected and analyzed by British Petroleum (BP).

The SAV Tier 2 assessments collected two additional types of samples. First, submerged oil sentinels or "pom poms" were used to more closely examine the presence of MC252 over time and at sites that were determined to be oiled based on SCAT. Sentinels were placed at varying depths within SAV beds at most of the Tier 2 sites (Figure 2b). Strips collected from the sentinels were sampled for PAHs and biomarkers. Second, the bioavailability and composition of mobile oil constituents in the water column were assessed in SAV beds at Chandeleur, Horn, and Petit Bois Islands, in Big Lagoon, and within Pensacola Bay with passive samplers. The passive samplers were low-density polyethylene plastic strips (~98 µm × 4.9 cm × 50 or 70 cm), known as polyethylene membrane sampling devices, or PEMDs (Carls et al., 2004). The locations of PEMDs are shown on Figure 2b. The details of the PEMD investigation are provided as Appendix B to this document and briefly summarized here in the Results section.

2.8.3 Characterization of SAV Health and Condition

In August and September, 2010 health and condition assessments of SAV beds in suspected exposure sites were conducted at: 1) Big Lagoon (FL), 2) Robinson Island, (AL), 3) Horn Island



(MS), 4) Petit Bois Islands (MS), and 5) Chandeleur Islands (LA) (Figures 5 through 11). At each location the sampling universe was selected using knowledge of SAV presence/absence based on prior surveys and mapping efforts (Handley et al., 2007; Fodrie and Heck, 2011) and local knowledge of the proximity of known SAV beds to oiled shorelines determined by SCAT surveys and surface oil trajectories. Final sample site locations were selected based on a nonrandom systematic approach to obtain the most comprehensive spatial coverage possible in the time allowed for sampling. The locations and total number of stations sampled at each of the five locations were; Big Lagoon, FL (n = 20), Robinson Island, AL (n = 10), Horn Island, MS (n = 14), Petit Bois, MS (n = 13), and Chandeleur Island, La (n = 27). As mentioned earlier, the Chandeleur Islands were further stratified into three strata based on the shoreline SCAT surveys: 1) heavy oiling, 2) moderate oiling, and no oiling (Figures 9, 10, and 11; also see Figure 3a). Figure 10 also depicts the locations of all Tier 1 samples which are labeled 1 through 12.

Field teams navigated in small vessels to each sampling station by loading their geographic coordinates into a Garmin GPS. Once the GPS waypoint was reached, a team of snorkelers entered the water and verified the presence or absence of SAV. If SAV was not present at the pre-determined waypoint the snorkelers conducted systematic circle searches around the waypoint until SAV was located, at which time the alternate waypoint was marked and stored in the GPS as the final sampling station.

At each sampling station the team recorded the time, water depth (nearest cm), SAV bed form (continuous vs. patchy), location of samples with respect to the SAV bed, and the general characteristics of the substrate type (sand, silt, etc.). The team also determined if there was oil visible on the water surface, the sediments, or on the SAV, and what form the oil was in (sheen, globules, tar or moose). Next, the team made a qualitative in situ visual assessment of the SAV shoot condition in the bed with regard to tissue color and structure, leaf senescence, evidence of shoot mortality, new shoot and rhizome growth, and the condition of apical meristems (Figure 12).

During the in situ assessments the team estimated the percent cover of SAV at each station using a modified Braun-Blanquet visual assessment technique (Braun-Blanquet, 1932) (Figure 13). At each station an observer haphazardly tossed three 0.25 m² quadrats into the bed and visually identified the species composition and estimated the percent cover of each species. In addition to SAV cover estimates, the team collected three plant/sediment cores in the SAV bed using 15 cm diameter polyvinyl chloride (PVC) pipe. The PVC cores were inserted 20 cm into the sediment, capped, and the entire core with plant and sediment material was extracted from the substrate and extruded into a mesh bag where it was gently shaken to remove sediment. During handling of the sediment cores the team observed whether oil was present on the SAV or in the sediment washed from the core. The plant contents of each core were then placed in a labeled plastic bag and stored in a cooler until processing. For processing the contents of each core, the team identified and separated the individual species present and recorded the number of shoots and



apical meristems of each species. During handling of the core material the team also made qualitative observations of the health and condition of leaves, whole shoots, rhizomes, roots, apical meristems, new shoot and rhizome growth, and epiphyte condition.

2.9 Tier 3 Post-Spill Sampling, Exposure Documentation, and SAV Health and Condition

2.9.1 Sample Site Selection

Using the provisional TPH sediment sample results of Tier 2, SCAT data and the preliminary fingerprint identification of MC252 oil within seagrass beds, the SAV TWG determined that the Chandeleur Islands had the most probable and potentially most severe exposure to MC252 oil. All Tier 3 samples were collected in the middle and southern regions of the Chandeleur Islands (Figures 14 and 15). During Tier 2 sample planning, these regions were categorized as the moderate and heavy oiled portions of the Chandeleur Islands, respectively, based on the SCAT classification of the adjacent shoreline. The northern portion of the Islands was categorized as "no oiling", and Tier 3 samples were not collected. However, as discussed later in the results section of this document, all portions of the Chandeleur Islands were exposed to MC252 oil as documented by surface oiling estimates from Synthetic Aperture Radar or "SAR" (Graettinger et al., 2015), shoreline oiling (Nixon et al., 2015) and positive (Match A and Match B) stranded oil results (Emsbo-Mattingly and Martin, 2015). SAR footprints are provided as Appendix C and discussed in Section 4.1.

The SAR, shoreline oiling, and forensic chemistry had not been completed before Tier 3 planning was finalized. In lieu of this information, the SAV TWG primarily used Tier 2 TPH sediment results to quantitatively identify the presence of oil related compounds, and secondarily used SCAT shoreline data to assess the degree of oiling at the following SAV sites: 1) Big Lagoon (FL), 2) Robinson Island, (AL), 3) Horn Island (MS), 4) Petit Bois Islands (MS), and 5) Chandeleur Islands (LA). The chemistry results showed that elevated concentrations of TPH (greater than 20 parts per million) were only present in the middle and southern regions of the Chandeleur Islands. SCAT data also showed relatively high levels of oiling near seagrass beds in these regions. In comparison, 26 of the 28 Tier 2 sediment samples collected within seagrass beds at Petit Bois and Horn Islands did not detect TPH. The only other detection of TPH among all other SAV TWG sediment samples was in Big Lagoon. Therefore, the SAV TWG limited the Tier 3 investigation to the middle and southern regions of the Chandeleur Islands and did not sample the four other Tier 2 sites

2.9.2 Oil Exposure Documentation

Tier 3 sampling of the middle and southern regions of the Chandeleur Islands took place between June 20 and June 22, 2011. 77 sediment and 53 tissue samples were collected following procedures outlined in the Tier 2 work plan (NOAA, 2011c). Most Tier 2 sample locations within these regions were resampled as part of Tier 3, and additional targeted samples within



SAV beds were also collected. Tier 1 sampling locations in the middle region of the Chandeleur Islands were also resampled as part of Tier 3. Preliminary evaluation of aerial imagery acquired in the fall of 2010 (see section below) was used to identify SAV beds and assist in Tier 3 sample planning and site selection beyond resampling Tier 1 and Tier 2 locations. These additional samples were selected to increase sampling coverage within the SAV beds (Figures 14 and 15).

2.9.3 Characterization of SAV Health and Condition

Field assessments and sample processing in Tier 3 followed the same protocols as Tier 2 with the exception that: 1) sampling efforts were focused on the moderately and heavily oiled strata of the Chandeleur Islands and, 2) the sampling intensity was increased (Figures 14 and 15). Instead of three visual assessments and three plant/sediment cores at each station, five visual assessments of SAV cover and five sediment cores were taken. Additionally, inshore, mid shore, and offshore stations were sampled to gain more comprehensive coverage of the SAV beds located on the shallow shelf along the western margin of the Chandeleur Islands.

2.10 Documentation of SAV Injury Using Aerial Imagery and SAV Change Analysis

2.10.1 General Plan and Objectives

The objective of the Tier 3 assessment was to supplement the point sampling by "scaling up" the injury assessment using high resolution aerial imagery to map and quantitatively evaluate changes in the areal abundance of SAV over the entire extent of the Chandeleur Islands where MC252 oil exposure was documented. Two factors prompted the expansion of the assessment's scale. First of all, MC252 oil did not reach the Chandeleur Islands in one single event, nor was it homogeneously distributed when it arrived (Graettinger et al., 2015). NAIP imagery from May 10, 2010 shows the heterogeneous distribution of oil on water as it first began to come ashore in the seagrass beds on the shallow shelf west of the Islands in May 2010 (Figure 16). As indicated by variability in the levels of oiling on the western shoreline, variation in the spatial distribution of oiling in the Chandeleur Islands continued for several weeks, arriving at different times, under different wind conditions, and at different tidal levels. These conditions likely resulted in a substantial amount of spatial variation and different degrees of exposure to the SAV on the shallow shelf west of the Islands. Oil arriving from the south and west, at periods of high tide and westerly winds, would have been readily transported further across the shelf toward the western shoreline of the Islands. Alternatively, oil arriving at falling tides would have been readily intercepted by the SAV canopies on the offshore shoals and in the shallow lagoons before it reached shore, if in fact it ever stranded on the shoreline. Oil arriving from the east and through the tidal passes between the Islands would have exposed narrow segments of the inner shelf as it was vectored west through the passes by high tides and onto the shallow shelf behind the Islands. Despite the ability to detect SAV exposure by point sampling, it is recognized that the point samples were limited with respect to inferences about SAV response under the wide range of conditions and spatial and temporal variability of the different exposure scenarios. The



high resolution and larger spatial coverage of the imagery provided an effective means of acquiring quantitative information about SAV response to chronic exposure, as well as any changes in SAV coverage in the large gaps between the individual sample points.

To quantitatively assess exposure at a broader spatial scale, sophisticated imagery analysis techniques were utilized to map the presence of SAV on three dates of high-resolution imagery from the fall of 2010, 2011, and 2012, near the end of the seagrass growing season when the meadows are normally at their maximum extent. There was no reliable source of pre-spill imagery so Fall (October) 2010 imagery was considered to represent as close to baseline pre-spill conditions as possible. Since the imagery was acquired nearly 5 months after initial oiling the possibility that there was acute stress to SAV cannot be ruled out, and the baseline underestimated the spatial coverage at the time of oiling in May and June, 2010. Change detection was then performed to determine the location and extent of SAV change in the two years following exposure.

The project area for the 2010 and 2011 mapping consisted of the shallow waters on the west side of the Chandeleur Islands, extending along the full length of the island chain where most of the SAV was located. New Harbor and North Islands, which are located further west, off the southern end of the Chandeleur Islands, were excluded from the mapping effort because field data were not collected around these islands during the SAV TWG sampling activities. No mapping was performed on the eastern (Gulf side) side of the islands where wave energy is generally too strong for SAV to establish and thrive. For the 2012 mapping, the project area was modified to focus on areas with the highest potential for oil exposure. Five Areas of Concern were identified within the Chandeleur Islands based on oil observed in imagery in combination with field data indicating heavy or moderate exposure to MC252 oil. NAIP imagery (1 meter resolution) acquired May 10, 2010, showed oil plumes in the water off the center and south end of the Chandeleur Islands (Figures 3a and 16). The shoreline SCAT data corresponding with these locations is shown in Figure 17 along with the five Areas of Concern that were used for seagrass mapping.

Aerial imagery for SAV mapping was acquired on three dates: in the fall of 2010, 2011, and 2012 (Figure 18). The five Areas of Concern that were delineated are shown in red over the 2012 imagery. The imagery was acquired through a *Technical Specifications and Scope of Work/Services for Aerial Imagery Acquisition and Image Processing in Support of the MC252 NRDA Process* (Handley et al., 2010) and *Mississippi Canyon 252 Assessment Workplan Concerning Aerial Imagery in the Northern Gulf of Mexico* (NOAA, 2013a) developed as a cooperative effort by the Aerial Imagery Technical Working Group (AITWG). The four band (Red, Green, Blue, NIR) imagery was acquired and ortho-rectified by Aerometric to meet the technical specifications of the aerial imagery acquisition. The 12-bit imagery, with a pixel resolution of 0.3 meters, was collected using multiple DMS sensors. ASPRS Class 2, 1:24,000 scale accuracy standards, which limit the RMS error in X or Y to 4 feet, were met. Review of



the imagery by the AITWG indicated that mismatches between adjacent images averaged less than 3 pixels and did not exceed 6 pixels. Overall, the quality of the imagery was very good, maintaining low sun angle, low wave height, and minimal sun glint in overlap. The frames selected for the mapping are summarized in Table 3 and shown in Figure 18. The overlapping frames were selected to avoid patches of glint as much as possible.

Complimentary information on SAV distribution, abundance, and species composition were obtained during field sampling at the Chandeleur Islands on June 2, 2010 in an attempt to collect data prior to oil exposure from the DWH spill (Fodrie and Heck, 2011). Field samples were collected again at the Chandeleur Islands in the late summer of 2010, between August 31 and September 2. Using the existing U.S. Geological Survey (USGS) seagrass maps from 1992 as the base, Tier 2 SAV sampling was done throughout the shallow waters of the western side of the Chandeleur Islands. SAV TWG acquired field samples at the Chandeleur Islands for Tier 3 between June 20 and 22, 2011. This survey collected sediment and tissue samples, as well as ground-truth information used in the seagrass classification process.

2.10.2 SAV Mapping

The purpose of the mapping was to delineate the aerial extent of SAV along the length of the Chandeleur Islands using advanced imagery analysis techniques. This SAV Imagery Analysis/Interpretation activity was considered a "hybrid" approach between automated Object-Based Imagery Analysis (OBIA) (Benz et al., 2004) and the traditional photo interpretation methods of all previous GOM seagrass mapping projects (Handley et al., 2007). The hybrid OBIA/interpretation activity was agreed upon collaboratively by the AITWG. Products resulting from the OBIA effort were reviewed by a USGS photo interpretation team with any uncertain SAV determinations noted and potentially corrected. It was anticipated that any systematic issues present in SAV classification would be worked through due to ongoing collaboration with the USGS lead (Larry Handley) throughout the SAV classification process. Any further attribution by USGS, such as species level determinations (if deemed necessary and possible), would be performed by a USGS photo interpretation team. Initially, the developed protocol required a minimum target mapping unit of 0.01 of an acre, far greater than most mapping efforts previously conducted; however, as the hybrid classification progressed the minimum target mapping unit was reduced in size to 0.001 acre (43 square feet) as the ability to discriminate seagrass from background sediment over the greatest part of the area was possible.

OBIA image segmentation was used to generate image objects which are polygons consisting of areas of similar spectral and textural characteristics as in the imagery (Benz et al., 2004). Image segmentation was done using Trimble's eCognition Developer software. The aerial imagery and principal components images derived from the aerial imagery were input into eCognition. The input images were then segmented based on parameters of scale and homogeneity specified by the analyst. The homogeneity parameters were based on color properties and shape properties of smoothness and compactness. The blue and green input bands were weighted heavier during the



segmentation process, because these shorter wavelengths penetrate the water better than longer wavelengths.

Nested image objects were produced at several scales as shown in Figure 19. Objects from the coarser scales of segmentation (i.e., larger objects) were used to classify and mask out the land areas, including the marshes. Water bodies in the interior of the Islands and the ocean waters on the Gulf side of the Islands, where little or no SAV is present, were also masked out. The remaining area, consisting of the ocean and tidal flats on the interior side of the Islands, was then segmented again in eCognition at a finer scale for mapping the SAV. The process of selecting appropriate scale and homogeneity parameters was done by the remote sensing analyst in collaboration with the AITWG chair (Larry Handley) from the USGS who has 30 years of experience photo interpreting and mapping SAV. This collaboration ensured that the final objects were of a scale and at a level of detail appropriate to discriminate discrete units of SAV for mapping.

Initially intent on mapping the presence of all seagrass in the meadows at the Chandeleur Islands, it became apparent that the hybrid mapping effort had high accuracy to delineate seagrass beds/patches with a density over an estimated 30% cover of seagrass shoots and leaves that present a distinct edge to the bed/patch. However, at less than approximately 30% seagrass cover, the photo interpretation signature shows slight discoloration of the sediment/sand without the development of a distinct edge. Therefore, the delineation of sparse (less than 30%) seagrass areas had limitations in the object based classification process. As a result, it was determined that the products of the OBIA process for seagrass mapping at the Chandeleur Islands present only those seagrass beds/patches with distinct edges, which is referred to as "discrete units of seagrass."

Once the final set of image objects was generated, various attribute values were calculated for each object. These attribute values were based on spectral and textural properties of the imagery within each object, as well as geometric properties of the objects themselves. A total of fifty-two attributes were derived for each image object. The image objects, along with their calculated attributes, were exported from eCognition as shapefiles which were then imported into an ArcGIS geodatabase.

The fine scale image objects were classified using Classification and Regression Tree (CART) statistical analysis (Breiman et al., 1984). The attributes from the objects generated for each image frame were used as input to the CART. Each image frame was processed separately because of spectral differences caused by sun glint and other environmental factors.



The classification scheme was binary, consisting of two classes: SAV and NOT SAV. Training samples⁷ of each class were identified throughout each image frame. The analysts worked collaboratively with the USGS team member to identify numerous training samples representing the range of conditions observed in the imagery, including variations in illumination, water depth, substrate, turbidity, and surface disturbance. Although the classification scheme was binary (presence/absence of SAV), samples of many other features were identified in order to be discriminated and removed from consideration as SAV, in particular the NOT SAV training samples, which consisted primarily of sand substrate sites. However, other non-SAV features were included. The training samples were input to the CART analysis and used to develop a classification model based on Random Forest decision tree logic (Breiman, 2001). The model was then applied to all objects in the image frame. Each object was assigned a class label and a probability value from the CART analysis. The probability values were based on the number of votes a class received from all the decision trees that went into the Random Forest model development. This initial classification was evaluated and a probability threshold was determined above which objects were classified with high confidence. All objects with probability values below this threshold were masked out and a second round of CART analysis was run using new sample sites selected from the objects being reclassified. After the second round of CART, the probability values from round 2 were compared with the probability values from round 1 on an object-by-object basis. The final class label was determined from the round with the higher probability value.

The results of the CART classification were inspected and edited using traditional photo interpretation methods to refine the classification. The classified image objects were overlaid on the imagery and photo interpretation was performed to identify image objects (polygons) representing errors of omission and commission. Corrections were made by editing the class label for these polygons. Only the class labels were edited. Object geometry was not altered from the initial object shapes derived from image segmentation.

The review and edit was an iterative and in-depth process, which involved both the mapping staff and outside review by USGS experts. The NewFields remote sensing staff made several rounds of reviews and edits. Each round of edits was subsequently quality checked by the USGS expert (Larry Handley) and NOAA expert (Jud Kenworthy) in SAV mapping of the Chandeleur Islands. Once the USGS expert signed off on the draft maps, they were sent to the National Wetlands Research Center where they were reviewed by a USGS photo interpreter who noted any additional edits to be made. This review was also quality checked by the USGS expert. The

⁷ Training samples are point locations identified on the imagery as having SAV present or Not Present. Several hundred locations were chosen throughout the area on the west side of the Chandeleur Islands to ensure that all parameters that would influence the spectral identification of SAV in the object-based classification were accounted for in the change detection analysis.



edits suggested by the USGS were made by NewFields staff and a final review and quality check was made by the USGS expert before the maps were finalized.

After the review process was completed, the SAV maps representing all of the image frames from each year were joined into a single map and a minimum target mapping unit was enforced. To do this, all adjoining polygons of the same class were merged. Next, any polygons less than the minimum target mapping unit of 0.001 acre (four square meters) were eliminated by dissolving them into the surrounding polygon, resulting in a standard minimum mapping unit.

2.10.3 Change Detection

The change detection analysis consisted of two parts: 1) change between 2010 and 2011 along the full length of the Chandeleur Islands, and 2) change between all three dates of mapping (2010, 2011, and 2012) within the five Areas of Concern.

For the initial change detection along the full length of the Chandeleur Islands, the minimum mapping unit SAV maps for 2010 and 2011 were overlaid and the polygons intersected. The output was clipped to the maximum common area of the two maps. Change in SAV between 2010 and 2011 was then calculated for each polygon and the acres of gains, losses, and no change were summarized.

For the second stage of change detection, the 2012 SAV map, consisting of the five Areas of Concern, was overlaid and intersected with the 2010/2011 data and clipped to the boundaries of the five Areas of Concern. Changes in SAV (acres of gains, losses, and no change) between the three dates were then determined by querying the dataset.

The objective of the change detection analysis was to identify areas of SAV loss resulting from oil exposure. Thus it was necessary to discriminate between natural losses and those potentially related to oil. To refine the analysis, Core Areas were identified and delineated within each of the Areas of Concern by the AI TWG chair (Larry Handley) that has mapped and studied the Chandeleur Islands for many years and is familiar with the natural processes of the Islands. These Core Areas represent areas where imagery interpretation cannot attribute the SAV losses to natural processes between 2010 and 2012.

Areas excluded from the Core Areas included outer edges of SAV meadows where natural erosion or burial may have occurred, areas adjacent to channels where scouring and sedimentation may have caused losses, areas where water column turbidity obscured the bottom, and overwash fans where significant sediment deposits were made by storms. In particular, large overwash fans created by Hurricane Isaac, which occurred a few weeks before the 2012 imagery was collected, were excluded from the Core Areas. By eliminating areas of potential natural losses from consideration, the Core Areas focused the analysis on areas where losses were more likely due to effects of oil exposure as documented by the 2010 May NAIP imagery, surface



oiling trajectories, SCAT surveys, and the SAV TWG sediment, stranded oil, plant, and animal tissue data.

The delineation of the Core Areas was an iterative process. Initially, they were determined solely based on interpretation of the 2010 and 2011 imagery. When 2012 post-Hurricane Isaac imagery became available, the Core Areas were refined to exclude overwash fan deposits from the hurricane. The final versions of the Core Areas were delineated after obtaining pre-Hurricane Isaac imagery from August 26, 2012 to compare to the September 2012 imagery. This imagery provided additional detail to further discriminate areas of loss due to the hurricane from those potentially due to oil exposure. After the Core Areas were overlaid and intersected with the combined 2010, 2011, and 2012 dataset, the change in SAV within the Core Areas between the three years was calculated and summarized.

2.10.4 SAV Injury Recovery Calculations

A scientific understanding of the seagrass injury and recovery process developed over several decades of research and the application of this research in NOAA's Mini 312 program was relied on to complete the injury assessment (www.darrp.noaa.gov/partner/mini312/index.html; also see Zieman, 1982; Williams, 1990; Fonseca et al., 2004; Kenworthy et al., 2002; Kirsch et al., 2005; Hammerstrom et al., 2006; Uhrin et al., 2011). Based on these studies, it is understood that seagrass injury recovery does not occur instantaneously and is a time dependent process. The extent of time needed to recover is contingent upon the type and size of the injury, the species composition, and the prevailing environmental conditions during recovery. During the time to recovery many of the ecological services (e.g.; primary productivity, nutrient cycling, and sediment stabilization) may continue to be absent or severely impaired. Therefore, to quantify the complete scope of the injury it is necessary to estimate the resource's time to full recovery (Fonseca et al., 2000).

Areas that were designated as persistent loss in the SAV change analysis were quantitatively documented (see previous section). Persistent loss areas where seagrasses were absent for two consecutive mapping intervals after 2010 oil exposure were designated as injured areas. To initiate the recovery analyses, firstly the size (perimeter and area) of each discrete persistent loss polygon identified in the SAV change analysis using ARC GIS was characterized (see previous section 2.10.3). 80,042 individual persistent loss polygons were identified in the Chandeleur Islands change analysis. Next, these polygons were sorted by area and their size distribution was examined. Based on previous experience with recovery dynamics of naturally disturbed and injured seagrass meadows of similar species composition, all of the persistent loss polygons that were < 100 m² in size excluded and a new distribution of individual polygon areas was created. An initial threshold of 100 m² was used, based on NOAA's experience with seagrass damage assessment and restoration in the Florida Keys National Marine Sanctuary (FKNMS) under the Mini 312 Program (www.darrp.noaa.gov/partner/mini312/index.html). In the Mini 312 Program NOAA conducted empirical studies and extensive assessments of injury and recovery in seagrass



beds in the FKNMS (Kenworthy et al., 2002; Whitfield et al., 2002; Whitfield et al., 2004; Fonseca et al., 2004; Kirsch et al., 2005; Hammerstrom et al., 2007). These scientifically peer-reviewed studies and the methods developed in them were successfully applied in seagrass damage settlements and trial litigation, and therefore served as both a scientific and legal basis for conducting this analysis. These studies determined that recovery times were relatively short for smaller injuries and compensation could not justify administrative costs, therefore most of the vessel injuries litigated in the Mini 312 program were $\geq 100 \text{ m}^2$, and the smaller injuries were resolved with set fees for the area damaged and no specific compensation was required for restoration.

For the next step in the analysis, the spatially explicit seagrass injury recovery modelling approach used by NOAA in the Mini 312 Program was adopted (Fonseca et al., 2004). This approach is ideally suited for this assessment because the spatial analytical method used to determine the persistent loss of seagrasses following MC252 oil exposure identified discrete unvegetated polygons (persistent loss polygons). These polygons have explicit areas and geometric characteristics that were similar to vessel grounding injuries assessed in seagrass beds in the Mini 312 program. To calculate recovery times, the model utilizes the spatial attributes of each individual polygon (total area, perimeter to area ratio) and assumes that all of the seagrass recovery in an injury (gap closure) occurs through lateral extension of seagrass shoots and rhizomes from the adjacent seagrass meadow. Known more generally as asexual reproduction and clonal growth, this process of rhizome extension and shoot production is one of the main forms of seagrass reproduction, expansion, and occupation of space (Tomlinson, 1974; Marba and Duarte, 1998; Fonseca et al., 2004). In the case of the Chandeleur Islands assessment, the injuries (individual persistent loss polygons) are unvegetated gaps in the seagrass cover with a shape (perimeter to area ratio ≈ 0.8) that are expected to recover primarily by vegetative reproduction and expansion of adjacent seagrass into the gaps.

3 Results

3.1 Tier 1 Baseline Sampling Results

3.1.1 Exposure Documentation

Tables 4 and 5 show average tPAH concentrations for sediment and tissue from all SAV TWG sampling events at Chandeleur Islands as well as average concentrations of petrogenic and pyrogenic PAHs. Tier 1 sediment and tissue results for the Chandeleur Islands, collected on June 2, 2010, demonstrated low concentrations of tPAH even though MC252 oiling had already reached some portions of the Islands by May 4, 2010 (Graettinger et al., 2015). The extent of Tier 1 sampling was limited to the moderately oiled regions of the Islands (Figure 3a). Concentrations of tPAH in sediment ranged from 7 to 105 ppb and averaged 40.8 ppb. Forensic chemistry did not identify any MC252 matches in these samples. Oiling of the Islands continued



after Tier 1 (Graettinger et al., 2015), especially in the middle and southern regions where oil on water was estimated to be between 6 to 10 days (Figure 3b).

SAV tissue tPAH concentrations from Tier 1 ranged from 0.6 to 7.4 ppb and averaged 3.4 ppb. External material rinsed from the SAV demonstrated an average concentration of 67,192 ppb and a maximum concentration of 535,598 ppb. Invertebrate tissue samples, which were mostly shrimp, averaged 5.6 ppb with a maximum value of 44.7 ppb.

3.2 Tier 2 Post-Spill Sampling, Exposure Documentation, and SAV Health and Condition Results

3.2.1 Exposure Documentation

Tier 2 samples were collected on August 31 through September 2, 2010, almost four months after the Chandeleur Islands were initially oiled. Sediment and tissue samples collected by the SAV TWG demonstrated a dramatic increase in concentrations compared to Tier 1 (Tables 4 and 5). Sediment tPAH concentrations ranged from 3.3 to 3,998 ppb and averaged 325 ppb which was eight times greater than Tier 1. Tier 2 sediment samples collected in the same, middle region as Tier 1 samples averaged 488 ppb for tPAH, which is 12 times that of the Tier 1 sediment samples. The increase in tPAH was mostly due to petrogenic PAHs which were almost 11 times higher than pyrogenic PAHs. Conversely, the ratio of petrogenic to pyrogenic PAHs in Tier 1 samples was less than two. Seventy percent of Tier 2 sediment samples were forensically matched (A or B) to MC252 oil compared to no MC252 oil matches for Tier 1 sediment samples.

SAV tissue tPAH concentrations ranged from 4.1 to 264 ppb and averaged 44.2 ppb which was 13 times higher than Tier 1 (Table 5). External material rinsed from the SAV demonstrated an average concentration of 174,020 ppb which was 2.6 times higher than Tier 1. Average SAV tissue and external material samples within the middle region of the Islands were 51.6 and 232,017 ppb, respectively, which was 15 and 3.5 times higher than that of Tier 1. Invertebrate tissue samples averaged 2,291 ppb, an increase of over 400 times compared to Tier 1. If invertebrate samples were limited to shrimp, Tier 2 tPAH concentrations were even higher, averaging 3,718 ppb, or over 660 times Tier 1 samples.

The dramatic increases in sediment and tissue tPAH concentrations and modest increase in external material indicate an accumulation of MC252 oil within SAV tissues as well as in sediment within SAV beds.

Nineteen of the 23 stranded oil samples collected along all regions of the Chandeleur Islands and within SAV beds were designated as Match A for MC252 oil. Only two stranded oil samples did not match MC252 oil. It is clear from the qualitative SAR, SCAT, and shoreline exposure data and quantitative analytical and forensic chemistry data that the Chandeleur Islands SAV beds and invertebrate populations were exposed to MC252 oil.



3.2.2 PEMD Results

The following summary of PEMD results was taken from Carls et al., 2011 which is included as Appendix B of this report.

PEMD results indicated PAHs were biologically available in SAV beds at low levels in August and September 2010, four months after initial oiling. All samples contained PAHs common in oil at concentrations above method detection limits. Distributions and gradients of naphthalenes through phenanthrenes (and possibly chrysenes) were likely caused by differential exposure to weathered DWH oil. Other PAH sources were present and variable across the sample area and contributed to total PAH concentrations; fluoranthene-pyrenes from an unknown, but possibly urban source were prominent near Pensacola and declined to the west. Conversely, perylene concentrations (probably produced in anoxic sediments) were relatively high in the Chandeleur Islands, but were below method detection limits further east.

Total PAH⁸ concentrations in passive samplers were low, ranging from 56 to751 ng/g (without perylene). These low environmental concentrations are not surprising given that the source oil was weathered and probably originated from oil stranded on nearby beaches. Although detected PAH concentrations were low, they do indicate chronic exposure through at least September 2010.

North-south and east-west total PAH gradients were observed in beds along the barrier islands. Total PAH concentrations were greatest in the southern portion of the Chandeleur Islands (mean = 373 ng/g) and generally declined towards the north (186 ng/g central and 174 ng/g north). Total PAH concentrations at Horn and Petit Bois Islands averaged 290 and 140 ng/g, respectively. The Pensacola Bay area also had relatively high PAH concentrations, but included an additional source of contamination not related to DWH. Concentrations in Big Lagoon, adjacent to Pensacola Bay, were elevated (mean 492 ng/g), were about the same at the mouth of Pensacola Bay, and dropped off inside the bay.

Total PAH concentrations in passive samplers tended to be greater inshore than offshore along the Chandeleur Islands. Concentrations in central and northern inshore samples were typically greater than in paired offshore samples, consistent with an oiling event and subsequent leaching of PAHs from stranded oil. However, offshore concentrations were about the same as inshore concentrations in the southern area, suggesting more extensive substrate oiling.

Based on the relative composition of several homologous PAH families (naphthalenes, fluorenes, dibenzothiophenes, phenanthrenes, and possibly chrysenes), the chemical evidence indicated that the PAH composition in the passive samplers was consistent with Macondo oil; percentages (without perylene) were reasonably similar to weathered Macondo oil, though the presence of

 $^{^{8}}$ The PAHs measured in Carls et al. (2011) differed from those listed in Table 2. Total PAH calculations are described on page 4 of Appendix B.



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other PAH sources influenced this comparison, particularly in Pensacola. Dissolution of PAHs and subsequent reuptake by passive samplers also likely influenced the observed composition, yet the overall patterns were quite similar. However, the pattern within chrysenes may have been altered by dissolution. All indications are that the Macondo oil that contaminated the MS and LA barrier island areas was weathered, consistent with the weathered pattern observed in passive samplers. The similarity of PAH composition was statistically supported with Principal Component Analysis (PCA) when data were restricted to likely oil components. Other unknown hydrocarbon sources added PAHs to the composition observed in SAV beds but this does not change the conclusion that Macondo oil was present. There were at least two other sources, a biogenic source that was present primarily in the Chandeleur Islands and a pyrogenic source that may represent urban soot, most prevalent in Pensacola. However, assignment of Macondo oil as the primary source oil is complicated because of composition differences among matrices (PAHs moved from oil to water to passive samplers), differential weathering, and by contributions from other PAH sources, hence the final determination of the source oil will depend on collateral verification from other samples such as sediment and vegetation collected at about the same places and times.

3.2.3 SAV Health and Condition Characterization Results

No visible oil was observed on the seagrasses or on the sediments within the seagrass beds at the five locations sampled in Tier 2. However, oil was observed as tar balls along portions of the shorelines of Horn and Petit Bois Islands and adjacent to SAV sampling stations. Likewise, oil was evident on adjacent shorelines (marsh and beach sites) near sampling stations in the moderately and heavily oiled sites in the Chandeleur Islands where it was observed as buried sheens in the intertidal sediments, tar balls and patties on surface sediments, and directly attached to plants (salt marsh spp. and mangrove spp.) in salt marsh fringe habitats.

As expected from previous seagrass surveys and documentation in the northern GOM, seagrass community composition displayed the expected biodiversity of species, depth distribution, and overall abundance at the five Tier 2 locations (Table 6). Inspection of seagrass shoot and rhizome condition along the outer edges of seagrass patches and continuous meadows indicated that seagrasses were actively growing prior to sampling and observations. New growth was evident as healthy rhizome apical meristems. Newly produced shoots of all seagrass species were observed along the expanding edges of seagrass patches, and there was no direct evidence of acute seagrass mortality within the established patches at any of the sites.

Relative seagrass distribution and abundance varied across the sampling sites according to the prevailing environmental conditions. Seagrasses were most abundant and continuously distributed in the protected waters of Big Lagoon, FL where T. testudinum was the dominant species by cover. The two species observed in Big Lagoon, T. testudinum at 18 stations and T. wrightii at 19 stations, co-occurred at 16 stations (Table 6). The average percent cover of T. testudinum was 78.6% ($\pm 5.1\%$) compared to a value of 48.9% ($\pm 6.6\%$) for T. wrightii.



Thalassia testudinum shoot density ranged from 398 to 3,400 shoots m⁻² compared to a range of 114 to 16,500 shoots m⁻² for *H. wrightii*. At Robinson Island near the mouth of Perdido Bay, *H. wrightii* was the only seagrass species observed, occurring as discrete, small patches, relatively large patches, and continuous alongshore meadows (Table 6). *Halodule wrightii* cover was 100%, with densities ranging from 6,800 to 16,000 shoots m⁻². On the northern shorelines of Horn and Petit Bois Islands, where exposure to a northerly wind fetch generates migrating underwater sand waves, the seagrasses were patchily distributed parallel to the shoreline among the sand waves. This seagrass community was comprised of much lesser amounts of *S. filiforme*. Here, the seagrass community was dominated by *H. wrightii* (Table 6) with cover ranging from 55 % to 69 %, while shoot density ranged from 833 to 6,100 shoots m⁻² with an average of 3,110 shoots m⁻². *Halodule* wrightii occurred at 25 of the 27 stations, S. *filiforme* occurred at only four stations, and there was no *T. testudinum* present at either of the Island sites.

All five species of seagrass known to occur in the northern GOM were observed in the Chandeleur Islands, but only three were actually sampled in the cores and quadrats (Table 6). Halodule wrightii was the most commonly encountered species in all three of the exposure strata, followed by R. maritima. Thalassia testudinum was observed at just two stations in the moderately oiled stratum. Although nearly all of the stations at the planned heavy and moderately oiled strata were sampled, only one of the pre-planned samples in the no oiled (northern region of the Chandeleur Islands) stratum was acquired. Activities associated with the construction of the temporary sand berm in the northern region of the Chandeleur Islands either buried the seagrasses at the original stations which were presented as Figure 3 of the Tier 2 Sampling Plan (NOAA, 2011b), or created conditions unsuitable for the growth and survival of seagrasses. Additionally, several of the most northerly stations were located in water depths and currents that exceeded the threshold for seagrass growth in the northern GOM. In order to supplement the original sampling plan in the no oiled stratum, three vegetated stations were arbitrarily located on the shallow shoal west of the original stations behind the newly constructed sand berm. Consequently, seagrass data was only acquired at four stations in the no oiled stratum.

Of the 21 stations that were sampled throughout the Chandeleur Islands, 17 were monotypic and four were mixed seagrass species stations with the co-occurrence of *H. wrightii* and *R. maritima*. Mixed species stations were observed in both the moderate and heavy oiled stratum. Seagrass cover (all species combined) was 58% (\pm 21%) in the heavily oiled stratum, 81% (\pm 14%) in the moderately oiled stratum, and 78% (\pm 9%) in the no oiled stratum. *Halodule wrightii* shoot densities ranged from \approx 2,000 shoots m⁻² in the heavily oiled stratum to \approx 9,100 shoots m⁻² in the moderately oiled stratum to \approx 3,000 shoots m⁻² in the moderately oiled stratum. No *R. maritima* was observed in the no oiled stratum. *Thalassia testudimum* densities were 738 and 909 shoots m⁻² at two stations in the moderately oiled stratum.



3.3 Tier 3 Post-Spill Sampling, Exposure Documentation, and SAV Health and Condition Results

3.3.1 Exposure Documentation

Based on the Tier 2 sampling and analysis results, further documentation of exposure in Tier 3 was focused only on the Chandeleur Islands. Tier 3 sediment and tissue samples were collected between June 20 and June 22, 2011, thirteen months after the Chandeleur Islands were first exposed to MC252 oil (Tables 4 and 5). Tier 3 samples were only collected in the middle (moderately oiled) and southern (heavily oiled) regions of the Chandeleur Islands. Sediment tPAH concentrations ranged from 0.6 to 425 ppb and averaged 69.6 ppb, which was 1.7 times greater than that of Tier 1 but only 21% that of Tier 2.

SAV tissue tPAH concentrations ranged from 0.4 to 132 ppb and averaged 10.0 ppb, which was three times higher than that of Tier 1 and 23% that of Tier 2. External material rinsed from the SAV demonstrated an average concentration of 49,237 ppb which was 0.7 times that of Tier 1. Invertebrate tissue samples averaged 7.0 ppb, which was an increase of 1.2 times compared to that of Tier 1 and 0.3% that of Tier 2.

On average, sediment and SAV tissue tPAH concentrations, more than one year after MC252 oil was documented in the Chandeleur Islands, were greater than Tier 1 samples and 39 to 23% of Tier 2 samples. A reduction in oil concentrations in external materials on SAV indicated that surface oiling from the DWH spill was no longer present, but internal tissue concentrations were still impacted by MC252 oil and residual concentrations of MC252 contaminants still resided within SAV bed sediment with four times more petrogenic PAHs present than in Tier 1. However, due to significant weathering of PAHs and the MC252 biomarkers (Emsbo-Mattingly and Martin, 2015), only two of 51 Tier 3 sediment samples forensically matched MC252 oil compared to 70% of Tier 2 samples.

3.3.2 SAV Health and Condition Characterization Results

The Tier 3 sampling effort focused on two regions of the Chandeleur Islands at 10 stations in the moderately oiled stratum and 14 stations in the heavily oiled stratum (Figures 14 and 15). During Tier 3 field observations no oil was observed on the seagrasses or sediments within the seagrass meadows were visibly observed; however, oil was observed on some of the shorelines in proximity to the nearshore stations. No evidence of seagrass mortality in visual observations was detected and there was evidence of new shoot and rhizome growth at the edges of the seagrass patches. Likewise, in the 120 seagrass/sediment cores that were processed, no oil was visibly observed and there was no evidence of acute or chronic stress to the plants. Each species had an abundance of healthy apical meristems and new shoot and rhizome growth was evident in all of the processed samples.

Four seagrass species were observed and quantitatively documented in the visual assessment quadrats and in the cores. The majority (61%) of the cores had just one species present, but



multiple species meadows were also observed in both strata (Figure 20 top panel). At least a third of the stations were comprised of two species meadows, consisting of different combinations of *H. wrightii*, *R. maritima*, *T. testudinum* and *S. filiforme* (Figure 20 bottom panel). Overall, *R. maritima* was the most frequently encountered species followed in order by *H. wrightii*, *T. testudinum* and *S. filiforme*.

There was some variability in the overall species abundance between the two strata. In the heavily oiled strata *H. wrightii* and *R. maritima* each occurred in eleven of the 14 stations sampled. *Halodule wrightii* was more frequently observed in the cores than any of the other three species (Figure 21, top panel). *Ruppia maritima* was the next most frequently encountered species. *Thalassia testudinum* was found at five of the fourteen stations and was the next most frequently encountered species. *Syringodium filiforme* was the least frequently encountered species and was observed in only one station. Where seagrasses occurred, total seagrass cover in the heavily oiled strata ranged from 72% to 100%, averaging 92.2% (±2%).

In the moderately oiled stratum, *R. maritima* was observed in nine of the 10 stations and was the most frequently encountered species (Figure 21, bottom panel). *Thalassia testudinum* was the next most frequently encountered species and was present in five of the ten stations. *Halodule* wrightii was less abundant than *T. testudinum* and present at three of the ten stations. Like in the heavily oiled strata, *S. filiforme* was the least abundant species. Total seagrass cover in the moderately oiled strata ranged from 45% to 100%, averaging 90.2% (±7.5%).

Ruppia maritima shoot densities ranged from 1,195 (± 308) shoots m⁻² in the heavily oiled strata to 2,654 (± 385) shoots m⁻² in the moderately oiled strata (Figure 22). Compared to pre-spill data for *R. maritima* sampled in the moderately oiled strata in June 2009 (Fodrie and Heck, 2011) and June 2010 (Ken Heck, Dauphin Island Sea Lab, unpublished data), there was a relatively lower shoot density in the heavily oiled strata in September 2011 during the Tier 3 survey. In Tier 3 the moderately oiled strata had similar densities to pre-spill densities in 2009. In 2010 (pre-spill), only one sample had *R. maritima*.

Halodule wrightii shoot densities in Tier 3 (June 2011) were higher than R. maritima, ranging from 3,451 (\pm 1,044) shoots m⁻² in the heavily oiled strata to 4,067 (\pm 3,840) shoots m⁻² in the moderately oiled strata (Figure 23). Compared to pre-spill data from the moderately oiled strata, these densities were similar to the one station sampled in 2010 that had H. wrightii (Fodrie and Heck, 2011) and higher than in 2011 (Ken Heck, Dauphin Island Sea Lab, unpublished data).

Thalassia testudinum shoot densities in Tier 3 ranged from 518 (\pm 72) shoots m⁻² in the heavily oiled strata to 739 (\pm 134) shoots m⁻² in the moderately oiled strata (Figure 24). Densities in the heavily oiled strata were slightly lower than both of the pre-spill samples, while densities in the moderately oiled strata were similar to the pre-spill data in 2010 (Fodrie and Heck, 2011) and 2011 (Ken Heck, Dauphin Island Sea Lab, unpublished data).



Syringodium filiforme was present at one station in each of the strata in Tier 3, where densities were 1,775 shoots m⁻² in the moderately oiled strata and 1,003 m⁻² in the heavily oiled strata. These densities are comparable to pre-spill densities observed for *S. filiforme* during pre-spill sampling in 2010 and 2011 (Fodrie and Heck, 2011; Ken Heck, Dauphin Island Sea Lab, unpublished data).

3.3.3 Change Detection Results

The results of the SAV mapping for the three years are shown in Figure 25. As described previously, the entire length of the Chandeleur Islands was mapped for 2010 and 2011, while 2012 mapping was limited to the five Areas of Concern where oil exposure was documented and it was hypothesized that these areas had the greatest possibility of SAV losses due to oil exposure.

The first phase of change detection identified changes that occurred along the full length of the island chain between fall 2010 and fall 2011. These changes are summarized in Table 7. During this time period, 483 acres of SAV were lost and 711 acres were gained. By plotting the change classes, the spatial location and concentration of the SAV losses could be observed.

The second phase of change detection identified changes that occurred between 2010, 2011, and 2012 in the five Areas of Concern. Figure 26 shows a map of changes for Area of Concern 1. In this figure, green symbolizes areas that were mapped as SAV on all three dates. These areas, therefore, experienced no change in SAV. Similarly, the areas with no coloration and the underlying imagery visibly represent areas that were mapped as NOT SAV on all three dates.

The yellow class represents areas where SAV was present (SAV) in 2010, absent (NOT) in 2011, and still absent (NOT) in 2012. For the purposes of this report, these areas are referred to as "persistent loss". In other words, there was an initial loss of SAV between 2010 and 2011, with continued absence (no recovery) in 2012. The red class, referred to as "delayed loss", indicates that SAV was present (SAV) in 2010, still present (SAV) in 2011, but was absent (NOT) in 2012. Orange represents areas where SAV was present (SAV) in 2010, absent (NOT) in 2011, and present again (SAV) in 2012. Thus these orange areas represent initial losses that subsequently revegetated. Because of the relatively quick rate of revegetation, these areas are not being considered as potentially impacted by oil. The three blue colors indicate areas that were NOT SAV in 2010 and therefore are not candidates for SAV loss from base year 2010.

As described in the Section 2.10.3, Core Areas were delineated to omit areas of natural processes that might be responsible for losses. For example, in Figure 25 the Core Area within Area of Concern 1 eliminates the overwash fans that were visible in the underlying 2012 post-Hurricane Isaac imagery. These fans resulted when waters washed over the Island during the hurricane, depositing fans of sediment on the western side of the Island, and burying existing SAV. The northern half of Area of Concern 1 was also excluded, because this part of the Island offered little physical protection to erosion and overwash burial and the beds of SAV in this area have



been suffering serious losses prior to 2010 and since. Figures 27-30 show the results of the change detection and the Core Areas for Areas of Concern 2-5. The changes across the three years are summarized, in acres, for the five Core Areas in Tables 8-12. In each table the persistent loss and delayed loss are highlighted in yellow and summed together as "Total Loss" at the bottom of the table.

The persistent and delayed losses within the Core Areas are being proposed by the SAV TWG as potential injury due to oil exposure from the DWH spill. These losses within the five Core Areas are summarized below in Table 13. A total of 111.7 acres were identified as persistent loss and 159.5 acres were classified as delayed loss. Therefore the Total Loss (persistent + delayed loss) for all five Core Areas was 271.2 acres.

3.3.4 SAV Injury Recovery

Initial inspection of the persistent loss polygons (see section 3.3.3) from October 2012 identified 80,042 individual polygons with a wide range of sizes and a large proportion of very small areas < 100 m². Many of these smaller areas would fall into a category referred to as "narrow gap group" injuries (Fonseca et al., 2004). Depending on the species, the smallest areas are likely to recover rapidly if there is no impairment from oil exposure or disturbance from extreme environmental conditions (e.g., severe storms). For the relatively faster growing species occurring in the Chandeleur Islands (*H. wrightii*, *S. filiforme* and *R. maritima*) the persistent loss areas < 100 m² are expected to recover in less than one year, or approximately one growing season. Based on this expectation and confirmation from prior experience in NOAA's Mini 312 damage assessment program, the smaller polygons were identified and excluded, and only persistent loss polygons exceeding 100 m² (0.0247 acres) were included. The initial analysis identified 583 individual persistent loss polygons with a total area of 51.08 acres. These polygons ranged in size from 0.02 to 2.18 acres, and the distribution was highly skewed with a median area of 0.04 acres. Only a few of these polygons were very large; six exceeded 1.0 acre in size.

For the next step, the seagrass recovery time in each of the 583 persistent loss polygons was calculated. These calculations were made with the following assumptions: 1) recovery rates would be based on available empirical data and the spatially explicit modelling approach described in Fonseca et al. (2004), 2) all calculations would be based on recovery of *H. wrightii*, the fastest growing seagrass species occurring in the Chandeleur Islands, 3) recovery times are not impaired by environmental conditions or any lingering oil exposure, and 4) only recovery times > one year would be considered relevant. The implications and uncertainties for each of these assumptions will be addressed in Section 4.3.

Of the 583 polygons the analysis identified, 131 individual polygons (33.85 acres) had recovery times ≥ one year. To summarize the model estimates, the recovery times for the individual polygons were binned by year (Table 14). Binning the acreage by the years to recovery shows that the distribution of acreages in the different time categories is highly skewed. Approximately



one third of the persistent loss polygons included in this analysis (11.24 acres) have predicted recovery times of between one and two years. Another 12.65 acres (37.4%) have recovery times of between two and ten years. The remaining 9.96 acres make up six large polygons with recovery times ranging from 14 to 26 years.

4 Discussion

4.1 Oil Exposure

The geographic size and ecological complexity of the DWH spill led to numerous response and damage assessment efforts. These efforts resulted in the compilation of a large number of exposure datasets associated with various impacted media. The primary exposure data investigated in this work are tPAH measurements.

As detailed earlier in this report, exposure of SAV to oil from the DWH spill was greatest at the Chandeleur Islands. In September 2010, average tPAH concentrations in sediment within SAV beds were eight times greater than the average concentration of samples collected in June 2010. The September 2010 concentrations were also eight times higher than the average ambient tPAH for undeveloped barrier islands in the GOM (Rouhani et al., 2015). It is important to note that the ambient tPAH concentrations developed by Rouhani equaled the average tPAH concentration of the Tier 1 Chandeleur sediment samples (41 ppb).

Some sediment concentrations within the Chandeleur Islands SAV beds were almost 100 times ambient levels. Nearby sediment samples along coastal vegetation on the back side of the Chandeleur Islands were as high as 3,300 times ambient levels and often two orders of magnitude higher than ambient levels. Assuming Tier 1 Chandeleur tissue samples were also representative of ambient levels, Tier 2 plant and invertebrate tissue tPAH concentrations were over 13 and 400 times higher than ambient conditions. Finally, over 70% of Tier 2 sediment samples were forensically matched (A or B) to MC252 oil.

SCAT surveys first documented oiling of the Islands on May 8, 2010 and visible oil on water was also evident in May 10, 2010 NAIP imagery. Review of individual SAR footprints demonstrate that the heaviest and most frequent oiling occurred from mid-June to mid-July 2010 (see Appendix C). Widespread light to heavy oiling throughout the Islands was confirmed during SCAT surveys on June 21st through the 28th and July 11th through the 13th. Heavy oiling of the Chandeleur Islands was last identified by SCAT teams in February 2011.

PEMD results indicated PAHs were biologically available in SAV beds at low levels in August and September 2010, four months after initial oiling. Total PAH concentrations were greatest in the southern portion of the Chandeleur Islands and generally declined toward the north.



Figures 31 and 32 demonstrate the Tier 1, 2 and 3 tPAH results for sediment and tissue samples, respectively, for the middle region of the Islands. Figures 33 and 34 demonstrate the Tier 2 and 3 tPAH results for sediment and tissue samples, respectively, for the southern region of the Islands. SCAT data is also shown in these figures.

4.2 SAV Assessment

The five locations sampled in the Tier 2 and Tier 3 assessments were characteristic of the wide range of seagrass community types found throughout the northern GOM (Handley et al., 2007). However, no single location could be considered representative of the other four; the seagrass communities at each location varied substantially in species composition, spatial distribution, and abundance.

4.2.1 North-Central Gulf SAV Assessment

Although in different combinations, multiple species meadows were observed in four of the five locations. Robinson Island, just inside of Perdido Pass and Horn Island in Mississippi Sound were the only sites where just one species (*H. wrightii*) occurred. Big Lagoon, the site furthest from the DWH wellhead, had relatively larger meadows consisting of two species: *T. testudinum* and *H. wrightii*. In Big Lagoon, the seagrass beds were continuously distributed behind the barrier island and along the mainland shoreline on the north side of the lagoon. Although SCAT data identified light oiling along shorelines near seagrass beds, preliminary TPH sediment results for Big Lagoon and Robinson Island were all non-detect except for one sample in Big Lagoon. Therefore, these coastal mainland sites were not sampled in Tier 3.

4.2.2 Petit Bois and Horn Islands SAV Assessment

At Horn and Petit Bois Islands in Mississippi Sound, the seagrass meadows consisted of primarily two species: H. wrightii and a small amount of S. filiforme. Located on a narrow shelf running parallel along the north shore of the barriers, these seagrass meadows were protected from the direct influence of the open Gulf, but exposed to the northerly wind fetch and wave energy from Mississippi Sound. The seagrass meadows consisted of various sized patches growing in shallow water (< 2.0 m) among dynamic underwater sand waves oriented nearly perpendicular to the shoreline. At the time of sampling, the meadows appeared to have the same distribution and biological characteristics described by Eluterius in 1971 and 1973 (see Eluterius, 1987). Sand waves migrating along the shallow shelf partially buried the seagrasses and prevented the formation of large continuous meadows, allowing only the most tolerant and fast growing species (*H. wrightii*) to survive the chronic physical disturbance. Periodic freshwater discharges into Mississippi Sound contribute additional stress to the seagrasses growing at Horn and Petit Bois Islands (Eluterius, 1987). Although SCAT data revealed only small segments of the very eastern and western ends of Horn and Petit Bois Islands to be either moderately or heavily oiled, the presence of tar balls on the shoreline suggests that oil reaching Mississippi Sound could have encountered the seagrass beds as it was transported shoreward. During the winter of 2010, weathered oil (e.g., tar balls) may have been redistributed in Mississippi Sound



and stranded on the north shore of Horn and Petit Bois Islands. The indirect transport mechanism and local physical conditions on the narrow shelf needed to entrain the oil within the seagrass beds, however, would have limited exposure. As mentioned earlier, over 90% of the preliminary TPH sediment results collected within and near seagrass beds at Horn and Petit Bois Islands were non-detect. Finally, analysis of post-oiling imagery did not show any changes in the aerial abundance of seagrasses at Horn and Petit Bois Islands between 2010 and 2011.

4.2.3 Chandeleur Islands SAV Assessment

The largest and most diverse seagrass distributions were observed in the Chandeleur Islands. Four species of seagrass (T. testudinum, H. wrightii, R. maritima, S. filiforme) were recorded in the quadrats and cores and a fifth species (H. engelmannii) was observed visually in the Tier 2 assessment (Table 6 and Figures 20 and 21). The meadows are all located on a broad and shallow shelf on the west side of the Chandeleur Islands (Figure 25), where they are protected from the easterly wind fetch and the direct influence of waves from the open Gulf. They are also protected by a back barrier sand bar, which shields westerly, southwesterly, and northwesterly winds coming across Chandeleur Sound. The shelf has a variable topography with substrates consisting primarily of sand, fine sand, and silt sediments. The central and inner portions of the shelf have numerous intertidal shoals and perennially submerged substrates (< -1-2 m), which are bisected by slightly deeper sloughs and basins in the tidal passes between the Islands. The outer portion of the shelf (western margin) is characterized by a slightly elevated sand bar which provides a north-south axis-oriented shoal that slopes out into the deeper open water of Chandeleur Sound. Generally, this shoal demarcates the most western distribution of seagrasses on the shelf. This shoal provides a baffle against wind and wave energy originating from Chandeleur Sound, which, in part, makes it responsible for creating the relatively lower energy conditions suitable for seagrass growth. In between the sand flats of the backside of the island chain and the back barrier sand bar is a lagoon with water depths ranging from <1m to nearly 4 m. This lagoon provides the Chandeleur Islands' primary seagrass habitat. At its widest point, the shallow shelf supporting the seagrass meadows extends as much as 1 to 1.5 km westward of the Islands, making these the largest meadows surveyed, and generally one of the largest areas of seagrass beds between Pensacola Bay, FL and Laguna Madre, Texas (Handley et al., 2007).

Seagrasses stabilize and trap sediments on the shelf and are intricately linked to the long term physical stability and survival of the barrier island system. The islands and shoals surrounding them are slowly migrating west towards the Mississippi Delta. The shelf behind the Islands provides a relatively stable foundation for the barriers, which helps maintain the elevation of the Islands above sea level and storm surge. Seagrasses are critically important in mitigating this process, stabilizing the shelf, and trapping additional sediments. This process is one of the most important ecological services provided by seagrass communities (Barbier et al. 2011) and is especially critical for the Chandeleur Islands due to its relatively starved sediment system.



As discussed earlier in this report, the Chandeleur Islands were impacted by multiple oiling events which occurred as early as May 4: 2010 and continued through July 2010. SCAT surveys identified extensive heavy oiling in the middle and southern regions of the Islands as early as May 8, 2010 and as late as February 2011. According to Graettinger et al. (2015), the Chandeleur Islands were the first shallow water habitats in the northern Gulf to experience exposure from MC252 oil due to its close proximity to the DWH wellhead and surface trajectories of the oil during the spill. This information suggests that the Chandeleur Islands were one of the most probable locations for acute and chronic MC252 oil exposure. The susceptibility of the Islands' seagrasses to MC252 oil exposure is attributed to several factors. First, chronic exposure was confirmed in September 2010 and June 2011 by high concentrations of tPAH in plant and animal tissue, sediment, and PEMD data. Second, the seagrass meadows of the Chandeleur Islands were the closest northern GOM seagrasses to the MC252 oil source (125 km). Third, they did not have the same extent of protection as some of the other seagrass communities in the northern GOM. The Chandeleur Islands are oriented north-south. Any MC252 oil transported northward from the source was less weathered and minimally impeded by the barrier islands, making them more physically exposed to oiling potential. Visible surface trajectories (Graettinger et al., 2015) showed oil coming into the Chandeleur Islands from the south and southwest across the outer shoal on the western boundary of the shelf and across the shallow seagrass beds to the beach and marsh shorelines (Figure 16). Oil was also transported from the open waters of the Gulf through several tidal passes between the Islands and onto the shallow seagrass beds on the shelf. Finally, the substantially lower energy, shallow water environments of the Chandeleur Islands fostered oil exposure. Aerial photography and SAV assessments at the Chandeleur Islands demonstrate seagrass canopies reached the water surface. Here, MC252 oil intercepted the seagrass canopy, settled onto the sediments among the seagrass meadows, or was stranded on the shoreline.

The next closest seagrass meadows are at Horn and Petit Bois Islands in Mississippi Sound (162 km from the DWH wellhead). These seagrass meadows are located on the north side of the east-west oriented islands and are on a very narrow shelf. As such, the islands deflected oil that was being transported north into Mississippi Sound and initially protected the seagrass meadows from direct exposure.

Tier 2 SAV assessment results affirmed initial concerns and objections to the incorporation of reference sites in the survey design, which was insisted upon by the Responsible Party. During the field assessment design, the SAV TWG discussed the establishment of reference sites, representative of GOM seagrass meadows, with which to compare potential oil exposure sites. At this time, oil was still being discharged from the DWH wellhead and the ultimate geographic extent of exposure was uncertain, making the establishment of reference sites problematic. Although the geographic scope of the five Tier 2 sites was relatively narrow and even included locations with no detected exposure at the time (e.g., Big Lagoon), the lack of a typical seagrass distribution in this region of the northern GOM made the establishment of reference sites inoperative. Even if sampling was conducted further east (Florida panhandle) or west (Laguna



Madre, TX), beyond the confirmed extent of oiling, finding a site representative of those areas exposed to MC252 oil in the Chandeleur Islands would have been unlikely due to their unique characteristics.

Instead, the abundance of individual species (shoot density) at the Tier 2 and 3 assessment sites was compared with data from other published SAV surveys in the northern GOM. In general, the post-spill Tier 2 and Tier 3 shoot densities of *T. testudinum*, *H. wrighti*, and *S. filiforme* fell within the species range (although wide) previously reported for locations in Mississippi (Mississippi Sound), Alabama (Grand Bay, Perdido Bay), Florida (Big Lagoon, St. Joe Bay) and Texas (Laguna Madre, Corpus Christi Bay, San Antonio Bay). However, these studies can only be used as a qualitative and imprecise estimate due to their wide range of environmental conditions (e.g. water depth, salinity) and differing sample times (seasonal abundance).

The delay in mobilizing and executing field sampling efforts after the initial spill also impacted the SAV TWG's assessment. For reasons beyond the group's control, Tier 2 field assessments were not initiated until the last week of August. The delay in sampling prevented direct observation of SAV response to acute exposure as oil first came ashore in May and experienced repeated oiling from mid-June to mid-July 2010. The delay also limited the collection of data on seagrass distribution, abundance, and health prior to exposure, rendering a statistically robust "before and after" (e.g., BACI) sampling design neither practical nor possible. The best empirical estimate of the SAV species composition and abundance in the Chandeleur Islands, prior to exposure, comes from the survey of 12 stations in the northern and central regions of the Chandeleur Islands on June 2, 2010 (Fodrie and Heck, 2011) and June 2011 (Ken Heck, Dauphin Island Sea Lab, unpublished data). Although this did not cover the entire scope of the area exposed to MC252 oil in the Chandeleur Islands, it provides important complementary information regarding species composition and abundance at the northern extent of exposure. Combined with the wider geographic sampling in the Tier 2 and Tier 3 assessments, the SAV community in the Chandeleur Islands exposed to MC252 oil was qualitatively and quantitatively characterized.

A long history of physical instability resulting from natural processes (severe storms, e.g.; Hurricane's Camille, George, and Katrina) and anthropogenic (sand starvation, freshwater discharge) disturbances has reduced the overall physical size of the Chandeleur Islands and steadily diminished the areal abundance of seagrasses (Poirrier and Franze, 2001; Franze, 2002; Poirrier and Handley, 2007; Handley et al., 2007). Still, the biodiversity and function of the Chandeleur Islands seagrass habitats are among the highest in the GOM (Eluterius, 1987; Handley et al., 2007; Fodrie and Heck, 2011). The Fodrie and Heck (2011) study in June 2010 and the Tier 2 samples in September 2010 confirmed the presence of diverse, spatially extensive, and functional seagrass communities, the same communities that were repeatedly exposed to MC252 oil.



Tier 3 sampling in June 2011, re-affirmed the abundance and biodiversity of seagrasses in the moderately (central) and heavily (southern) oiled regions of the Chandeleur Islands. Tier 3 SAV assessments were performed in offshore, inshore, and mid-shore, seagrass beds - across the entire shelf west of the Islands, providing more comprehensive coverage of the areas where exposure in Tier 2 was identified. In Tier 3, like in Tier 2, various-sized patches and relatively larger, contiguous meadows of seagrasses were observed among various-sized gaps of unvegetated substrate. No direct evidence of seagrass stress or mortality was observed in Tier 3, confirmed by the collected and processed live samples and examined cores. However, there were evident differences in the seagrass community between Tier 2 (September 2010) and Tier 3 (June 2011). For instance, a substantially greater presence and abundance of Ruppia maritima samples were observed in Tier 3. Additionally, extensive flowering and seed production of Ruppia maritima was observed in Tier 3 and not in Tier 2. This can be attributed to the variations in sexual reproductive efforts among, and even within, a species, which is influenced by environmental conditions and season. Ruppia maritima is one of the most fecund seagrasses in the northern GOM. It is known to rely on flowering and seed production in late spring and early summer in the northern GOM in order to replenish biomass. Flowering and seed production are important for both meadow maintenance and spatial dispersal of Ruppia maritima as well as other species. Ruppia meadows are frequently referred to as either ephemeral or annual where all of the biomass senesces at the end of the growing season and is replaced in the next year by seed germination and seedling growth.

In the Chandeleur Islands, all seagrass canopies extend vertically above the sediments and up into the water column, making them susceptible to MC252 oil exposure. Probable oil exposure increased during low tides when the leaf canopies reached the water surface and oil was entrained on the shelf. The *Ruppia* canopy and flower development during the Tier 3 event was robust and taller than any of the other species, such that *Ruppia* occupied the entire water column nearly everywhere it was observed and during most tidal stages.

This abundance of *Ruppia* and extensive flowering was mostly likely the case in May and June 2010 when oil was first transported into the near shore environment of the Islands. Consequently, *Ruppia* biomass and sexual reproduction were one of the most severely exposed biological components of the shelf ecosystem. The relatively lower abundance of *Ruppia* in Tier 2 could have been the result of acute stress caused by MC252 oil exposure; however, intra-annual variation in growth could also be a cause (see Eluterius, 1987).

Alternatively, the robust abundance of *Ruppia* in June 2011 could have resulted from the emergence of seedlings that were stored in a sediment seed bank. Sediment seed banks can be derived from flowering events prior to oiling. At least two other species present in the Chandeleur Islands, *H. wrightii* and *S. filiforme*, are known to produce seeds that can be retained in a sediment seed bank. These seeds can germinate in future growing seasons (McMillan, 1991; Inglis, 2000). Since MC252 oil exposure was detected in the sediments, we cannot rule out the



possibility that the seagrass seed banks were impacted; however, the effect of oil on the longevity of seagrass seeds residing in the sediment seed bank is currently unknown.

Field sampling of SAV metrics was designed to assess seagrass health and condition and interpret point samples with respect to oil exposure. However, these assessments were limited to areas where seagrass was present. As mentioned earlier, Tier 2 assessments were conducted more than 2 months after initial exposure, preventing direct observation and documentation of acute seagrass stress and mortality during oiling. Moreover, Tier 2 and Tier 3 sampling events were 9 months apart. In this time, changes in seagrass abundance and distribution could have occurred as a result of chronic exposure, which would not have been detected by the limited spatial and temporal scope of the point samples.

Areal coverage of seagrass was not continuous and heterogeneously distributed across the shallow shelf west of the Islands based on observations made during the SAV assessments and analysis of aerial imagery. The seagrass meadows ranged in size from small isolated patches a few meters in diameter to relatively larger and continuous meadows hundreds to several thousand square meters in size. Patches of healthy, growing seagrasses were interspersed among unvegetated areas (gaps). These patches varied in size. The presence of unvegetated areas among healthy seagrass meadows is not uncommon where either bioturbation (e.g., sediment excavation, grazing) or some form of physical disturbance (e.g., waves, prop scarring) disrupts seagrass meadows. It is possible, however, that some of the unvegetated patches observed in September 2010 were not the result of acute stress and mortality. The formation and persistence of new unvegetated patches following initial exposure can, in time, be a long term indication of seagrass response to acute and/or sustained stress as a result of oil exposure. Consequently, the injury assessment was expanded to a larger spatial scale, incorporating aerial imagery and change detection.

Aerial imagery of the Chandeleur Islands was used to supplement point sampling and increase the scale and scope of inference to assess seagrass meadows. Imagery acquired under the AITWG work plan (NOAA, 2013a) in the fall of 2010, 2011, and 2012, was utilized. Traditional remote sensing and change analysis investigations interpret and measure the areal seagrass coverage at each time interval, and then calculate an absolute change in acres between two intervals. Factors potentially responsible for the change in coverage are assumed to be homogeneously distributed across the entire interpreted area. This is not the case for the Chandeleur Islands. Seagrass meadows are not continuous and are often patchy in areas and, oiling, as shown by SCAT data, SAR oil on water estimates and tPAH sampling results, was clearly heterogeneous. Additionally, there could be areas of seagrass loss due to exposure and areas of gain that were not exposed. Therefore, documenting both categories is more representative of a response to heterogeneous oiling than a simple calculation of total change across the entire area.



The seagrass change analysis was performed by stratifying seagrass meadows into five Areas of Concern based on available SCAT data (Michel et al., 2014), May 10, 2010 NAIP aerial imagery which identified early oiling trajectories, Tier 2 and 3 tPAH data, and May to July surface oil trajectories (Graettinger et al., 2015). In order to minimize uncertainty, areas where changes in seagrass coverage could be attributed to natural processes (e.g.; island overwash fans and erosion from Hurricane Issac) and problematic interpretation signature areas (turbidity, deep water, sun glint) were excluded from the change analysis

The seagrass injury assessment calculated 111.7 acres of persistent loss of areal seagrass coverage in the Chandeleur Islands that could not be attributed to natural processes or interpretation error, but to oil exposure from the DWH spill. The baseline imagery used in this analysis came from October 2010, almost five months after oil first reached the Chandeleur Islands. This means that the 111.7 acres of seagrass loss is most likely a conservative estimate. Additionally, 159.5 acres of areal seagrass coverage were classified as delayed loss. Delayed loss may have been the result of either chronic exposure, latent effects of oil on sediment biogeochemical processes, seagrass mortality, and/or seagrass recruitment.

4.3 SAV Recovery Calculations

Assumptions used in the recovery calculations likely overestimate seagrass recovery in the Chandeleur Islands. First, recovery is based on unimpaired vegetative regrowth into the injured polygons from adjacent side populations of seagrass. In the Tier 2 and Tier 3 SAV assessments, healthy seagrass populations were observed among the unvegetated areas (persistent loss polygons), which would contribute to recovery. However, the calculations credit recovery to side populations that may not exist, as some of the persistent loss polygons are not bordered by seagrass; for example, along the inner or outer edges of meadows or adjacent to deep edges or shallow shoals devoid of vegetation.



into the persistent loss polygons. Therefore, the modeled recovery rates are likely higher than the growth rates of transplanted seagrasses. So, although the calculations do overestimate recovery, the *H. wrightii* growth rates in the northern Gulf are under-estimated as Fonseca et al. (1987) suggest.

The assumption that all calculations are based on one species, *H. wrightii*, also leads to an overestimation of recovery. As indicated by the Tier 2 and 3 assessments, five species of seagrass occur in the Chandeleur Islands and all five may have been injured by exposure. One species, *H. engelmamii*, is rare and has no published growth data. Therefore, this seagrass species was not considered in the modelling. Two other seagrasses documented in the Chandeleur Islands, *H. wrightii* and *S. filiforme*, are relatively fast spreading, opportunistic species. According to empirical studies in South Florida and the Caribbean basin (Duarte, 1991; Gallegos et al., 1994; Marba and Duarte, 1998; Fonseca et al., 2004), these two species have similar growth rates. Therefore, the model did not distinguish between them in the recovery estimates. No comparable growth data and modeling studies were available for *R. maritima*. However, *R. maritima* and *H. wrightii* are both relatively fast growing, opportunistic species. Therefore, *H. wrightii* growth data were assumed a reasonable surrogate for *R. maritima*. Additional uncertainties associated with the life history of *R. maritima* are addressed later in the discussion.

The last species found at the Chandeleur Islands is T. testudinum, commonly referred to as Thalassia. This species is especially important in regards to the model assumptions. Where Thalassia occurs it frequently constitutes the largest amount of biomass and provides many important ecological services, such as fishery habitat, sediment stabilization, nutrient recycling, and carbon sequestration (Zieman, 1982, Zieman and Zieman, 1989; Fodrie and Heck, 2011). However, quantitative assessments of the relative abundance and spatial distribution of the different seagrass species in the Chandeleur Islands have never been conducted. The best estimates of the relative abundance of the seagrasses in the Chandeleur Islands are derived from ground truthing observations associated with prior seagrass mapping efforts in the northern GOM (Handley et al., 2007), Tier 2 and 3 samples, and surveys by Fodrie and Heck (2011). It is estimated that as much as 20% of the seagrass cover in the Chandeleur Islands could be monotypic *Thalassia* meadows while another 30% of the meadows could be *Thalassia* mixed with either or all of the other species (Larry Handley personal communication). In another, more recent study, benthic core samples obtained in conjunction with trawl surveys of fishes in the northern and central regions of the Chandeleur Islands documented *Thalassia* in seven of thirteen stations sampled on June 2, 2010, and in four of thirteen stations sampled on June 20, 2011 (Fodrie and Heck, 2011; Ken Heck, Dauphin Island Sea Lab, unpublished data). In 2010, all of the cores with *Thalassia* were monotypic, while in 2011 *Thalassia* co-occurred with *R. maritima* in three of the four stations. The Fodrie and Heck (2011) study did not include sites in the southern region of the Islands. Only Tier 2 and 3 sample data and limited spatial information on the relative abundance of the different species are available for this region. However, it is clear



that *T. testudimum* was exposed to MC252 oil in the Chandeleur Islands, but, the areal coverage potentially affected by oil exposure for each species is uncertain.

Uncertainty regarding the relative abundance of *T. testudinum* in the *H. wrightii*-based model could lead to a large underestimate of recovery times. This is mainly due to the fact that *T. testudinum* is the slowest growing species in the seagrass community (Zieman, 1982; Williams, 1988; Williams, 1990; Duarte, 1991; Marba and Duarte, 1998). Rates of growth into disturbance gaps are documented to be as much as three to four times slower for *T. testudinum* than for *H. wrightii* and *S. filiforme* (Kenworthy et al., 2000; Kenworthy et al., 2002; Fonseca et al., 2004; Hammerstrom et al., 2007). Therefore, the largest underestimates of recovery time in the Chandeleur Islands would apply mainly to persistent loss polygons in monotypic meadows where natural recovery depends solely on the adjacent *T. testudinum* populations. For example, if the 0.7 acre persistent loss polygon in Table 14 were located in a monotypic *T. testudinum* meadow and the recovery rate calculation was scaled to that of *T. testudinum*, recovery time would increase from 8 to 30 years.

Alternatively, if it is assumed that mixed species meadows serve as the source of regrowth into the injured gaps, the faster growing species should precede the slower growing *Thalassia* and initiate seagrass recovery. In this case the modeling approach used approximates the natural process of recovery often reported for mixed sub-tropical and tropical seagrass communities of *Halodule, Syringodium* and *Thalassia* (Zieman, 1982; Williams, 1990). Assuming there are no differences placed on the "relative values" of the individual species, modeling the re-growth of *Halodule* or any other fast growing species should be an acceptable "best case scenario" for recovery in the Chandeleur Islands. This has, in fact, been a precedent set in NOAA's seagrass Damage Assessment and Restoration Program in the Florida Keys National Marine Sanctuary (FKNMS). The temporary substitution by faster growing species (e.g., *Halodule* for *Thalassia*) has been considered acceptable for restoring the ecological services provided by seagrasses, whether or not the injured community is a mixed assemblage of species or a monotypic meadow (Fonseca et al., 2004).

The recovery calculations relied exclusively on asexual reproduction and vegetative re-growth (Fonseca et al., 2004). This increased uncertainty because it didn't include sexual reproduction and fragment dispersal. In addition to clonal growth, seagrasses flower and reproduce sexually (McMillan, 1976). Flowering plants develop seeds that maintain and disperse populations (Orth et al., 2006). All of the seagrasses growing in the Chandeleur Islands are known to reproduce and disperse sexually. But fecundity and seed dispersal varies widely between species and even among populations of the same species (McMillan, 1976; Whitfield et al., 2004; Hammerstrom et al., 2006; Orth et al., 2006). Seed dispersal and its contribution to recovery could be vital for some of the species at the Chandeleur Islands. However, this information has yet to be quantitatively incorporated into seagrass recovery models and could not be included in recovery estimates. Even though this information could not be included, it should be noted that oil



exposure in the Chandeleur Islands occurred during the early stages of the seagrass growing season in 2010 (May and June) when flower development would have been vulnerable. The impairment of flower development could have resulted in limited seed production and seeds are needed to initiate recovery in subsequent growing seasons. Hence, the impairment of flower development may have been partially responsible for the persistent and delayed losses identified in 2011 and 2012.

Of all the species growing in the Chandeleur Islands, *R. maritima* is the most fecund. In shallow water, *R. maritima* flowers often grow up to the water surface where they may have been more vulnerable to May and June 2010 oil exposure than other species. Oil exposure was documented in the sediments where seeds of *H. wrightii*, *S. filiforme*, and *R. maritima* are normally dispersed, stored, and germinated. Some species, including *H. wrightii* and *S. filiforme*, may store seeds for several years in sediment seed banks, which supply propagules for future recruitment and recovery once disturbance stress has decreased (McMillan, 1981; McMillan, 1991; Whitfield et al., 2004; Hammerstrom et al., 2006). The initial period of oiling (May and June 2010) and subsequent sediment oil exposure may have also affected buried seed banks, as well as seed germination and seedling recruitment of all seagrass species.

In summary, the change analysis performed in the five Areas of Concern at the Chandeleur Islands identified 80,042 individual persistent loss polygons totaling 111.7 acres. A large portion of these polygons ($\approx 93\%$) was very small, unvegetated areas ($< 100 \text{ m}^2$). Without additional natural disturbances (e.g. severe storms, bioturbation), or lingering effects from oil exposure, these small gaps in the seagrass cover would likely become revegetated by the relatively faster growing species (H. wrightii, S. filiforme, and R. maritima) in one growing season. If any of the persistent loss polygons were located in monotypic T. testudinum meadows, the total amount of acreage shown in Table 14 would be underestimated and the time to recovery would need to be increased by a factor of 3.86, which is the relative difference in the modeled growth rates of T. testudinum and H. wrightii. This would increase the acreage for the one to ten year recovery categories. Since specific species could not be associated with individual persistent loss polygon without a high degree of uncertainty, recovery was conservatively modeled based on rates for the fastest growing species (H. wrightii). This underestimates recovery times of T. testudinum dominated communities, but approximates recovery in mixed species meadows that include the three relatively faster growing species (H. wrightii, S. filiforme, and R. maritima). These three species are estimated to account for as much as 70% of the seagrass cover in the Chandeleur Islands. Finally, the "time to recovery" estimates are even more conservative than suggested above, because the model is based on empirical data from sub-tropical and tropical seagrass ecosystems where growing seasons are longer, water temperatures are higher, and rates of growth are faster. In all, the model estimates are the "best case scenario" for seagrass recovery in the Chandeleur Islands.



4.4 Restoration

The SAV TWG recommends implementing a restoration program for the Chandeleur Island in order to promote seagrass growth and maintain the integrity of the Islands. This program may also include the restoration of dunes and vegetation. In 2000, restoration efforts were conducted at the Islands to stabilize 364 acres of unvegetated washover deposits on 22 overwash fan sites. Smooth cordgrass (*Spartina alterniflora*) plantings were successfully used to stabilize unvegetated areas. The 2000 restoration program highlights the opportunities for similar and more intensive restoration efforts, especially in the areas where seagrasses were lost during the DWH oil spill.

There are several reasons why restoration efforts should be focused on the Chandeleur Islands versus other locations in the GOM. First, the Islands and surrounding waters are considered pristine as demonstrated by baseline, Tier 1 sampling results. The seagrasses of the Chandeleur Islands are isolated from chemical and nutrient contamination, unlike many of the other shallow coastal areas within the Gulf that are adjacent to human populations and urban runoff.

Second, the seagrass beds off the Chandeleur Islands are unique and extremely productive. They are the most biodiverse and largest seagrass beds in north central GOM and are part of the Breton National Wildlife Refuge which is the second-oldest refuge in the U.S. National Wildlife Refuge System. These Islands are prolific environments where hundreds of species of finfish, crustaceans, and wildlife flourish. The heavily vegetated interiors of this fragmented chain are veritable sanctuaries, where juvenile fish, crabs, and shrimp can find refuge, nursery, and feeding grounds, increasing their odds of survival in the Gulf. The Islands' seagrasses also provide habitat and food for green sea turtles and support the overwintering of Red Head ducks.

Third, for generations, recreational anglers have enjoyed world-class fishing associated with seagrass productivity at the Chandeleur Islands. Multiple day and overnight trips from Louisiana, Mississippi and Alabama to the Islands are frequented by anglers from all over the U.S and the world. Anglers can fish the grass flats, coves and inlets and can encounter huge schools of surface feeding redfish, speckled trout, and flounder. Tarpon and sharks are also found and are both prized sportfish known for their aggressive fighting. The seagrasses not only benefit the recreational fishery but commercial as well, as they provide a nursery ground for shrimp and other crustaceans that are caught in GOM waters.

Lastly and most importantly, the seagrasses have helped to create a protective barrier that has stabilized the Islands for hundreds of years. The Islands create a buffer zone between the Gulf and the mainland of Louisiana, protecting the New Orleans area from wind and storm surges associated with hurricanes. The existence of seagrass beds in the Chandeleur Islands is made possible by two critical factors; 1) the presence and persistence of emergent land features (the islands) above sea level that baffle wave and current energy, and 2) a source of sediment to



maintain suitable water depth (≤ 2 m) on the leeward platform where the seagrasses occur. The emergent islands and the platform are a coupled geological unit (barrier island system) slowly migrating west into Chandeleur Sound. The leeward platform is the foundation upon which the islands are perched and maintained above sea level. The seagrasses play an important role in this process, functioning as a stabilizing feature on the submerged platform and helping to maintain its elevation as well as the Islands.

5 References

Barbier, E.B., Hacker, S.D., Kennedy, C., Koch, E.M, Stier, A.C., and B.R. Silliman. 2011. The value of estuarine ecosystems. Ecological Monographs 81:169-193.

Benz, U.C., P. Hofman, G. Willhauch, I. Lingenfelder, and M. Heymen. 2004. Multi-resolution, object-oriented fuzzy analysis of remote sensing data for GIS-ready information. ISPRS Journal of Photogrammetry & Remote Sensing 58:239-258.

Borowitzka, M.A., Lavery, P.S. and Mike van Keulen. 2006. Epiphytes of seagrasses. In: Larkum, A.W.D., Orth, R. J., and C. M. Duarte (eds.) Seagrasses: Biology, Ecology and Conservation. pp 441-461.

Breiman, L., J. H. Friedman, R. A. Olshen, and C. J. Stone. 1984. *Classification and regression trees*. Monterey, Calif., U.S.A.: Wadsworth, Inc.

Breiman, L. Random Forests. Machine Learning, 45(1): 5-32, 2001.

Braun-Blanquet, J. 1932. Plant sociology - the study of plant communities. Koeltz Scientific Books, Koenigstein, West Germany.

Carls, M.G., Holland, L.G., Short, J.W., Heintz, R.A., Rice, S.D., 2004. Monitoring polynuclear aromatic hydrocarbons in aqueous environments with passive low-density polyethylene membrane devices. Environmental Toxicology and Chemistry 23, 1416-1424.

Carls, M.G. and Meador, J.P. 2009. A perspective on the toxicity of petrogenic PAHs to developing fish embryos related to environmental chemistry. Human and Ecological Risk Assessment 15(6): 1084-1098.

Carls, M. G, Larsen, M. L., Lindeberg, M, and S. D. Rice. 2011. Biologically available polynuclear aromatic hydrocarbons in submerged aquatic vegetation near barrier islands after the Deepwater Horizon blowout. Report prepared by the National Oceanic and Atmospheric Administration, National Marine Fisheries Service Auke Bay Laboratories, 17109 Point Lena Loop Road, Juneau, AK 99801. 26 pp.

Continental Shelf Associates, Inc., Martel Laboratories, Inc. 1985. Florida Big Bend Seagrass Habitat Study Narrative Report. Final Report for the Minerals Management Service, Metairie, Louisiana, No. 14-12-0001-30188.



Continental Shelf Associates, Inc., Martel Laboratories, Inc. 1987. Assessment of hurricane damage in the Florida Big Bend Seagrass Beds. A final report for the U.S. Department of the Interior, Minerals Management Service, New Orleans, Louisiana, No. 14-12-0001-29036.

Dawes, C.J., Philips, R.C. and G. Morrison. 2004. Seagrass communities of the Gulf Coast of Florida: Status and Ecology. Florida Fish and Wildlife Conservation Commission Fish and Wildlife Research Institute and the Tampa Bay Estuary Program. St. Petersburg, FL. iv + 74pp.

Duarte, C.M. 1991. Allometric scaling of seagrass form and productivity. Marine Ecology Progress Series 77:289-300.

Duarte, C.M., Middelburg, J.J., and N. Caraco. 2005. Major role of marine vegetation on the oceanic carbon cycle. Biogeosciences 1: 1–8.

Eluterius, L. N. 1987. Seagrass ecology along the coasts of Alabama, Louisiana, and Mississippi. Florida Marine Research Publications 42:11-24.

Embso-Mattingly, S.D. 2015. Chemical Fingerprinting Methodology and the Classification of Nearshore Samples Used in the Deepwater Horizon NRDA. NewFields report to NOAA.

Embso-Mattingly, S.D. and C. Martin. 2015. Distribution and Weathering of Macondo oil in the Nearshore Soils, Sediments, and Tissues Collected Between Spring 2010 to Spring 2012 Based on Chemical Fingerprinting Methods. NewFields report to NOAA.

Fodrie F. J., and K.L. Heck Jr. 2011. Response of Coastal Fishes to the Gulf of Mexico Oil Disaster. PLoS ONE 6(7): e21609. doi:10.1371/journal.pone.0021609

Fonseca, M.S., Thayer, G. W. and W.J. Kenworthy. 1987. The use of ecological data in the implementation and management of seagrass restorations. Florida Marine Research Publications 42:175-188.

Fonseca, M.S., B.E. Julius, and W. Judson Kenworthy. 2000. Integrating biology and economics in seagrass restoration: How much is enough and why? Ecological Engineering 15:227-237.

Fonseca, Mark S., Whitfield, Paula E., Kenworthy, W. Judson, Colby, David, R., and Brian E. Julius. 2004. Use of two spatially explicit models to determine the effect of injury geometry on natural resource recovery. Aquatic Conservation: Marine and Freshwater Ecosystems 14:281-298.

Fourqurean, J. W., Willsie A., Rose C. D., and L. M. Rutten. 2001. Spatial and temporal pattern in seagrass community composition and productivity in south Florida. Marine Biology 138:341–354.

Fourqurean, J.W., Robblee, M.B., Durako, M.J., Hall, M.O., and L.E. Hefty. 2002, Seagrass distribution in south Florida: A multi-agency coordinated monitoring program, in Porter, J.W., and Porter, K.G., eds., The Everglades, Florida Bay, and Coral Reefs of the Florida Keys an ecosystem sourcebook, pp. 497–522.



Franze, C.D. 2002, Barrier island seagrass and geomorphic interactions: a case study of hurricane damage and efficacy of restoration efforts, at the Chandeleur Islands: New Orleans, La., University of New Orleans, M.S. thesis, 69 p.

Gallegos, M.E., Merino, M., Rodriguez, A., Marba, N. and C.M. Duarte. 1994. Growth patterns and demography of pioneer Caribbean seagrasses Halodule wrightii and Syringodium filiforme. Marine Ecolgy Progress Series 109:99-104.

Gacia E, Duarte C. M., and J.J. Middelburg J.J. 2002. Carbon and nutrient deposition in a Mediterranean seagrass (Posidonia oceanica) meadow. Limnology and Oceanography 47:23–32

Green, E.P. and F. Short, F. 2003. World Atlas of Seagrasses. University of California Press, Berkley, CA, 310 pp.

Graettinger, G., Holmes, J., Garcia-Pineda, O., Hess, M., Hu, C., Leifer, I., MacDonald, I., Muller-Karger, F., Svejkovsky, J. and Swayze, G. 2015. Integrating Data from Multiple Satellite Sensors to Estimate Daily Oiling in the Northern Gulf of Mexico during the Deepwater Horizon Oil Spill.

Hammerstrom, K.K., W. Judson Kenworthy, M.S. Fonseca, and P.E. Whitfield. 2006. Seed bank, biomass and productivity of Halophila decipiens, a deep water seagrass on the west Florida continental shelf. Aquatic Botany 84:110-120.

Hammerstrom, KK, W.J. Kenworthy, P.E. Whitfield, M. Merello. 2007. Response and recovery dynamics of seagrasses Thalassia testudinum and Syringodium filiforme and macroalgae in experimental motor vessel disturbances. Marine Ecology Progress Series 345:83-92.

Handley, L., Altsman, D., and R. DeMay, eds. 2007. Seagrass Status and Trends in the Northern Gulf of Mexico: 1940-2002: U.S. Geological Survey Scientific Investigations Report 2006-5287 and U.S. Environmental Protection Agency 855-R-04-003, 267 p.

Handley, L. 2010. Technical Specifications and Scope of Work/Services for Aerial Imagery Acquisition and Image Processing in Support of the MC252 NRDA Process.

Hatcher, A. I. and A. W. D. Larkum. 1982. The effects of short term exposure to Bass Strait crude oil and Corexit 8667 on benthic community metabolism in Posidonia australis beds Hook.F. dominated microcosms. Aquatic Botany 12:219-227.

Inglis, G.J. 2000. Variation in the recruitment behavior of seagrass seeds: implications for population dynamics and resource management. Pacific Conservation Biology 5:251-259.

Iverson, R. L. and H. E Bittaker. 1986. Seagrass distribution and abundance in eastern Gulf of Mexico waters. Estuarine and Coastal Shelf Science 22:577-602.

Jackson, J. B. C., J. D. Cubit, B. D. Keller, V. Batista, K. Bums, H. M. Caffey, R. L. Caldwell, S. D. Garrity, C. D. Getter, C. Gonzalez, H. M. Guzman, K. W. Kaufmann, A. H. Knap, S. C. Levings, M. J. Marshall, R. Steger, R. C. Thompson, and E. Weil. 1989. Ecological effects of a major oil spill on Panamanian coastal marine communities. Science 243:37-44.



Kenworthy, W. J. and G. W. Thayer. 1984. Production and decomposition of the roots and rhizomes of seagrasses Zostera marina and Thalassia testudinum in temperate and subtropical marine ecosystems. Bull. Mar. Sci. 35: 364-379.

Kenworthy, W.J., Fonseca, M.S., Whitfield, P.E. and K. Hammerstrom. 2000. Experimental manipulation and analysis of recovery dynamics in physically disturbed tropical seagrass communities of North America: implications for restoration and management. Proceedings of the Fourth International Seagrass Biology Workshop. Biologia Marina Mediterranea 7:385-388.

Kenworthy, W.J., Fonseca, M.S., Whitfield, P.E. and K. Hammerstrom. 2002. Analysis of seagrass recovery in experimental excavations and propeller-scar disturbances in the Florida Keys National Marine Sanctuary. Journal of Coastal Research 37:75-85.

Kirsch, K.D., K.A. Barry, M.S. Fonseca, P.E. Whitfield, S.R. Meehan, W. Judson Kenworthy, and B.E. Julius. 2005. The Mini-312 Program-an expedited damage assessment and restoration process for seagrasses in the Florida Keys National Marine Sanctuary. Journal of Coastal Research SI40:109-119.

Larkum, A.W.D., Orth, R.J., and C.M. Duarte, eds. 2006. Seagrasses: Biology, Ecology, and Conservation. Amsterdam, The Netherlands: Springer, 691 pp.

Marba, N. and C.M. Duarte. 1998. Rhizome elongation and seagrass clonal growth. Marine Ecology Progress Series 174:269-280.

McMillan, C. 1976. Experimental studies on flowering and reproduction in seagrasses. Aquatic Botany 2:87-92

McMillan, C. 1981. Seed reserves and seed germination for two seagrasses, Halodule wrightii and Syringodium filiforme, from the western Atlantic. Aquatic Botany 11:279-296.

McMillan, C. 1991. The longevity of seagrass seeds. Aquatic Botany 40:195-198.

McRoy, C. P. and S.L. Williams. 1977. Sublethal effects on seagrass photosynthesis. Final Report to NOAA Outer Continental Shelf Environmental Assessment Program, Contract# 03-5-022-56.

Michel J., Owens E.H., Zengel S., Graham A., Nixon Z., et al. 2013. Extent and Degree of Shoreline Oiling: Deepwater Horizon Oil Spill, Gulf of Mexico, USA. PLoS ONE 8(6): e65087. doi:10.1371/journal.pone.0065087.

National Oceanic and Atmospheric Administration (NOAA). 2000. Shoreline Assessment Manual. 3rd Edition. August.

National Oceanic and Atmospheric Administration (NOAA). 2010. Mississippi Canyon 252 Oil Spill Submerged Aquatic Vegetation Tier 1 Pre-Assessment Plan Pre-Impact Baseline Characterization. Prepared for the MC252 NRDA Submerged Aquatic Vegetation Technical Working Group of the Mississippi Canyon 252 Trustees. Draft Version 7.0. September 28.



National Oceanic and Atmospheric Administration (NOAA). 2011a. Mississippi Canyon 252/Deepwater Horizon Scope of Work for Emergency Restoration Project: Response Impacts to Seagrasses within Alabama, Florida, Louisiana and Mississippi Coastal waters

National Oceanic and Atmospheric Administration (NOAA). 2011b. Mississippi Canyon 252 Oil Spill Submerged Aquatic Vegetation Tier 2 Pre-Assessment Post Spill Exposure Characterization Plan. Prepared for the MC252 NRDA Submerged Aquatic Vegetation Technical Working Group of the Mississippi Canyon 252 Trustees. Final Version. November 8

National Oceanic and Atmospheric Administration (NOAA). 2011c. Tier 3: Injury Assessment plan for Submerged Aquatic Vegetation: Chandeleur Islands, Louisiana. Prepared for the MC252 NRDA Submerged Aquatic Vegetation Technical Working Group of the Mississippi Canyon 252 Trustees. Draft Version 1.0. May 10.

National Oceanic and Atmospheric Administration (NOAA). 2012a. Natural Resources Damage Assessment April 2012 Status Update for the Deepwater Horizon Oil Spill. April. Available at: http://www.gulfspillrestoration.noaa.gov/wp-content/uploads/FINAL_NRDA_StatusUpdate_April2012.pdf

National Oceanic and Atmospheric Administration (NOAA). 2012b. Analytical Quality Assurance Plan, Mississippi Canyon (Deepwater Horizon) Natural Resource Damage Assessment. Version 3.1. November 12.

National Oceanic and Atmospheric Administration (NOAA). 2013a. Assessment Plan Concerning Aerial Imagery in the Northern Gulf of Mexico.

National Oceanic and Atmospheric Administration (NOAA). 2013b. *Shoreline Assessment Manual*. 4th Edition. August.

Nadeau, R. J. and E.T. Bergquist. 1977. Effects of the March 18, 1973, oil spill near Cabo Rojo, Puerto Rico, on tropical marine communities. International Oil Spill Conference Proceedings: March 1977, Vol. 1977, No. 1, pp. 535-538.

Nixon, Z., Zengel, S., & Michel, J. (2015). Shoreline Oiling from the Deepwater Horizon Oil Spill.

Ogden, J.C., and J.C. Zieman. 1977. Ecological aspects of coral reef-seagrass bed contracts in the Caribbean. Proceedings of the 3rd international symposium on coral reefs. University of Miami 3:377-382.

Orth, Robert J., Tim J.B. Carruthers, William C. Dennison, Carlos M. Duarte, James W. Fourqurean, Kenneth L. Heck, Jr., A Randall Hughes, Gary A. Hendrick, W. Judson Kenworthy, Suzanne Olyarnik, Fred T. Short, Michelle Waycott, and Susan L. Williams. 2006. A global crisis for seagrass ecosystems. Bioscience 56:987-996.



Peirano, A., V. Damasso, M. Montefalcone, C. Morri, and C. N. Bianchi. 2005. Effects of climate, invasive species and anthropogenic impacts on the growth of the seagrass Posidonia oceanica (L.) Delilein Liguria (NW Mediterranean Sea). Marine Pollution Bulletin 50:817-822.

Poirrier, M.A., and C.D. Franze. 2001 Seagrass restoration in the Chandeleur Islands after Hurricane Georges. Final report: New Orleans, La., University of New Orleans Department of Biological Sciences, 46 p.

Poirrier, M.A. and L.R. Handley. 2007. Chandeleur Islands. In: Handley, L., Altsman, D., and DeMay, R. (eds.) Seagrass Status and Trends in the Northern Gulf of Mexico: 1940-2002: U.S. Geological Survey Scientific Investigations Report 2006-5287 and U.S. Environmental Protection Agency 855-R-04-003 pp 63-72.

Ralph, P. J. and M. D. Burchett. 1998. Impact of petrochemicals on the photosynthesis of Halophila ovalis using chlorophyll fluorescence. Marine Pollution Bulletin 36:429-436.

Rasheed, M.A., K.R. Dew, S.P. Kerville, L.J. McKenzie and R.G. Coles. 2006. Seagrass distribution, community structure and productivity for Orman Reefs, Torres Strait – March and November 2004. DPI Information Series QI06088 (DPI, Cairns), 38 pp.

Romero, J., Lee, K-S, Perez, M, Mateo, M.A., and T. Alcoverro. 2006. Nutrient dynamics in seagrass ecosystems. In: Larkum, A.W.D., Orth, R. J., and C. M. Duarte (eds.) Seagrasses: Biology, Ecology and Conservation. pp 227-254.

Rouhani, S., M.C. Baker, M. Zhang, J. Oehrig, I.J. Zelo and S.D. Emsbo-Mattingly. 2015. Nearshore Exposure to DWH Oil: A Conceptual Model.

Sandulli, R., C. N. Bianchi, S. Cocito, C. Morri, A. Peirano, and S. Sgorbini. 1998. An experience of 'basilage' in monitoring the effects of the Haven oil spill on some Ligurian Posidonia oceanic meadows. Oebalia 24:3-15.

Scarlett, A., T. S. Galloway, M. Canty, E. L. Smith, J. Nilsson, and S. J. Rowland. 2005. Comparative toxicity of two oil dispersants, Superdispersant-25 and Corexit 9527, to a range of coastal species. Environmental Toxicology and Chemistry 24:1219-1227.

Short, F.T., D.M. Burdick and J.E. Kaldy. 1995. Mesocosm experiments quantify the effects of eutrophication on eelgrass, Zostera marina. Limnology and Oceanography 40:740-749.

Short, F.T., Carruthers, T.J.B., Dennison, W.C., Waycott, M., 2007. Global seagrass distribution and diversity: a bioregional model. Journal of Experimental Marine Biology and Ecology 350, 3–20.

Thorhaug, A. and J. Marcus. 1987. Oil spill cleanup: the effect of three dispersants on three subtropical/tropical seagrasses. Marine Pollution Bulletin 18:124-126.

Tomlinson, P.B. 1974. Vegetative morphology and meristem dependence. The foundation of productivity in seagrasses. Aquaculture 4:107-130.



U.S. Coast Guard (USCG). 2011. BP Deepwater Horizon Spill: Incident Specific Preparedness Review (ISPR). January 2011. Washington, DC: U.S. Department of Homeland Security.

U.S. District Court. 2014. In re: Oil Spill by the Oil Rig "Deepwater Horizon" in the Gulf of Mexico, on April 20, 2010, No. MDL 2179, Section 7 (Revised September 9, 2014) ("Findings of Fact and Conclusions of Law: Phase One Trial"), Figure 1. United States District Court for the Eastern District of Louisiana.

U.S. District Court. 2015. In re: Oil Spill by the Oil Rig "Deepwater Horizon" in the Gulf of Mexico, on April 20, 2010, No. MDL 2179, 2015 WL 225421 (La. E.D. Jan. 15, 2015) ("Findings of Fact and Conclusions of Law: Phase Two Trial"). United States District Court for the Eastern District of Louisiana.

Uhrin, A.V., Kenworthy, W.J., and Fonseca, M.S. 2011. Understanding uncertainty in seagrass injury recovery: an information-theoretic approach. Ecological Applications 21:1365–1379.

Wang, Z.; Stout, S.; Fingas, M. 2006. Forensic Fingerprinting of Biomarkers for Oil Spill Characterization and Source Identification. *Environmental Forensics*. Vol. 7. pp 105-146.

Whitfield, P.E., Kenworthy, W.J., Fonseca, M.S., Hammerstrom, K. 2002. The Role of a Hurricane in expansion of disturbances initiated by motor vessels on subtropical seagrass banks. Journal of Coastal Research 37:86-99.

Whitfield, P.E., W. Judson Kenworthy, Michael J. Durako, Kamille K. Hammerstrom, and Manuel F. Merello. 2004. Recruitment of Thalassia testudinum Seedlings into Physically Disturbed Seagrass Beds. Marine Ecology Progress Series 267:121-131.

Williams, S.L. 1988. Disturbance and recovery of a deep-water Caribbean seagrass bed. Marine Ecology Progress Series 42:63-71.

Williams, S.L. 1990. Experimental studies of Caribbean seagrass bed development. Ecological Monographs 60:449-469.

Zelo, I and K. Benson, 2011. Submerged Oil Characterization Across Multiple Habitats for Assessment of Persistent Exposures in Nearshore Sediments, Deepwater Horizon Oil Spill (DWHOS).

Zieman, J.C. 1982. The ecology of the seagrasses of south Florida: a community profile. U.S. Fish and Wildlife Service, Office of Biological Services, Washington, D.C. FWS/OBS-82/25.

Zieman, J. C., R. J. Orth, R. C. Phillips, G. W. Thayer, and A. Thorhaug. 1984. The effects of oil on seagrass ecosystems. Pages 37-64 in J. Cairns and A. L. Buikema, editors. Restoration of habitats impacted by oil spills.

Zieman, J.C. and R. T. Zieman. 1989. The ecology of seagrass meadows of the west coast of Florida: a community profile. Biological Report 85(7.25). U.S. Fish and Wildlife Service, Washington, D.C. 155 pp.





Submerged Aquatic Vegetation Exposure to Deepwater Horizon Spill

Tables

Table 1. Illustration of the SAV TWG assessment timeline and the activities and locations associated with each of the three Tiers.

Proximity to Spill	May/June 2010	July/August/September 2010			June 2011	Aug	2011-May 2012	
	Base Line – Pre-oil	Exposure – Post-oil			Injury Analysis			
	Tier 1	Tier 2	Phase 1	Phase 2	Phase 3	Tier 3	Phase 1	Phase 2
Closest	Chandeleur Islands , LA	Chandeleur Islands , LA	X	X	X	Chandeleur Islands, LA	X	х
	Hom Island, MS	Horn Island, MS	X	X				X*
	Petit Bois Island, MS	Petite Bois Island, MS	Х	X		1		X*
	Cat Island, MS	Cat Island, MS	X					
	Ship Island, MS	Ship Island, MS	X			1		
	Grand Bay, AL	Grand Bay , AL	X					
	Dauphin Is, AL	Dauphin Islands, AL	X					
	Perdido Pass (Robinson Is), AL	Perdido Pass (Robinson Is), AL	X	X				
	Pensacola Bay (Big Lagoon), FL	Pensacola Bay (Big Lagoon), FL	X	X				
	Pensacola Bay opening, FL							
	Mobile Bay, AL							
7	Gulf Islands National Seashore (Big Lagoon) FL							
	Gulf Islands National Seashore Santa Rosa Island FL							
4	Big Lagoon (outside NPS) FL							
	Choctawhatchee Bay, FL							
Farthest	St. Andrew Bay ,FL							
	St. Joseph Bay ,FL							
LOOP	Dry Tortugas National Park ,FL							
CURRENT	Florida Keys, FL							
	AERIAL IMAGERY							\rightarrow
	19 Sites		8 sites	5 sites	1 site	1 site	1 site	3 sites
	All areas that were in threat of oiling by observing NOAA trajectories. Aerial Imagery flown from LA to FL pan handle starting in May 2010	Phase 1: Documentation of oil on shorelines triggered use of submerged oil sentinels; Phase 2: Confirmation of oil within the SAV beds by oil sentinels and triggered rapid assessment; and Phase 3: Confirmation of exposure within SAV beds by analytical collected during rapid assessment.				Phase 1: Increased sampl document physical chang fauna and chemistry sam determine duration of ex on southern and central C	ges to beds and ples to posure. Focus	Phase 2: Aerial imagery/change analysis on Chandeleurs.* Petite Bois and Horn added.



Table 2. List of Total PAH (tPAH) Compounds

tPAH Compounds								
Acenaphthene C1-Naphthalenes		C4-Dibenzothiophenes						
Acenaphthylene	C1-Naphthobenzothiophenes	C4-Fluoranthenes/pyrenes						
Anthracene	C1-Phenanthrenes/anthracenes	C4-Naphthalenes						
Benzo(a)anthracene	C2-Chrysenes	C4-Naphthobenzothiophenes						
Benzo(a)fluoranthene	C2-Dibenzothiophenes	C4-Phenanthrenes/anthracenes						
Benzo(a)pyrene	C2-Fluoranthenes/pyrenes	Chrysene						
Benzo(b)fluoranthene	C2-Fluorenes	Chrysene+Triphenylene						
Benzo(b)fluorine	C2-Naphthalenes	Dibenzo(a,h)anthracene						
Benzo(b+j)fluoranthene	C2-Naphthobenzothiophenes	Dibenzo(a,h+a,c)anthracene						
Benzo(e)pyrene	C2-Phenanthrenes/anthracenes	Dibenzofuran						
Benzo(g,h,i)perylene	C3-Chrysenes	Dibenzothiophene						
Benzo(j+k)fluoranthene	C3-Dibenzothiophenes	Fluoranthene						
Benzo(k)fluoranthene	C3-Fluoranthenes/pyrenes	Fluorene						
Biphenyl	C3-Fluorenes	Indeno(1,2,3-c,d)pyrene						
C1-Chrysenes	C3-Naphthalenes	Naphthalene						
C1-Dibenzothiophenes	C3-Naphthobenzothiophenes	Naphthobenzothiophene						
C1-Fluoranthenes/pyrenes	C3-Phenanthrenes/anthracenes	Phenanthrene						
C1-Fluorenes	C4-Chrysenes	Pyrene						



Table 3. Dates of acquisition and number of frames used to map seagrasses in the Chandeleur Islands, LA.

Date	# frames for mapping
October 8,	
2010	14 frames
October 24,	
2011	17 Frames
September	
23, 2012	10 frames *

^{*} Five smaller subset areas were mapped for 2012 rather than the whole extent of the Chandeleurs.



Table 4. Tiers 1, 2 and 3 tPAH concentrations for sediments in the Chandeleur Islands. Also shown are average ratios between Tiers, % reduction from Tier 3 to Tier 2 and the ratio of Petrogenic (Pet) to Pyrogenic (Pyr) PAH.

	Average (ppb)	e Concenti	ration	Average	% Red.	
Measure	Tier 1	Tier 2	Tier 3	Tier 2/1	Tier 3/1	Tier 3/2
tPAH	40.84	325.01	69.63	7.96	1.71	21%
PetPAH	27.02	297.57	49.99	11.01	1.85	17%
PyrPAH	13.64	27.25	18.78	2.00	1.38	69%
Pet/Pyr	1.98	10.92	2.66			



Table 5. Tiers 1, 2, and 3 tPAH concentrations for SAV plant, external material and animal tissue tPAH concentrations in the Chandeleur Islands. Also shown are average ratios for Tiers 2 and 3 compared to Tier 1 and the ratio of Petrogenic (Pet) to Pyrogenic (Pyr) PAH.

Average Concentration (ppb) for Plant Tissue, External Material and Whole Body Invertebrates								Average	Ratios C	ompared	to Tier 1				
	Leaves & Stems		ıs	Ext	External Material Whole Body		Leav Ste		Exte Mat		Whole	Body			
Measure	Tier 1	Tier 2	Tier 3	Tier 1	Tier 2	Tier 3	Tier 1	Tier 2	Tier 3	Tier 2	Tier 3	Tier 2	Tier 3	Tier 2	Tier 3
tPAH	3.36	44.23	10.04	67,192	174,020	49,237	5.58	2,290.98	6.96	13.15	2.99	2.59	0.73	410.7	1.25
PetPAH	1.28	38.03	8.16	28,710	139,842	38,532	0.17	2,132.03	4.25	29.72	6.38	4.87	1.34	12,589	25.12
PyrPAH	1.85	6.16	1.88	38,434	34,075	10,946	4.23	158.38	2.71	3.32	1.02	0.89	0.28	37.5	0.64
Pet/Pyr	0.69	6.18	4.34	0.75	4.10	3.52	0.04	13.46	1.57						



Table 6. Characterization of the seagrass species community composition showing the number of stations sampled with each species and mean water depths at the five Tier 2 sampling stations. The Chandeleur Islands, La were stratified into categories of oil exposure as; heavily oiled (H), moderately oiled (M), and no oil (N).

LOCATION	# STATIONS	MEAN DEPTH (m)	Thalassia testudinum	Halodule wrightii	Syringodium filiforme	Ruppia maritima
Big Lagoon (Fl)	20	0.77	18	19	0	0
Robinson Island (Al)	10	0.74	0	10	0	0
Horn Island (MS)	14	1.25	0	14	0	0
Petit Bois Island (MS)	13	1.35	0	11	4	0
Chandeleur Islands (La) H	10	0.47	0	7	0	3
Chandeleur Islands (La) M	10	0.42	2	6	0	3
Chandeleur Islands (La) N	4	1.1	0	4	0	0



Table 7. Changes (loss, gains, or no change) in SAV areal coverage (acres) in the Chandeleur Islands, LA between 2010 and 2011.

Change 2010 to 2011							
2010	2011	Acres					
SAV	SAV	1902.54	No Change				
SAV	NOT SAV	482.81	Loss				
NOT SAV	SAV	710.71	Gain				
NOT SAV	NOT SAV	17496.59	No Change				
		20592.65	Total Acres				
		482.81	Total Loss				



Table 8. Change (losses and gains) and no change in SAV areal coverage (acres) in Core Area 1 in the Chandeleur Islands, LA between 2010 and 2012. Delayed loss and persistent loss are calculated separately and summed to compute total loss.

Core Area 1									
2010	2011	2012 A	res L	oss Classes					
SAV	SAV	SAV	25.37						
SAV	SAV	NOT SAV	41.17	Delayed Loss					
SAV	NOT SAV	SAV	0.99						
SAV	NOT SAV	NOT SAV	13.73	Persistent Loss					
NOT SAV	SAV	SAV	7.31						
NOT SAV	SAV	NOT SAV	19.99						
NOT SAV	NOT SAV	SAV	3.69						
NOT SAV	NOT SAV	NOT SAV	53.69						
			54.91	Total Loss					
			165.96	Total Acres					

Table 9. Change (losses and gains) and no change in SAV areal coverage (acres) in Core Area 2 in the Chandeleur Islands, LA between 2010 and 2012. Delayed loss and persistent loss are calculated separately and summed to compute total loss.

Core Area 2									
2010	2011	2012	Acres	Loss Classes					
SAV	SAV	SAV	176.44						
SAV	SAV	NOT SAV	28.68	Delayed Loss					
SAV	NOT SAV	SAV	21.98						
SAV	NOT SAV	NOT SAV	35.76	Persistent Loss					
NOT SAV	SAV	SAV	13.28						
NOT SAV	SAV	NOT SAV	9.84						
NOT SAV	NOT SAV	SAV	22.59						
NOT SAV	NOT SAV	NOT SAV	156.41						
			64.44	Total Loss					
			464.99	Total Acres					



Table 10. Change (losses and gains) and no change in SAV areal coverage (acres) in Core Area 3 in the Chandeleur Islands, LA between 2010 and 2012. Delayed loss and persistent loss are calculated separately and summed to compute total loss.

Core Area 3									
2010	2011	2012	Acres	Loss Classes					
SAV	SAV	SAV	149.39						
SAV	SAV	NOT SAV	19.26	Delayed Loss					
SAV	NOT SAV	SAV	12.84						
SAV	NOT SAV	NOT SAV	17.35	Persistent Loss					
NOT SAV	SAV	SAV	17.81						
NOT SAV	SAV	NOT SAV	16.05						
NOT SAV	NOT SAV	SAV	21.17						
NOT SAV	NOT SAV	NOT SAV	171.78						
			36.61	Total Loss					
			425.64	Total Acres					

Table 11. Change (losses and gains) and no change in SAV areal coverage (acres) in Core Area 4 in the Chandeleur Islands, LA between 2010 and 2012. Delayed loss and persistent loss are calculated separately and summed to compute total loss.

Core Area 4									
2010	2011	2012	Acres	Loss Classes					
SAV	SAV	SAV	159.12						
SAV	SAV	NOT SAV	32.74	Delayed Loss					
SAV	NOT SAV	SAV	9.10						
SAV	NOT SAV	NOT SAV	14.80	Persistent Loss					
NOT SAV	SAV	SAV	11.66						
NOT SAV	SAV	NOT SAV	10.61						
NOT SAV	NOT SAV	SAV	9.33						
NOT SAV	NOT SAV	NOT SAV	123.27						
			47.55	Total Loss					
			370.62	Total Acres					



Table 12. Change (losses and gains) and no change in SAV areal coverage (acres) in Core Area 5 in the Chandeleur Islands, LA between 2010 and 2012. Delayed loss and persistent loss are calculated separately and summed to compute total loss.

Core Area 5								
2010	2011	2012 A	cres L	oss Classes				
SAV	SAV	SAV	277.88					
SAV	SAV	NOT SAV	37.64	Delayed Loss				
SAV	NOT SAV	SAV	14.80					
SAV	NOT SAV	NOT SAV	30.06	Persistent Loss				
NOT SAV	SAV	SAV	35.79					
NOT SAV	SAV	NOT SAV	23.60					
NOT SAV	NOT SAV	SAV	24.34					
NOT SAV	NOT SAV	NOT SAV	210.31					
		67.70	Total Loss					
			654.40	Total Acres				

Table 13. Summary of SAV Persistent and Delayed Losses (acres) for each of the five Core Areas and the total loss.

Area	Persistent Loss (ac)	Delayed Loss (ac)	Total Loss (ac)
Core Area 1	13.73	41.17	54.91
Core Area 2	35.76	28.68	64.44
Core Area 3	17.35	19.26	36.61
Core Area 4	14.8	32.74	47.55
Core Area 5	30.06	37.64	67.7
Total	111.7	159.5	271.2



Table 14. Model based estimates of years to recovery for seagrass in persistent loss polygons. Acreages are binned by years to recovery and the number of polygons in each of the categories is shown.

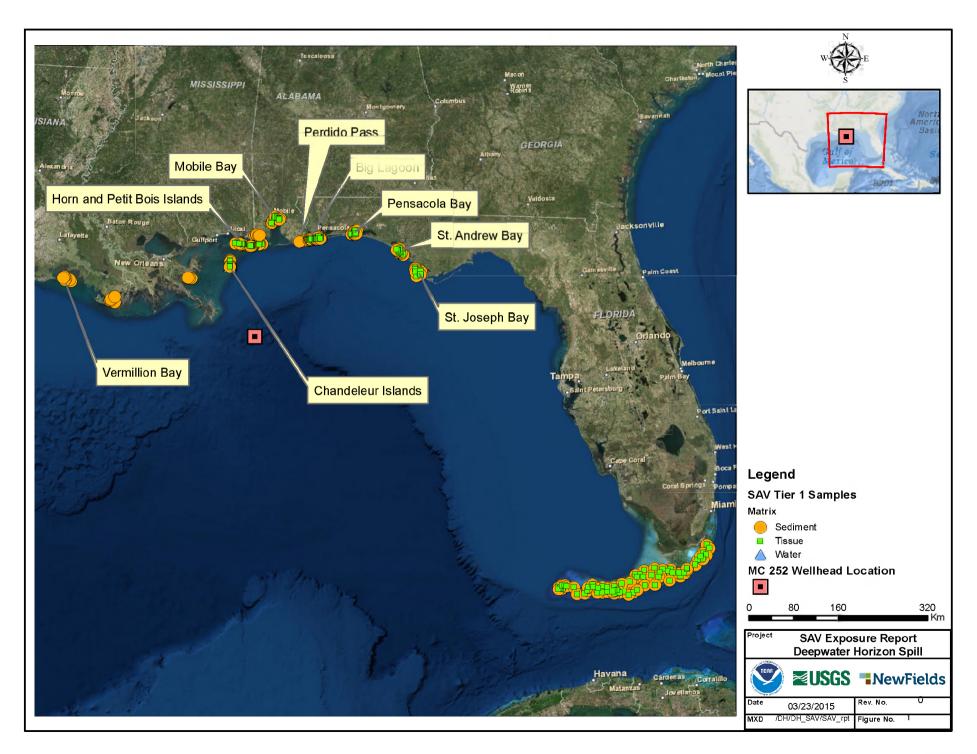
	Number of Polygons	Total Acres	Years to Recovery
	1	2.18	26
	1	1.91	23
	1	1.89	23
	1	1.53	18
	1	1.29	15
	1	1.16	14
	1	0.70	8
	1	0.59	7
	3	1.58	6 - <7
	3	1.31	5 - <6
	6	2.25	4 - <5
	9	2.53	3 - <4
	19	3.69	2 - <3
	83	11.24	1 - <2
Total	131	33.85	Not Applicable

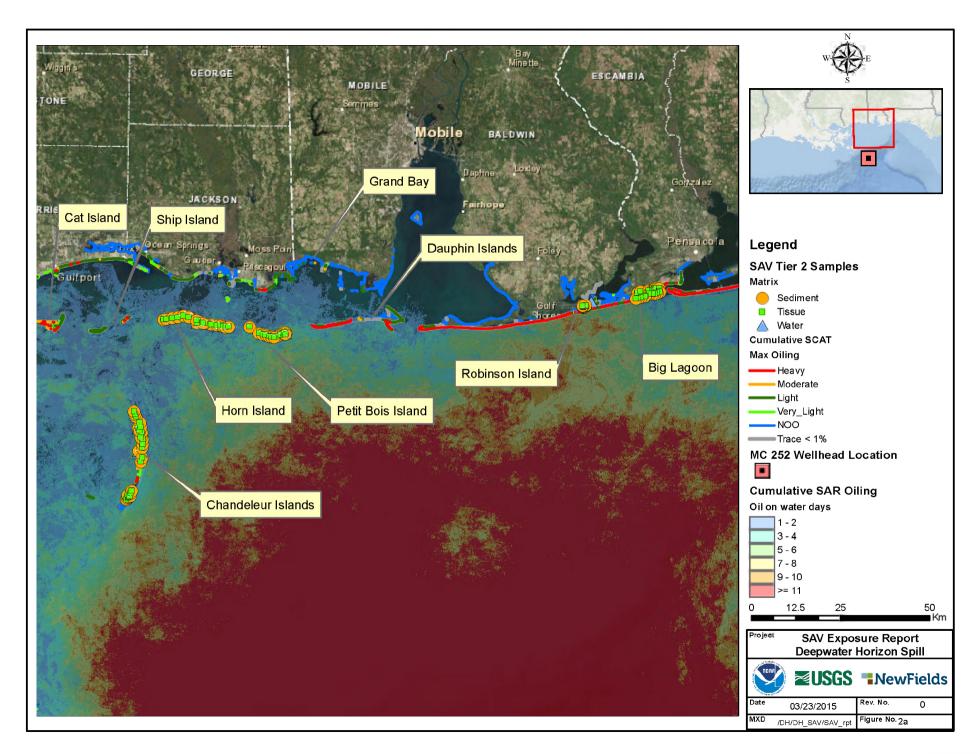


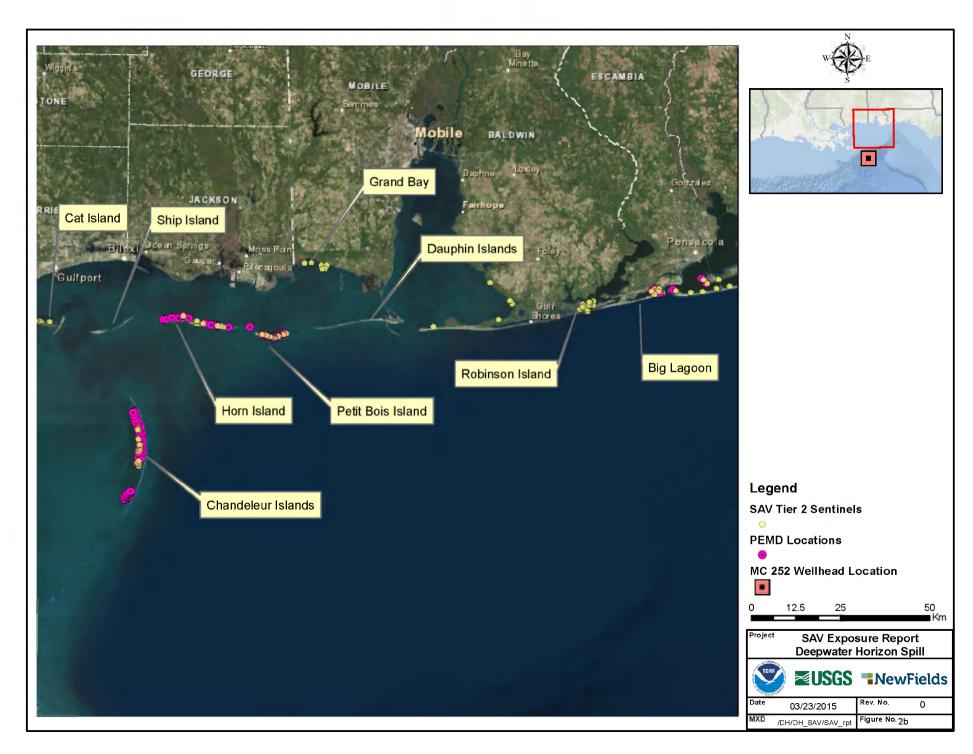


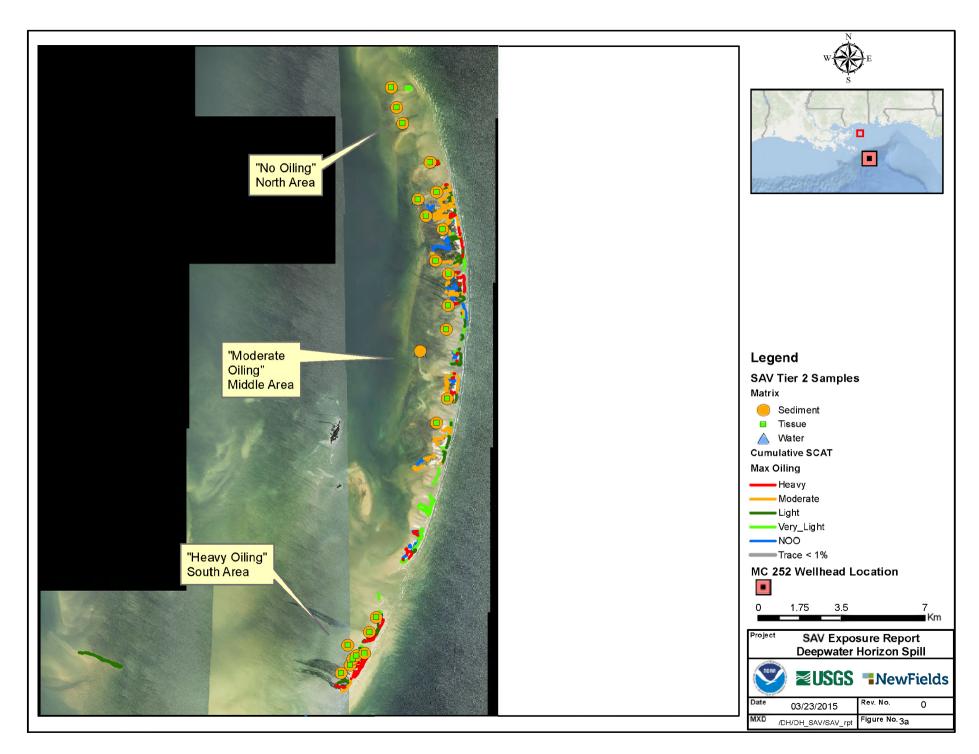
Submerged Aquatic Vegetation Exposure to Deepwater Horizon Spill

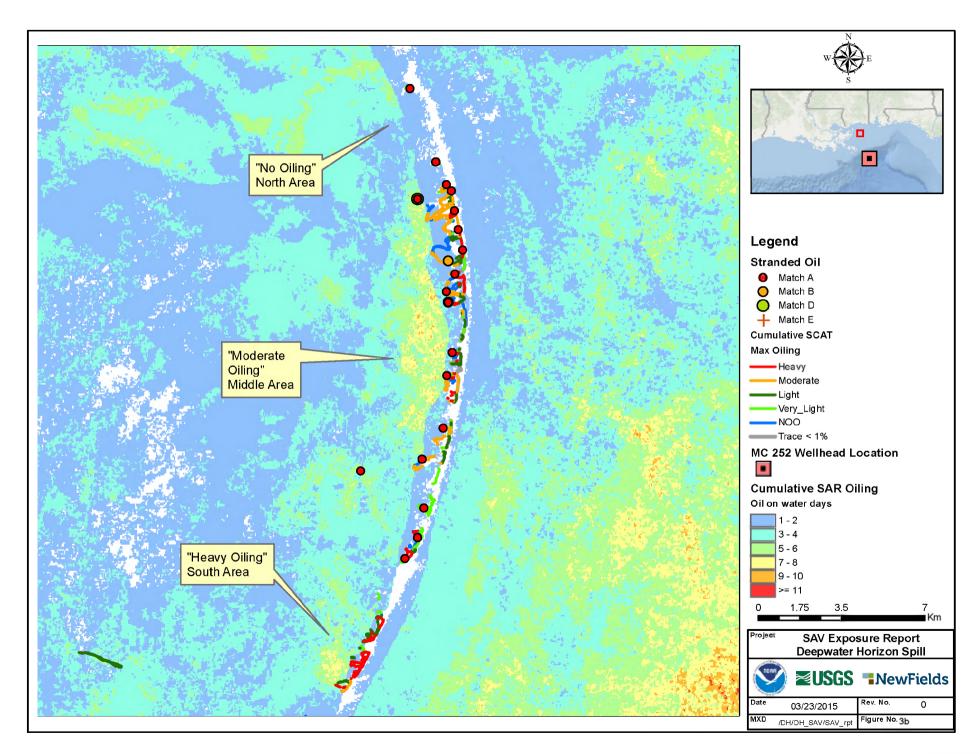
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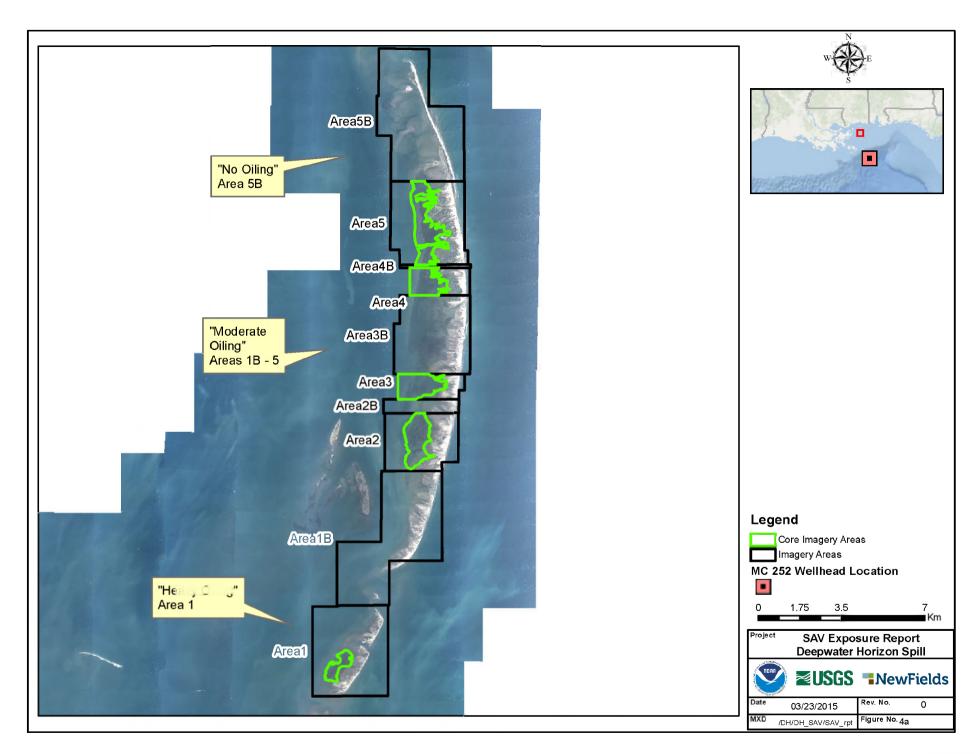


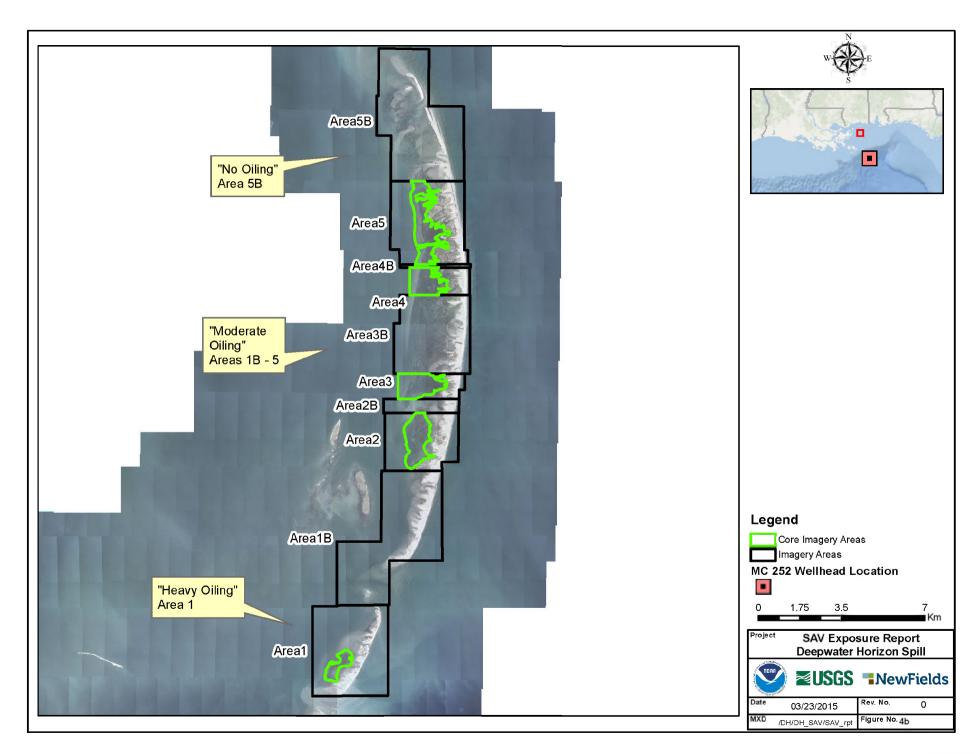


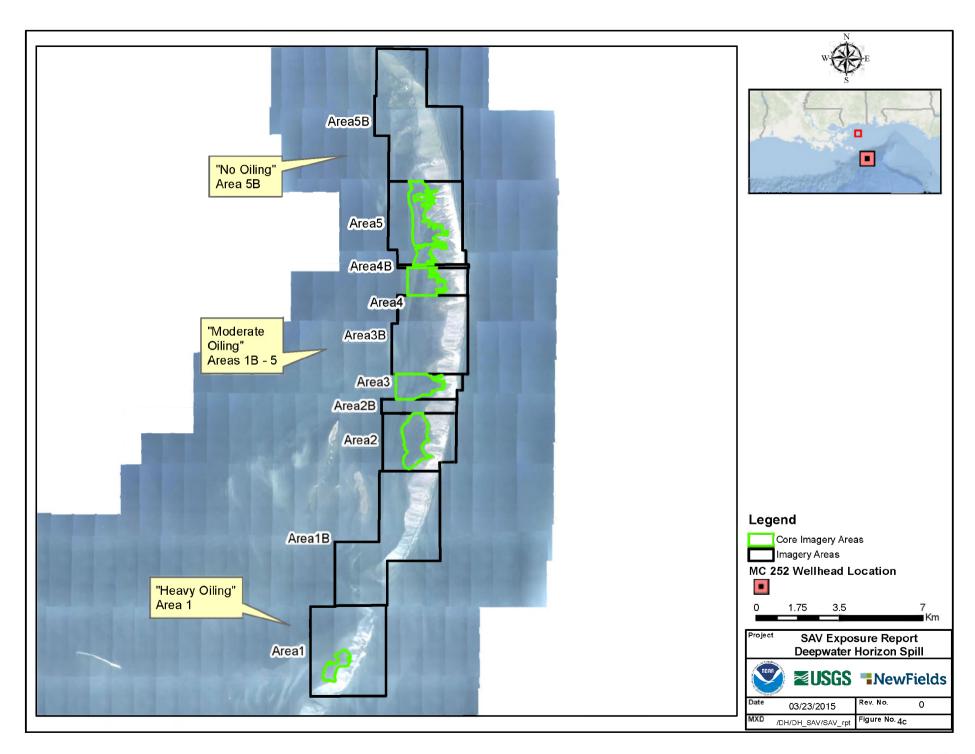


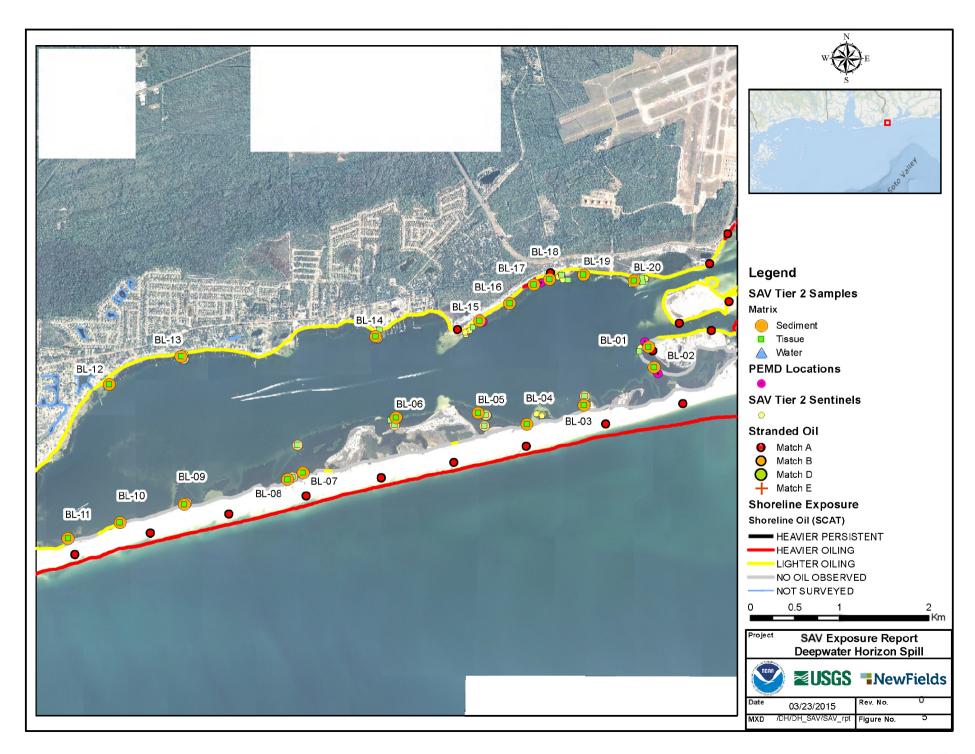


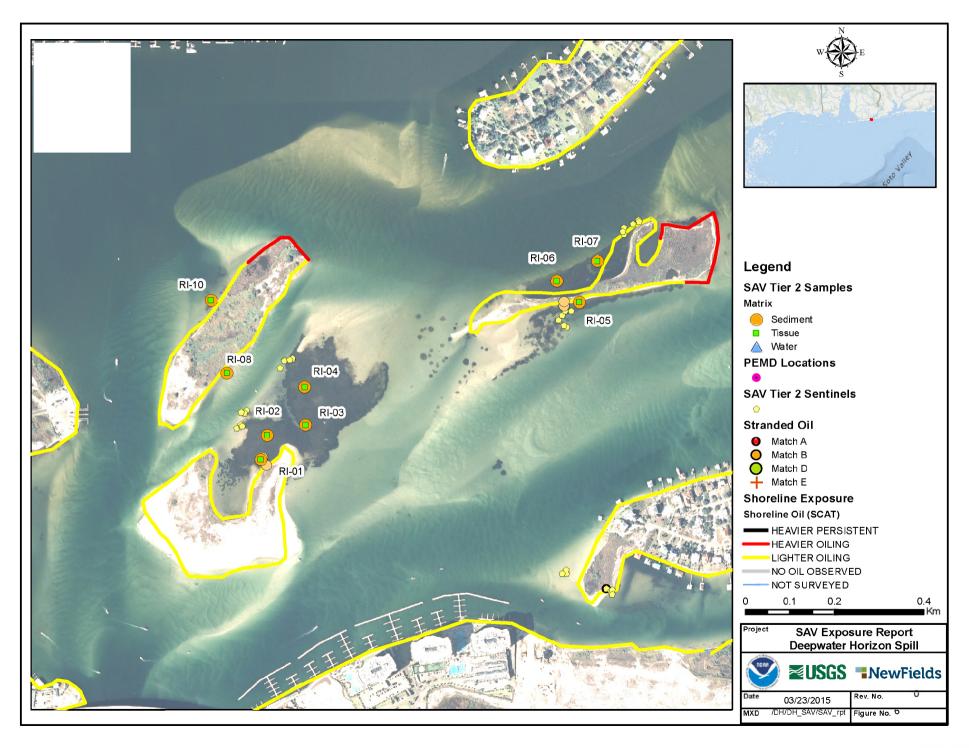


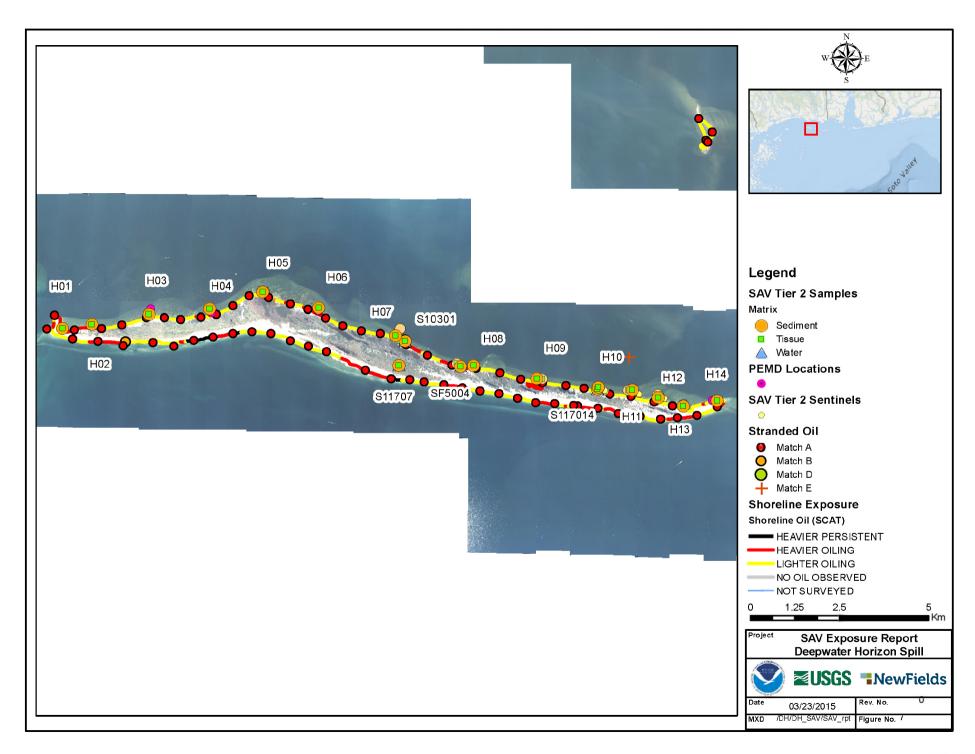


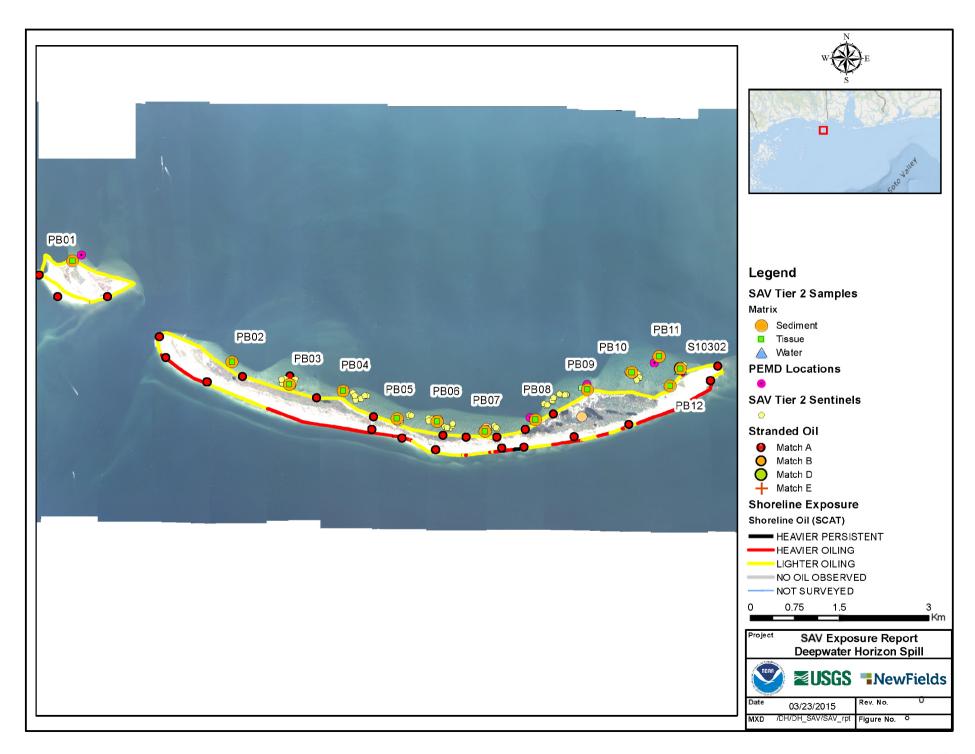


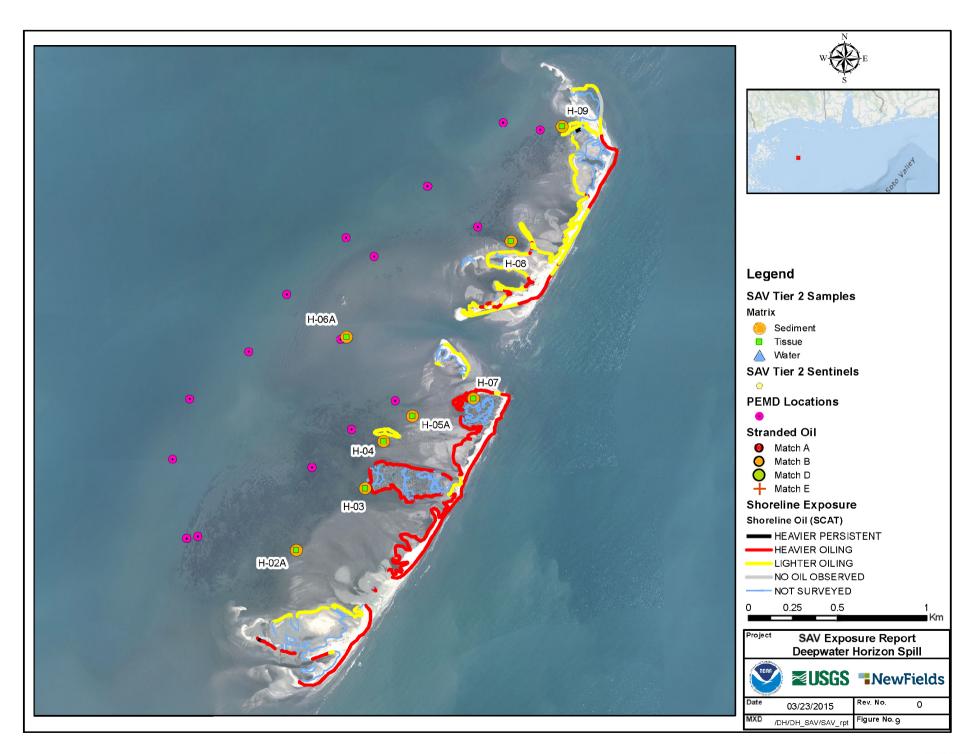


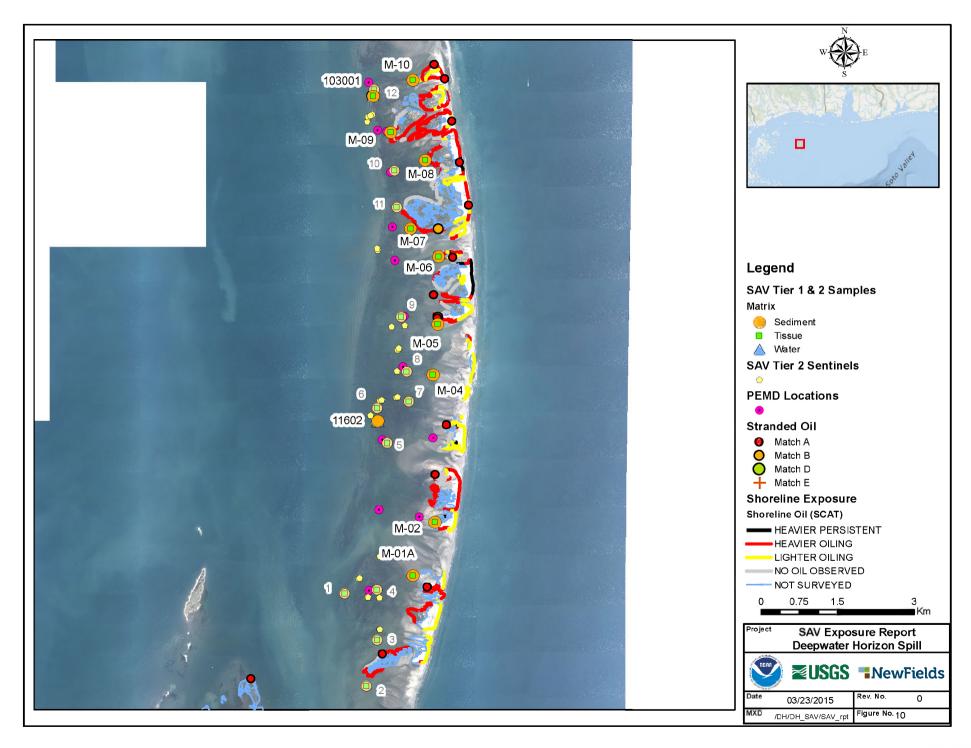


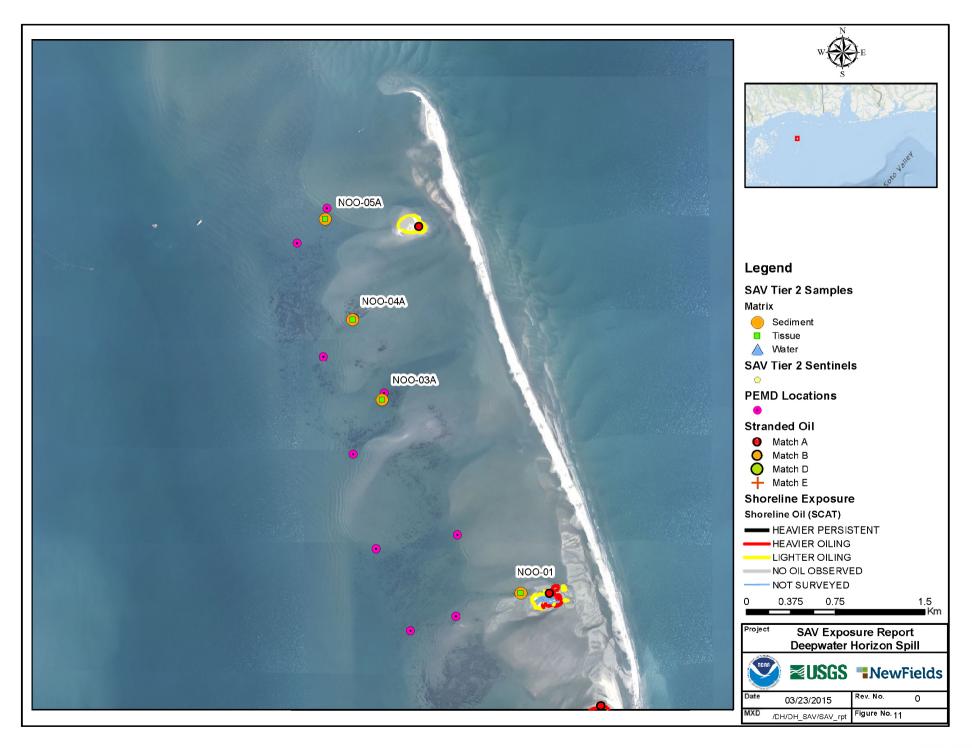












Thalassia testudinum

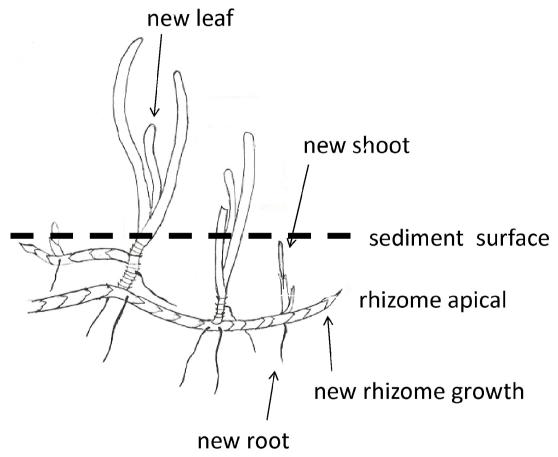


Figure 12. Illustration of a clonal fragment of Thalassia testudinum with intact shoots showing new leaf, shoot, rhizome and root growth and the rhizome apical meristem.

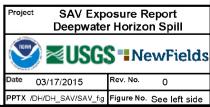
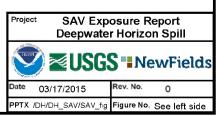
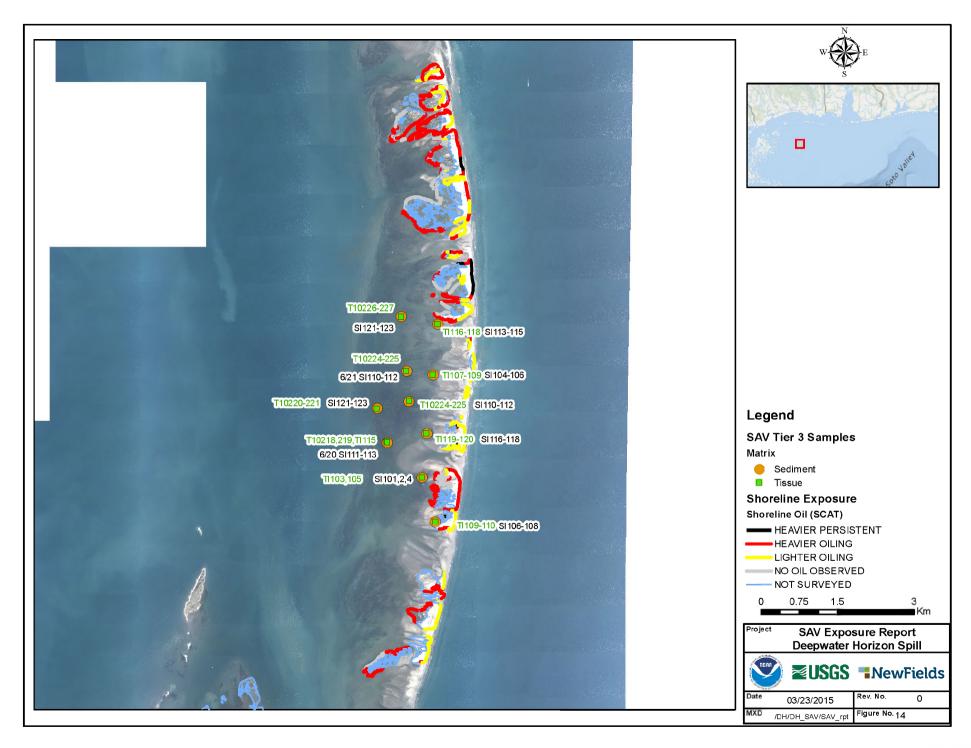
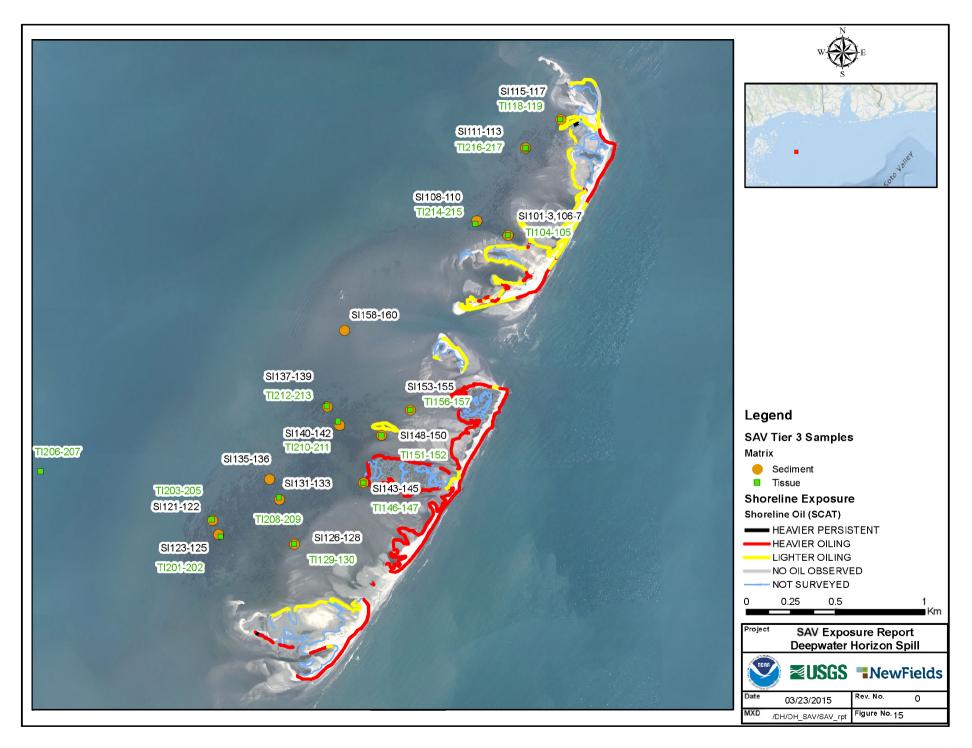


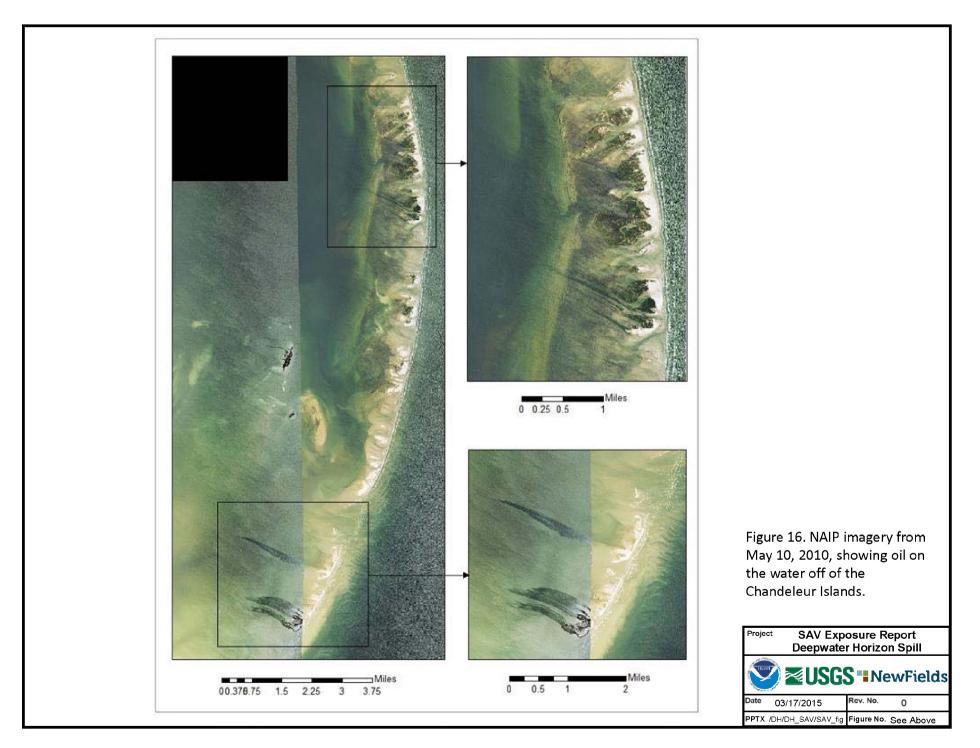


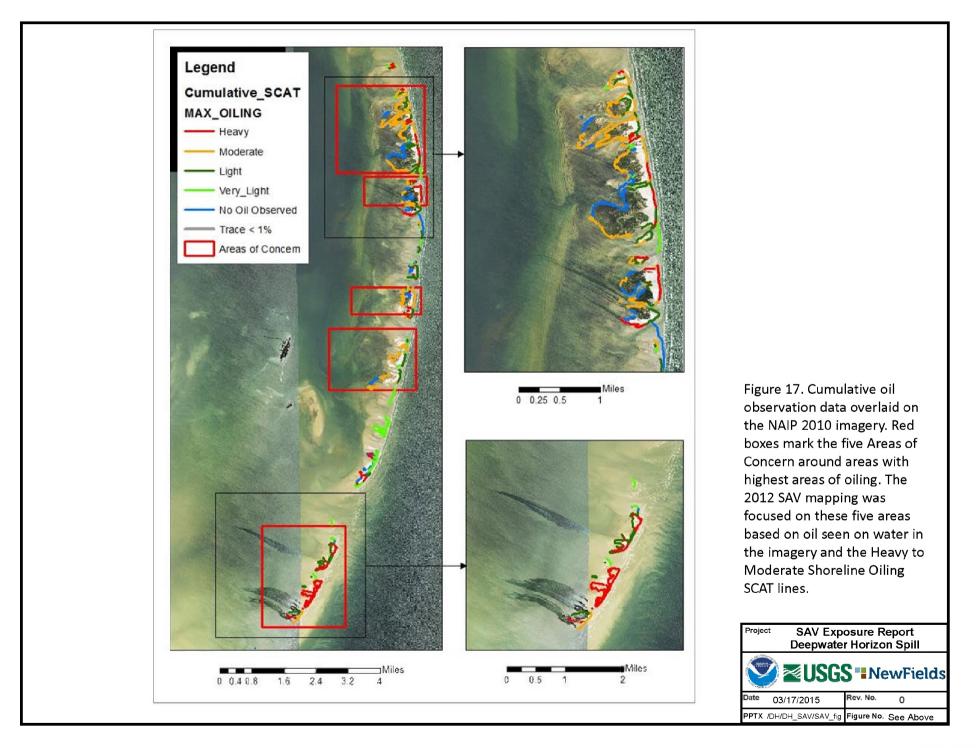
Figure 13. Photograph of a Braun-Blanquet quadrat used to visually assess the species composition and cover of seagrasses.

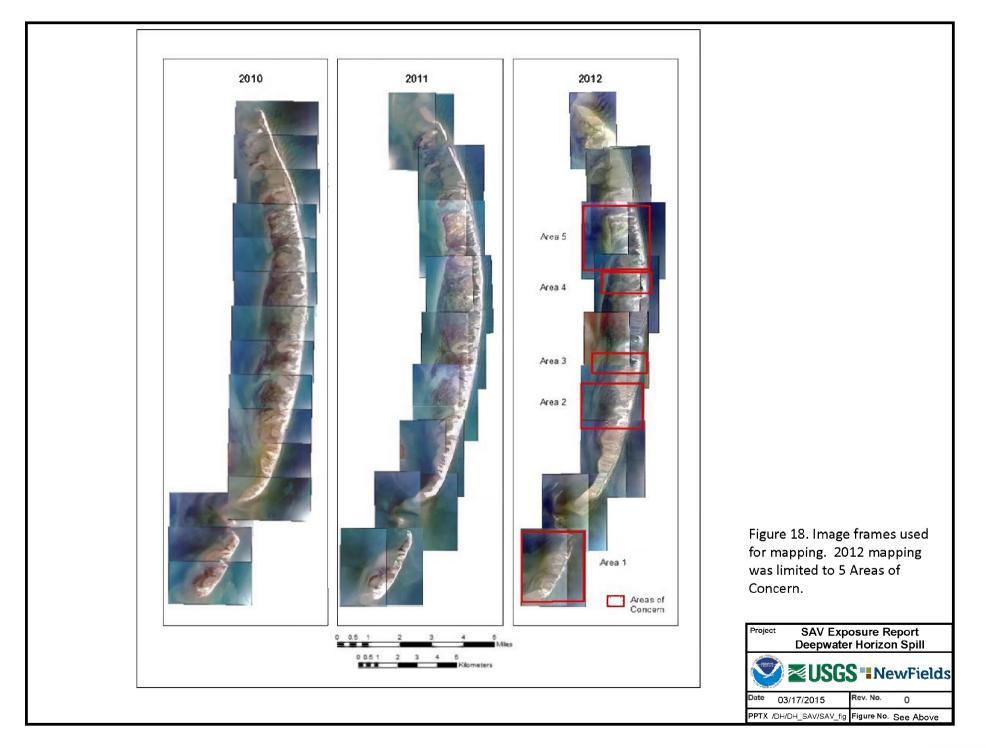












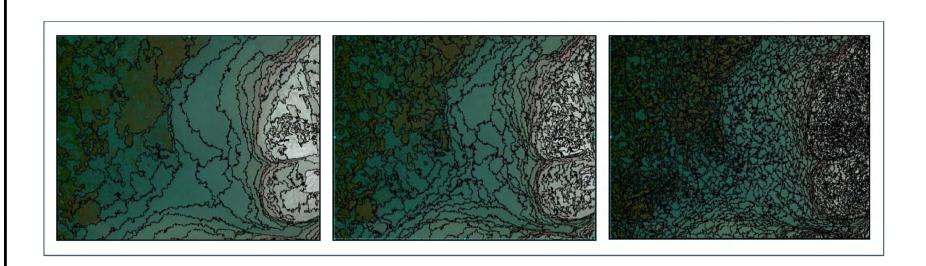
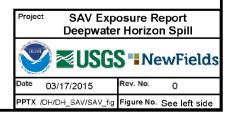
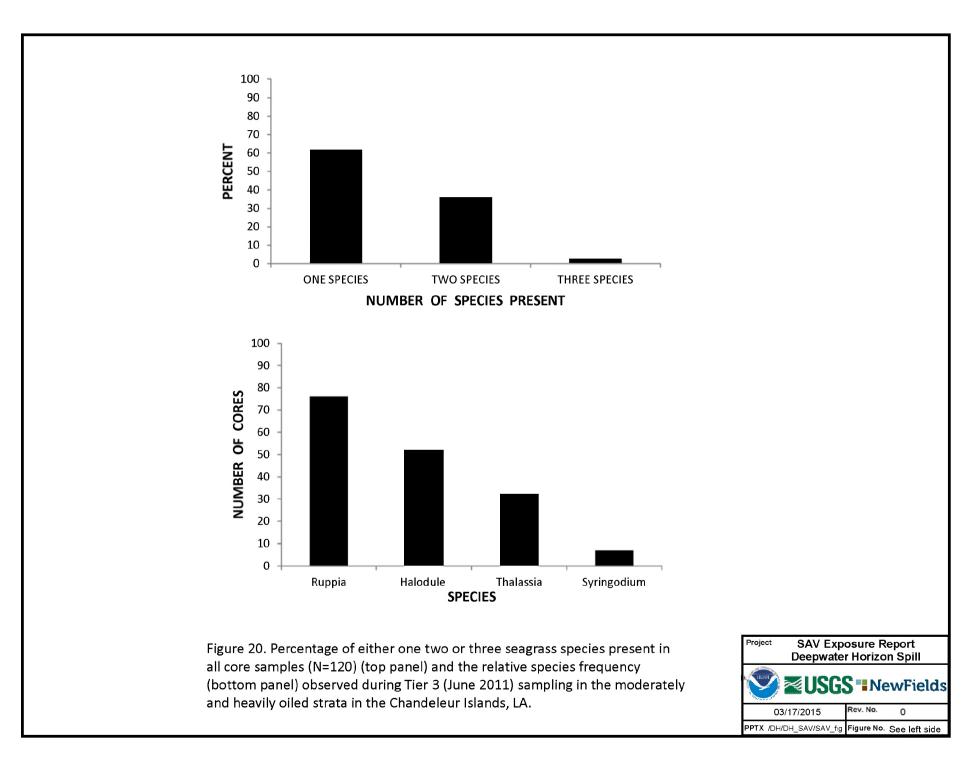
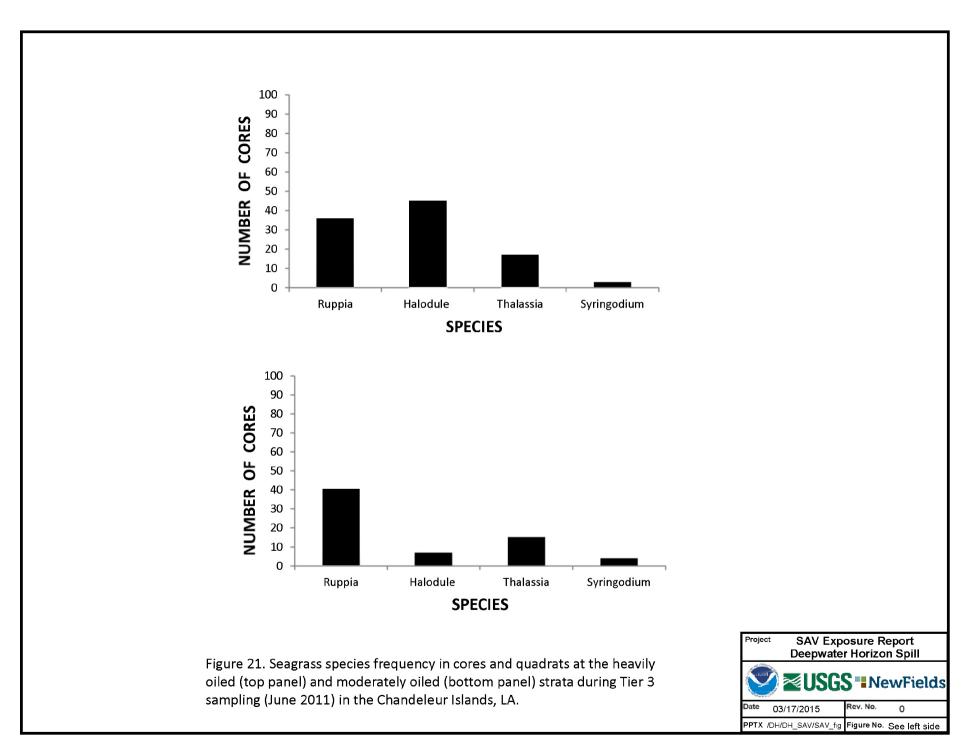
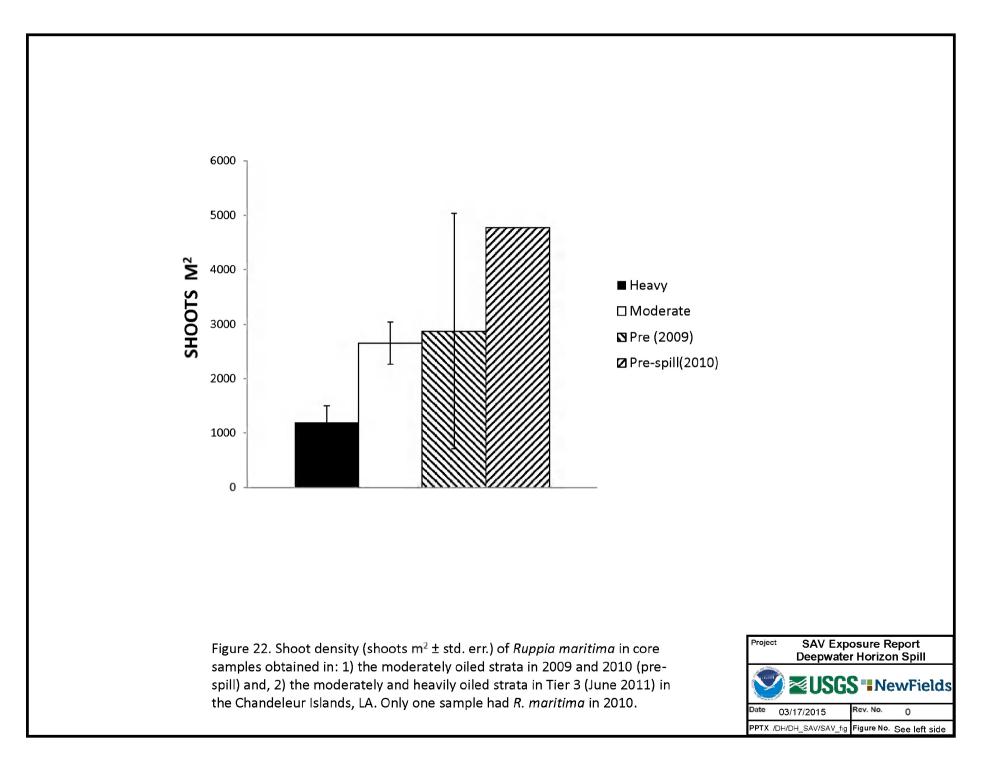


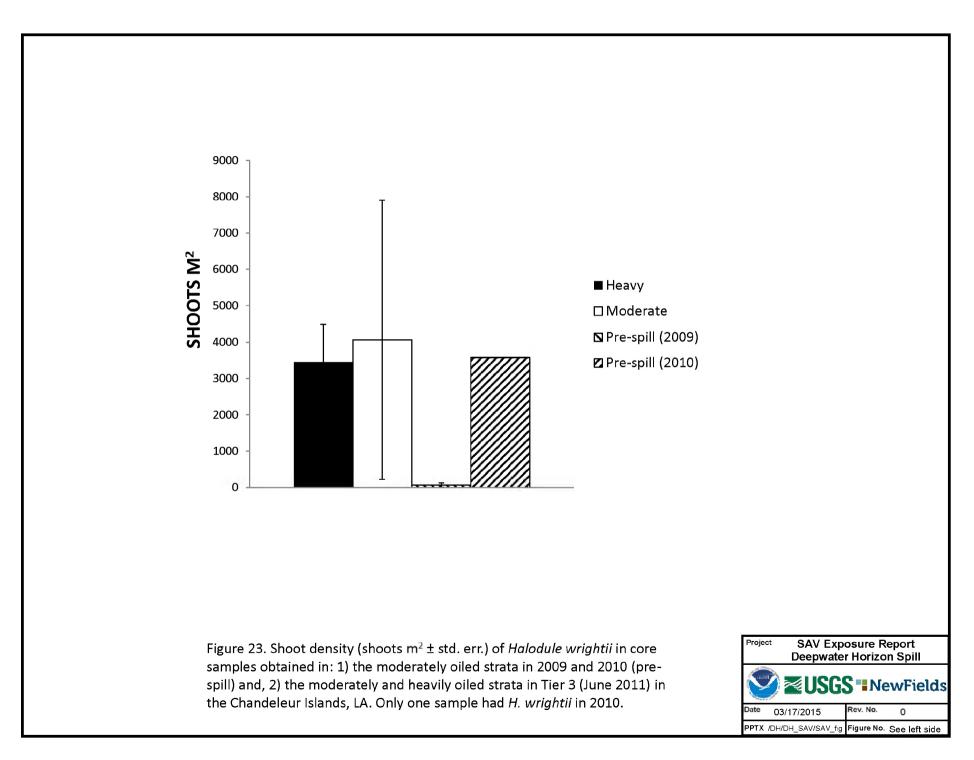
Figure 19. Nested image objects generated by the image segmentation.

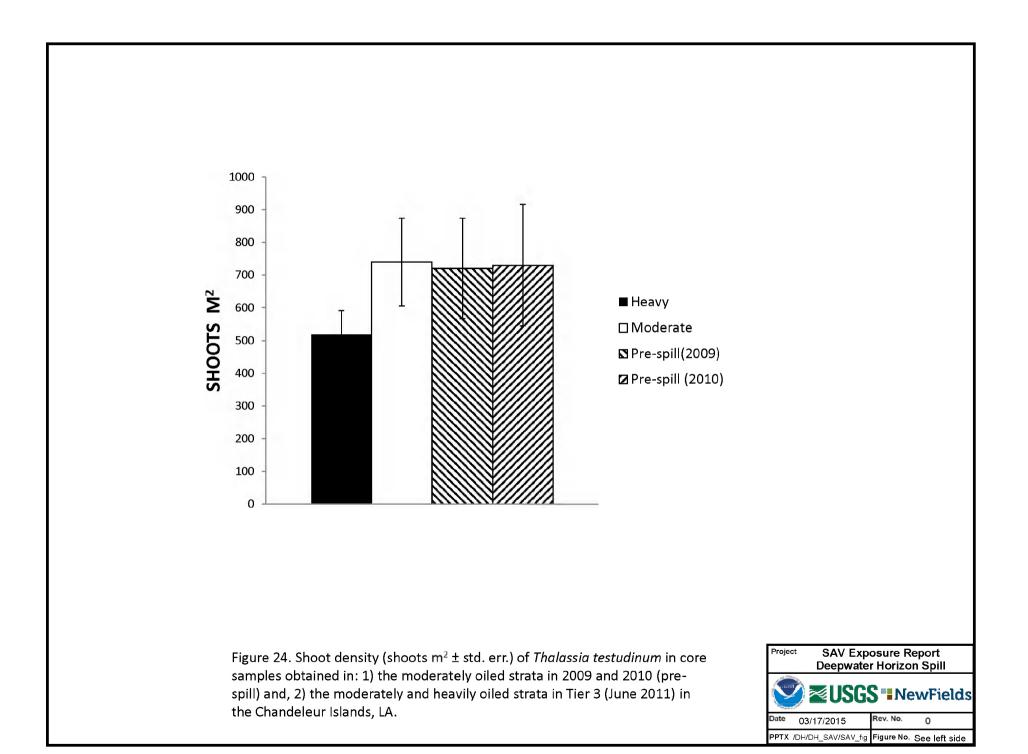


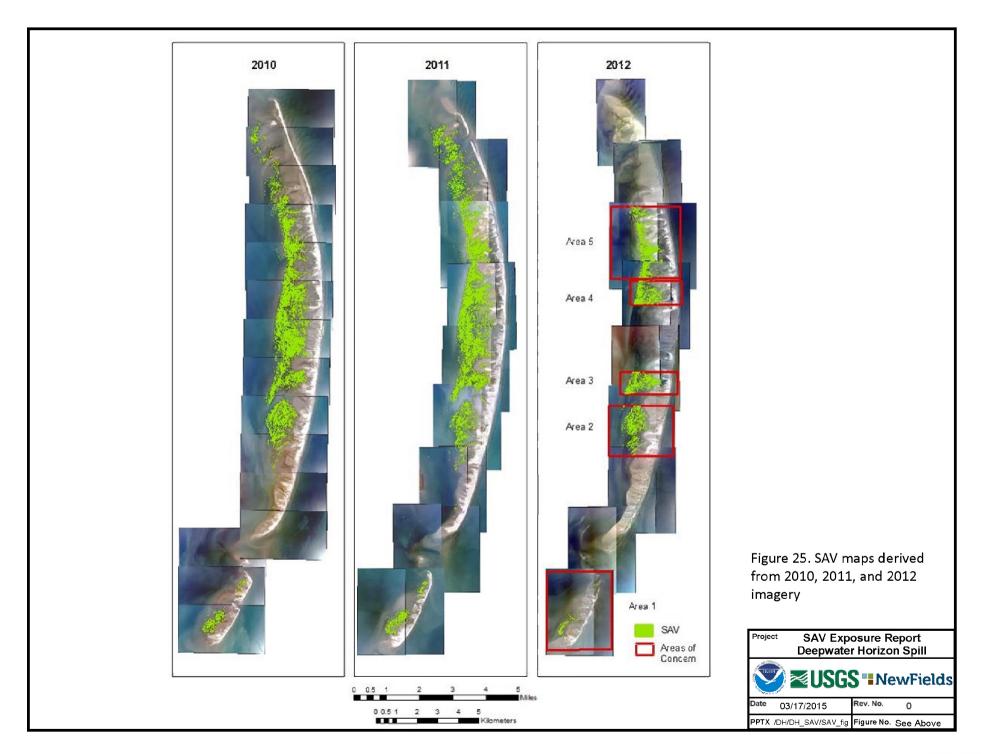












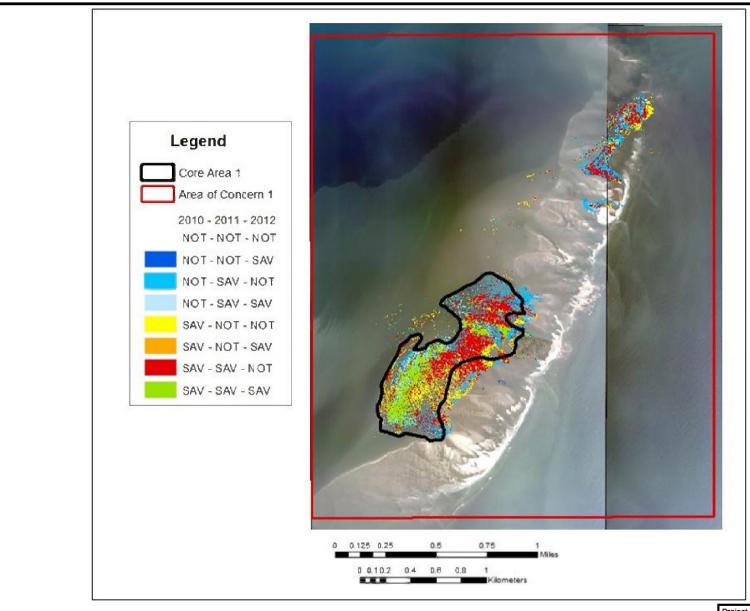


Figure 26. Change in SAV in Area of Concern 1. Legend indicates the presence or absence of SAV over 3 years (2010, 2011, and 2012). Core Area is outlined in black.



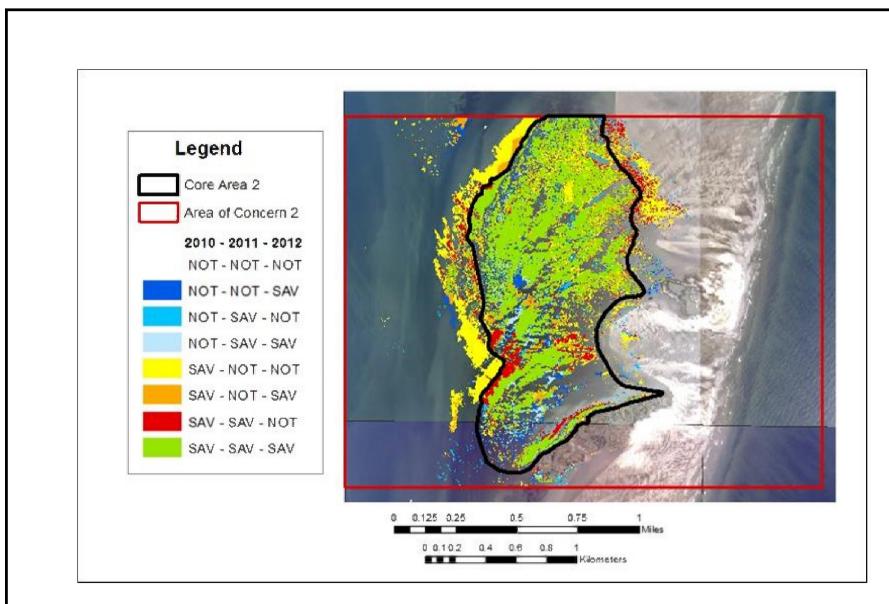


Figure 27. Change in SAV in Area of Concern 2. Legend indicates the presence or absence of SAV over 3 years (2010, 2011, and 2012). Core Area is outlined in black.



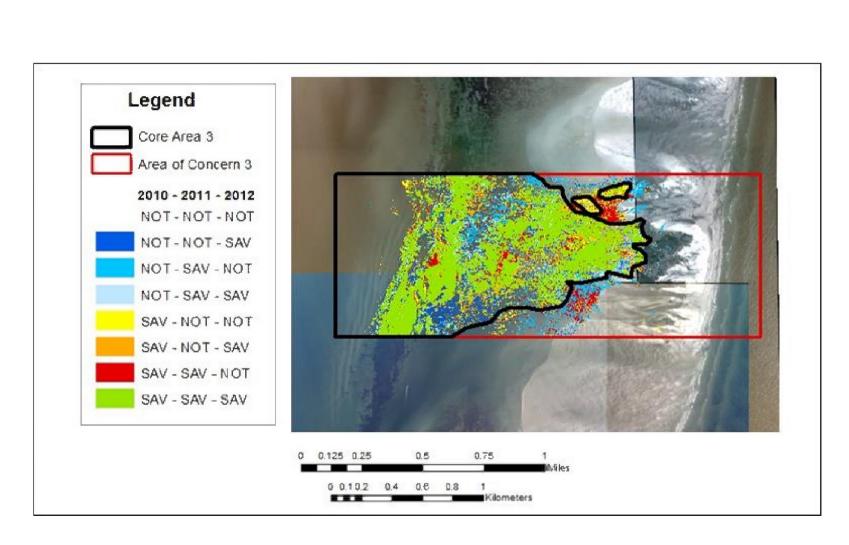


Figure 28. Change in SAV in Area of Concern 3. Legend indicates the presence or absence of SAV over 3 years (2010, 2011, and 2012). Core Area is outlined in black.



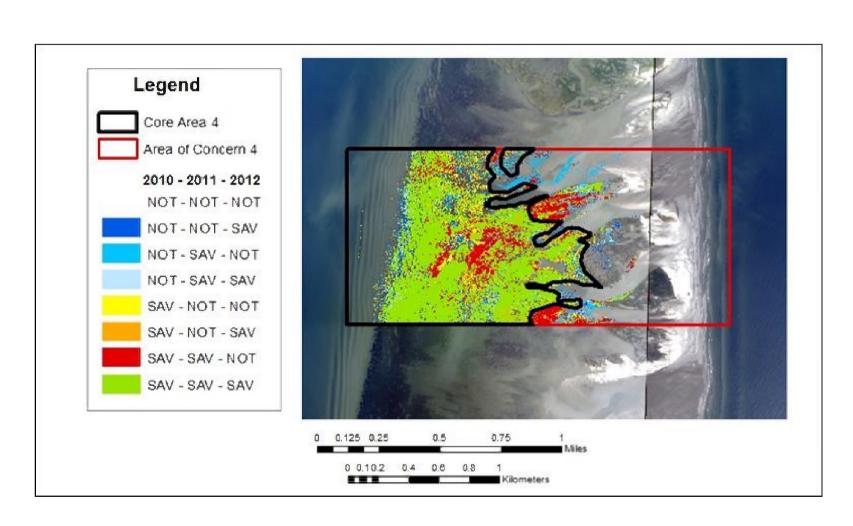


Figure 29. Change in SAV in Area of Concern 4. Legend indicates the presence or absence of SAV over 3 years (2010, 2011, and 2012). Core Area is outlined in black.

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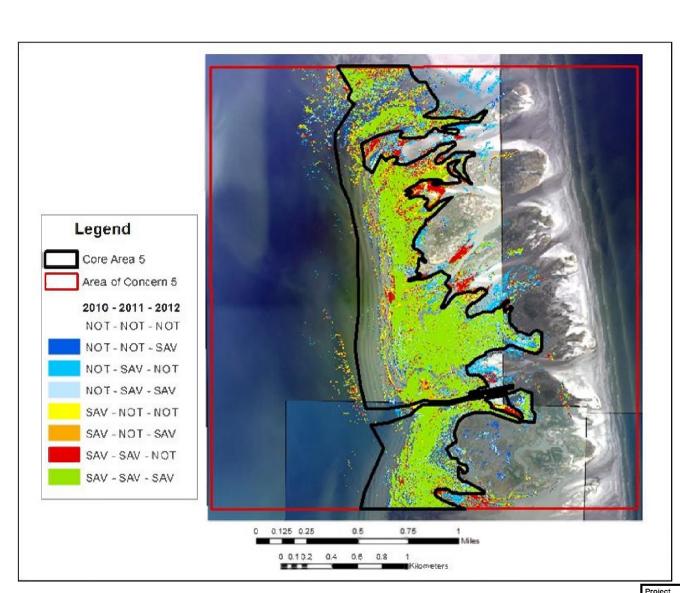
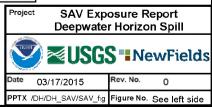
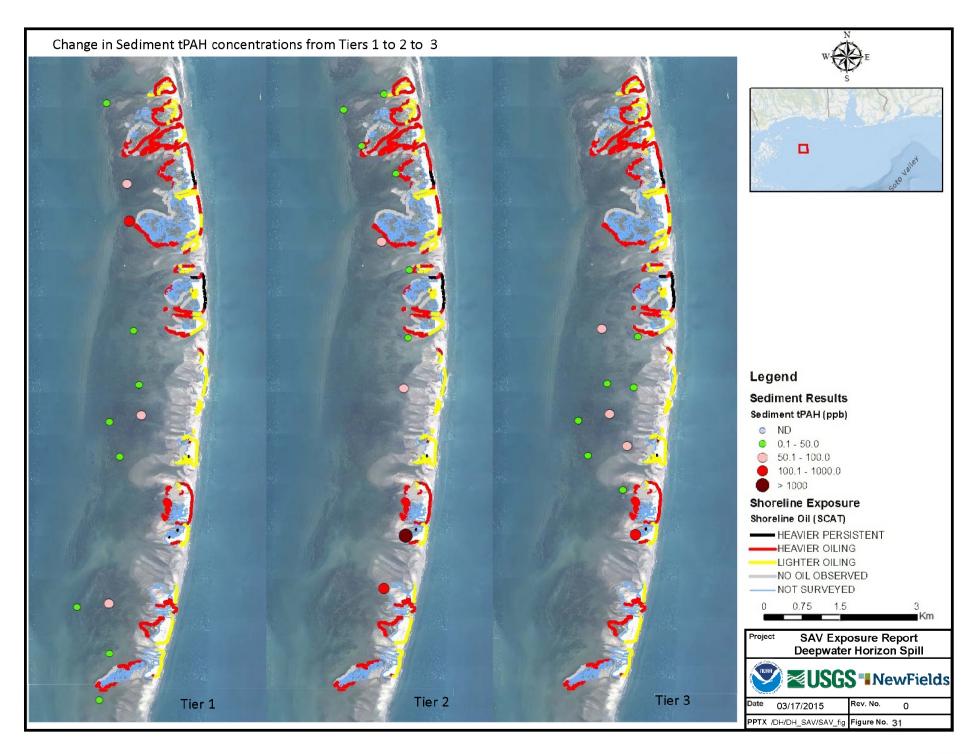
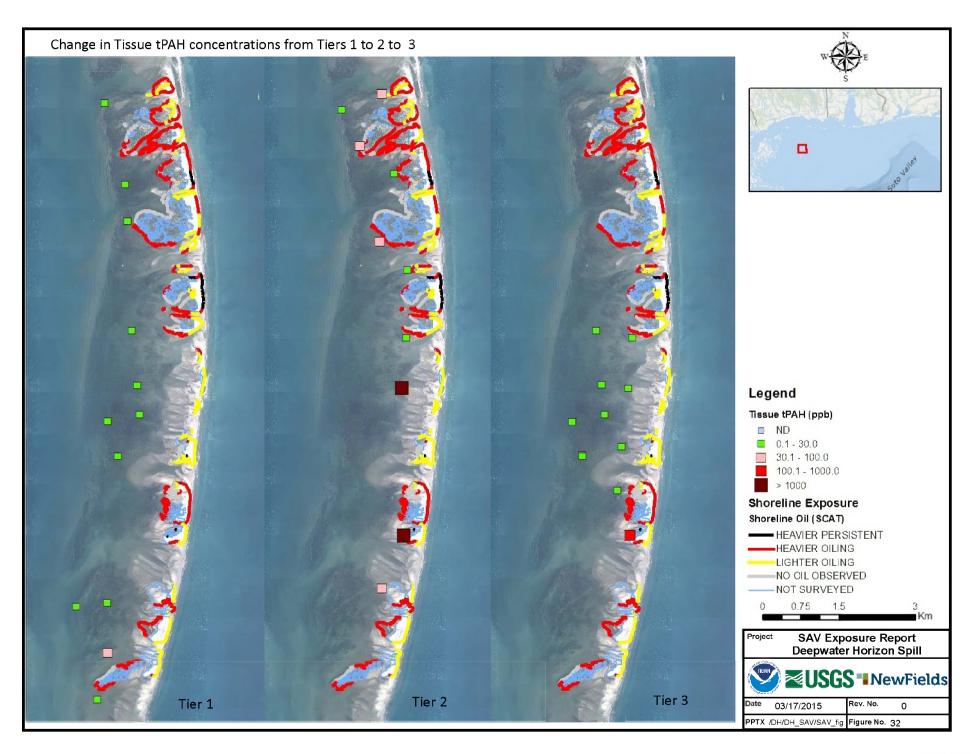
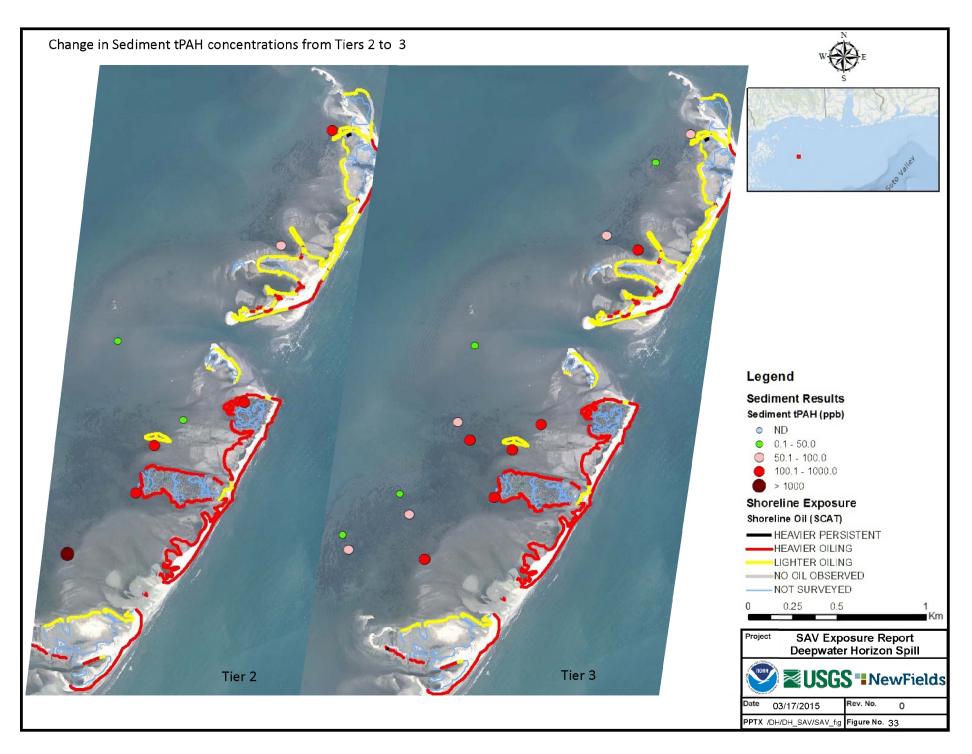


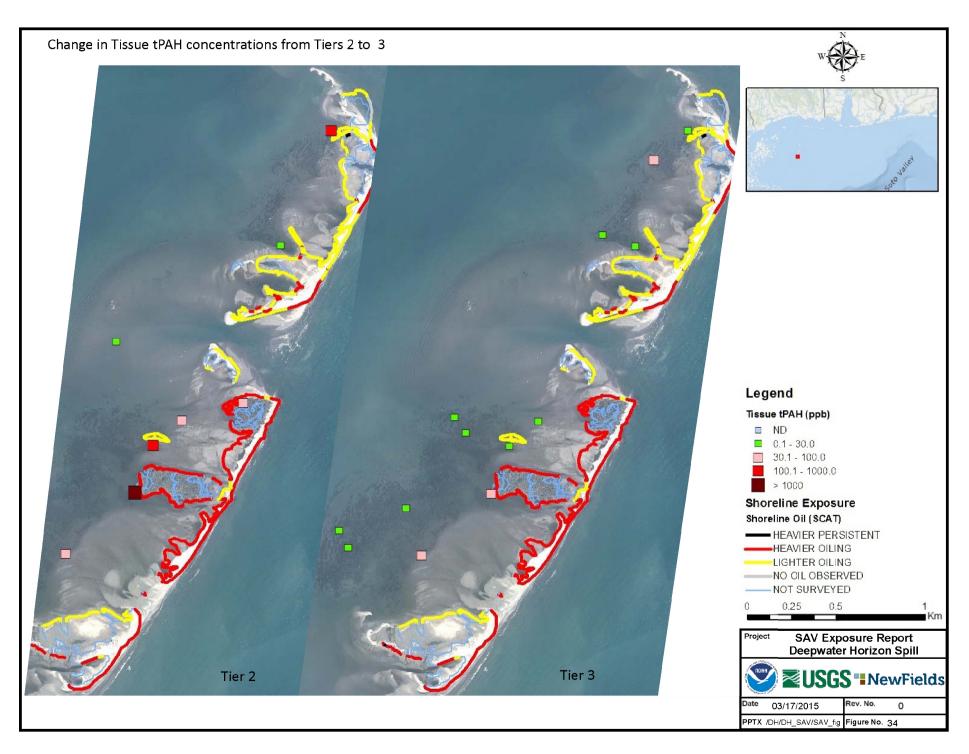
Figure 30. Change in SAV in Area of Concern 5. Legend indicates the presence or absence of SAV over 3 years (2010, 2011, and 2012). Core Area is outlined in black.









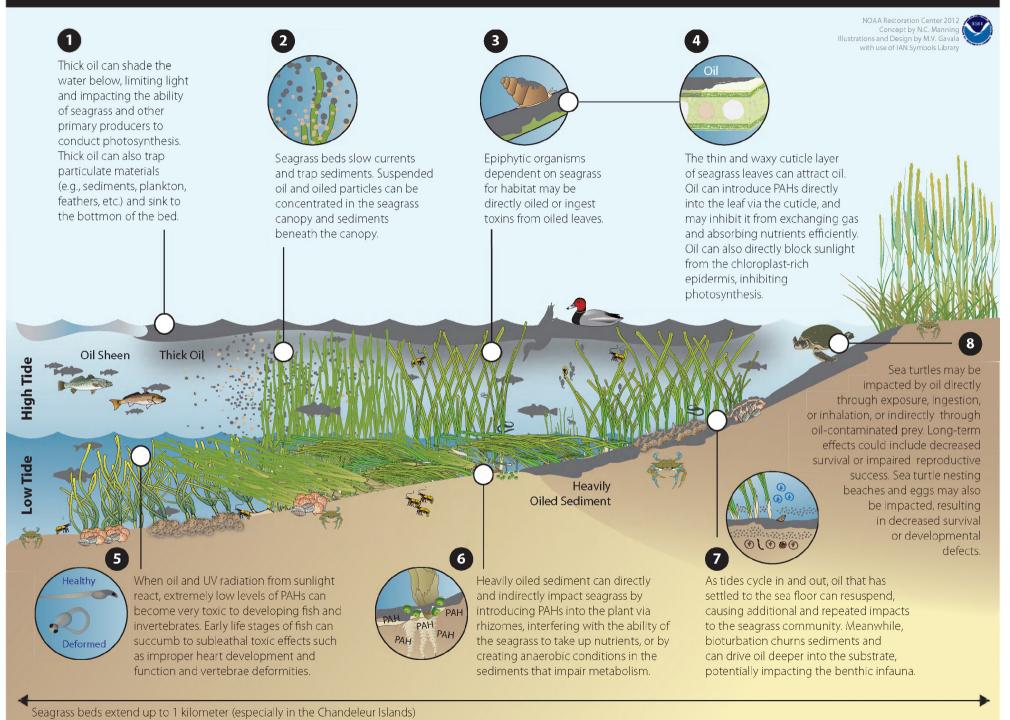




Submerged Aquatic Vegetation Exposure to Deepwater Horizon Spill

Appendix A – Conceptual Model

Potential Impacts of Oil to Seagrasses and Associated Organisms





Submerged Aquatic Vegetation Exposure to Deepwater Horizon Spill

Appendix B - Passive Sampling of Dissolved Oil Using PEMDs

Biologically available polynuclear aromatic hydrocarbons in submerged aquatic vegetation near barrier islands after the Deepwater Horizon blowout

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Abstract

Deepwater Horizon (DWH) oil was biologically available in portions of submerged aquatic vegetation near the inner sides of Mississippi barrier islands along the northern Gulf of Mexico after the 2010 blowout. This dissolved oil was detected with passive samplers (low-density polyethylene membrane devices, PEMDs) deployed for 2 weeks in September 2010, about a month after active oiling ceased. Total polynuclear aromatic hydrocarbon (PAH) concentrations were not high in any samplers but were greatest in the southern portion of the Chandeleur Islands (mean 373 ng/g), an area of relatively high beach oiling, and generally declined toward the north. Concentrations in central and northern samples were typically greater inshore than in paired offshore samples, consistent with an oiling event and subsequent leaching of PAHs from stranded oil. Offshore concentrations were about the same as inshore concentrations in the southern area, suggesting more extensive oiling; we predict this will be evident in sediment samples. Measured across Horn and Petit Bois Islands, biologically available oil concentrations were relatively low to the west (mean 190 ng/g), highest in the central area (373 ng/g), and declined to the east (140 ng/g). Concentrations in Big Lagoon, adjacent to Pensacola Bay, were elevated (mean 492 ng/g), were about the same at the mouth of Pensacola Bay, and dropped off inside the bay. Non-DWH sources were evident and variable across the sample area and contributed to total PAH concentrations. Fluoranthene-pyrene content in the Pensacola area was likely from an urban source; this was less prominent to the west (intermediate in Petit Bois and Horn Island samples and least in Chandeleur samples). Conversely, perylene concentrations, probably produced in anoxic sediments, were relatively high in the Chandeleur Islands but below method detection limits in all samples further east. Distributions and gradients of naphthalenes through phenanthrenes (and possibly chrysenes) reflect the bioavailability of PAH during the September 2011 sampling, are consistent with observed beach oiling, and were likely caused by differential DWH oiling over a period of time. However, assignment of DWH as the primary source oil is complicated because of composition differences among matrices (PAHs moved from oil to water to passive samplers), differential weathering, and by contributions from other PAH sources, hence will depend on verification from other samples such as sediment and vegetation collected at about the same places and times.

Introduction

The purpose of this study was to determine if toxic oil constituents known as polynuclear aromatic hydrocarbons (PAHs) were biologically available in nearshore submerged aquatic vegetation (SAV) along the inner sides of barrier islands of the north-central area of the Gulf of Mexico (Louisiana, Mississippi, Alabama, and Florida) as a result of the Deepwater Horizon (DWH) blowout. To accomplish this, passive samplers were deployed to accumulate PAHs from seawater in SAV beds, thereby increasing concentrations by a factor of 10^4 to 10^5 (Carls et al., 2004) and allowing cumulative detection over the 10 d deployment period. Deployments were in September 2010, about a month after the active shoreline oiling ceased.

The barrier islands studied (Chandeleur, Horn, and Petti Bois Islands, and Perdido Key near Pensacola) were formed as part of the Mississippi River delta (Fig. 1). These islands are ephemeral; they support some terrestrial vegetation (including trees on the more stable islands) but have generally been shrinking and migrating. Their formation and movement is driven by interaction between sea level, sedimentation, meteorlogical, and oceanographic conditions and they tend to undergo rapid change as a result of hurricane activity (Schmid 2003).

Submerged aquatic vegetation (SAV) beds (or communities) are important components of the barrier island ecosystems. They remove nutrients and pollutants from coastal runoff, reduce sediment erosion, and provide food and nursery habitat for many aquatic animals, including fish, crustaceans, waterfowl, turtles, and manatees (Neckles et al., 1997). The SAV beds are shallow, often less than 2 m, and are vulnerable to physical change and chemical contamination.

The DWH well, located in the Gulf of Mexico (about 28.7367° N latitude, 88.3872° W longitude), blew out on April 20, 2010. By May 1 the slick spread north to the southern portion of the Chandeleur Islands; the central and northern portions were likely oiled for the first time on May 8 (NOAA/NOS/OR&R 2010). The slick expanded and contracted multiple times over the next several weeks and eventually extended north to the other barrier islands; it likely reached Petit Bois Island on June 2, 2010; the Pensacola area on June 5; and Horn Island on June 12 (NOAA/NOS/OR&R 2010). Reoiling of the Chandeleur and northern barrier islands continued until about July 27 as the slick moved about with north-south and east-west motions. Oil was last forecast near the Pensacola area on July 10, 2010 and in the other study islands on July 27, 2010 (NOAA/NOS/OR&R 2010).

There was concern that the SAV beds had been exposed to DWH oil and that exposure would persist as a result of stranded, sequestered oil. This passive sampler project provided a method to measure the possibility of continued contamination after oiling ceased by controlled, time-specific deployment. Specifically, two samplers were deployed at each site, one near shore, and one offshore. This was repeated at numerous sites along Chandeleur, Petit Bois, and Horn Islands, and near and within Pensacola Bay. Our specific objectives were to determine the bioavailability of PAH in these sites by measuring relative PAH concentrations in the passive samplers. The PAH composition and weathering parameters for each area aided judgment of whether DWH was responsible for the PAH contamination.

Methods

Sampler preparation and deployment

The bioavailability and composition of mobile oil constituents was assessed in SAV beds along Mississippi River barrier islands (Chandeleur, Horn, and Petit Bois Islands), in a lagoon behind Perdido Key, and within Pensacola Bay with passive samplers. These passive samplers were low-density polyethylene plastic strips ($^{\sim}98~\mu m \times 4.9~cm \times 50~or~70~cm$), known as polyethylene membrane sampling devices, or PEMDs (Carls et al., 2004). The plastic was purchased without additives (Brentwood Plastics, St. Louis, MO, USA), cut as indicated, immersed in pentane and alternately sonicated for 15 min and soaked for 30 min (two cycles), and rinsed with clean pentane after a final 15 min extraction. Laboratory, field, and trip blanks were included for quality control. All samplers were double-wrapped in aluminum foil and double-bagged in ziplock bags for shipping to and from the field. Samplers were frozen before shipment to the field and after receipt at the laboratory.

The passive samplers were deployed using large links of chain as anchors and fastened to buoy lines. They were deployed September 4 to 6, 2010, about a month after active oiling ceased and were retrieved 10 d later (September 14 to 16, 2010). Pensacola samples (including the lagoon behind Perdido Key) were deployed September 19, 2010 and retrieved September 29, 2010. Paired passive samplers were deployed at every site in two different ways, bare and armored. Armored samples (50 cm strips) were housed in aluminum canisters (11.5 cm diameter \times 6.6 cm) with perforated aluminum endplates (3 mm holes spaced 4.8 mm apart). Bare plastic (70 cm) strips were heat-sealed to solvent-cleaned halibut clips. Armored samplers were attached 50 cm above anchors with a carabiner and bare samplers were attached 25 to 30 cm above them. Deployment was successful for both gear types and there were no significant differences in sampling effectiveness.

Hydrocarbon measurement and analysis

Passive samplers were wiped clean to remove gross surface contamination and cut into 25 cm sections. These sections were placed in centrifuge tubes and spiked with 500 μ l of a deuterated surrogate recovery standard (Table 1). The spike solvent (hexane) was allowed to evaporate and the plastic was extracted in a sonic bath with 100 ml of 80:20 mixture of pentane:dichloromethane for 120 min (three 20 min sonications with a 30 min rest between each sonication). The plastic was immediately rinsed with pentane as it was removed after the final sonication. The extracts were dried with sodium sulfate, concentrated to 1 ml in hexane, and purified on a chromatography column (1.5 g 5% deactivated silica gel). Samples were eluted with 22 ml of a 1:1 mixture of pentane and dichloromethane. Extracts were spiked with an internal standard, hexamethyl-benzene, and stored at -20° C pending analysis.

The aromatic fraction of passive sampler extracts were analyzed for PAHs by a gas chromatograph equipped with a mass selective detector (GC/MSD). The data were acquired in selected ion monitoring (SIM) mode and concentrations were determined by the internal standard method (Short et al., 1996; Carls et al., 2004). Experimentally determined method detection limits (MDLs) were 0.2 to 3.9 ng/g. The accuracy of the PAH analyses was about \pm 15% based on comparison with National Institute of Standards and Technology values, and precision expressed as coefficient of variation was usually less than about 20%, depending on the PAH. Total PAH (TPAH) concentrations were calculated by summing

concentrations of individual PAH [naphthalenes (N0 to N4), biphenyl (BP), acenaphthylene (AC), acenaphthene (AE), fluorenes (F0 to F4), dibenzothiophenes (D0 to D4), phenanthrenes (P0 to P4), anthracene (AN), fluoranthene (FL), pyrene (PY), fluoranthenes/pyrenes (A1 to A4), benzo(a)anthracene (AA), chrysenes (C0 to C4), benzo(b)fluoranthene (BB), benzo(k)fluoranthene (BK), Benzo(e)pyrene (BE), Benzo(a)pyrene (BA), indeno(1,2,3-cd)pyrene (IC), dibenzo(a,h)anthracene (DB), benzo(ghi)perylene (BZ)]. Perylene was not included in total PAH (TPAH) estimates to avoid inclusion of biogenic sources. Significant (above MDL) perylene concentrations were observed only in the Chandeuler Islands, where it accounted for 4 to 52% (mean 21%) of all PAHs. Relative PAH concentrations were calculated as the ratio of PAH_i/TPAH.

Principal component analysis (PCA) was used to explore PAH composition among all field samples. Source oil and weathered oil samples were included in these analyses. One analyte, IC, that was consistently not detected, was excluded from PCA. Exclusion of perylene from PCA had little influence and no meaningful influence on the results.

Results and Discussion

The evidence collected with passive samplers indicates DWH was biologically available in SAV beds at low levels. All samples contained PAHs common in oil at concentrations above method detection limits. Distributions and gradients of naphthalenes through phenanthrenes (and possibly chrysenes) were likely caused by differential exposure to weathered DWH oil. Other PAH sources were present and variable across the sample area and contributed to total PAH concentrations; fluoranthene-pyrenes from an unknown, but possibly urban source were prominent near Pensacola and declined to the west. Conversely, perylene (probably produced in anoxic sediments) concentrations were relatively high in the Chandeleur Islands but were below method detection limits further east.

Low PAH concentrations were detected in all passive samplers

PAH concentrations in passive samplers were low, ranging from 56 to 751 ng/g (without perylene). No spurious contamination was detected in field and trip blanks and TPAH in blanks was significantly less than in all environmental samples. These low environmental concentrations are not surprising given that the source oil was weathered, probably from oil stranded in nearby beaches. Aqueous PAH concentrations in September 2010 were likely less than peak concentrations; these passive samplers concentrate PAHs from seawater by a factor of 10^4 to 10^5 (Carls et al., 2004). Although detected PAH concentrations were low, they do indicate chronic exposure through at least September 2010.

North-south and east-west TPAH gradients were observed in beds along the barrier islands. Total PAH concentrations were greatest in the southern portion of the Chandeleur Islands (mean 373 ng/g) and generally declined toward the north (186 ng/g central and 174 ng/g north; Fig. 2). Measured across Horn and Petit Bois Islands, biologically available oil concentrations were relatively low to the west (mean 190 ng/g), highest in the central area (373 ng/g), and lower to the east (140 ng/g; Fig. 2). The Pensacola Bay area also had relatively high PAH concentrations, but included an additional source of contamination (see discussion in the composition section). Concentrations in Big Lagoon, adjacent to

Pensacola Bay, were elevated (mean 492 ng/g), were about the same at the mouth of Pensacola Bay, and dropped off inside the bay (Fig. 2).

Total PAH concentrations in passive samplers tended to be greater inshore than offshore along the Chandeleur Islands. Concentrations in central and northern inshore samples were typically greater than in paired offshore samples, consistent with an oiling event and subsequent leaching of PAHs from stranded oil (Fig. 2). However, offshore concentrations were about the same as inshore concentrations in the southern area, suggesting more extensive substrate oiling (Fig. 2). Paired inshore-offshore sampling was restricted to the Chandeleur Islands.

PAH composition in passive samplers

All samples contained PAHs common in oil at concentrations above method detection limits; naphthalenes, fluorenes, dibenzothiophenes, phenanthrenes, and chrysenes. Of these, relative phenanthrene concentrations were greatest. Naphthalene, fluorene, dibenzothiophene, and phenanthrene patterns were consistent with a weathered petrogenic (oil) source (Fig. 3 and Appendix 1), and this was confirmed with modeling (Appendix 2). Although passive samplers are ideal for detecting biologically available PAHs in the water column, they are not the best substrate for comparing composition to source oil and identifying the source. Oil- and oil-contaminated sediments are the best substrates for identifying source oil. Nevertheless, PAH composition in the passive samplers was consistent with DWH oil, and sediments collected from the same areas should corroborate DWH as the source.

Chrysene composition in passive samplers across the study area was consistent with a pyrogenic source, yet may have also originated from DWH oil. Parent chrysene concentrations were largest and chrysene concentrations fell with alkyl substitution (Fig. 3), thus composition had a pyrogenic character, different than in oil (and DWH oil, where parent chrysene concentrations were lower than some of the alkyl-substituted chrysene concentrations; Fig. 4). Hence, chrysenes may have originated from a non-oil source (such as soot from combustion processes). Alternatively, the inversion may be a sampling artifact; the pattern can reverse from petrogenic to pyrogenic because parent (and least-substituted) congeners are more soluble in water than more-substituted congeners, thus aqueous transfer from source oil to PEMD may alter the composition pattern. This has been demonstrated in the laboratory with Alaska North Slope crude oil under some circumstances (Appendix 3) and we are planning to determine if this is the case for DWH oil when sampled by passive samplers. Although the origin of chrysenes in the study area cannot be definitively assigned to DWH oil, their relative total concentrations were consistent with weathered DWH oil across the sample area and did not vary with presumptive soot contamination (see next section).

Regional PAH composition in passive samplers

Composition of PAHs varied among collection sites and included at least two non-DWH sources. Perylene concentrations were substantial in the Chandeleur Islands and consistently above method detection limits, indicating the presence of a contemporary, biogenic source (Fig. 3). (Perylene was also observed further to the west in Black, Barataria, and Timbalier Bays in another study; Carls et al.,

unpublished data.) In contrast, perylene concentrations were below method detection limits in other locations (Horn and Petit Bois Islands, and Pensacola; Fig. 3). A second non-DWH source was prominent in the Pensacola area, evident by the greatest relative and absolute abundance of fluoranthenes and pyrenes (mean 131 ng/g); these compounds were least represented in the Chandeleur Islands (mean 10 ng/g). Two higher molecular weight compounds, benzo(b)- and benzo(k)-fluoranthene were present above method detection limits in Pensacola samples but not elsewhere (Fig. 3).

Regional variation in PAH composition was also clearly evident when summarized by PCA. Chandeleur, Horn, and Petit Bois Island samples formed two overlapping clusters (Fig. 5). Pensacola samples formed a distinct cluster (Fig. 5). Rough north-south and east-west gradients were evident across the Chandeleur, Horn, and Petit Bois clusters. A second PCA analysis restricted to naphthalenes through phenanthrenes for the purpose of focusing only on DWH oil components (and excluding other possible regional inputs) clustered all SAV sites more closely together and included weathered DWH oil within this cluster (Fig.6). Again, north-south and east-west gradients were evident (Fig.6), consistent with the more general PCA (Fig. 5) and the trends evident in PAH composition plots (Fig. 3). Weathered DWH oil, both mousse and tar, was most closely related to Chandeleur Island samples (Fig. 6). Tar balls from an unknown source, collected at the eastern end of Petit Bois Island, had a very high perylene content (68 to 74%) and did not cluster directly with any other samples (Fig. 6).

Oil weathering

Weathering varied geographically. Modeled weathering, w, (Short and Heintz 1997) indicated the presence of weathered oil throughout the Chandeleur Islands, though w was not estimable in some locations (Fig. 7). Weathering was generally least in the northern Chandeleur islands. Offshore samples in the south were often more weathered than inshore counterparts. Greater weathering in the southern than northern Chandeleur Islands is also consistent with PCA results; DWH tar was more weathered than DWH mousse; tar clustered with southern samples and mousse with northern samples (Fig. 6). Weathering was typically not estimable by modeling in Horn and Petit Bois Islands and Pensacola (Fig. 7).

Was DWH the source of oil in passive samplers?

The preceding paragraphs demonstrated that the composition and chemical behavior of some PAHs sequestered in the passive samplers was consistent with DWH oil but that is not absolute proof that DWH was the source. Additional lines of evidence are necessary to draw definitive conclusions. These additional lines of evidence include knowledge of DWH oil timing and distribution from slick observations, visual assessment of stranded shoreline oil (SCAT surveys), and direct chemical measurement of oil, tar, and oiled sediment samples. Direct measurement of DWH oil in the same area provides chemically unaltered information about DWH composition and weathering.

To recap the chemical evidence of DWH oil contained in passive samplers, the relative composition of several homologous PAH families (naphthalenes, fluorenes, dibenzothiophenes, phenanthrenes, and possibly chrysenes) was consistent with DWH oil (compare Fig. 4 to Fig. 3); percentages (without perylene) were reasonably similar to weathered DWH oil, though the presence of other PAH sources

influenced this comparison, particularly in Pensacola. Dissolution of PAHs and subsequent reuptake by passive samplers also likely influenced the observed composition, yet the overall patterns were quite similar. However, as previously discussed, the pattern within chrysenes may have been altered by dissolution. All indications are that the DWH oil that contaminated the Mississippi barrier island area was weathered (Fig. 4), consistent with the weathered pattern observed in passive samplers (Fig. 3). The similarity of PAH composition in weathered DWH oil and passive samplers deployed in SAV beds was statistically supported with PCA when data were restricted to likely oil components (Fig. 6). Other unknown hydrocarbon sources added PAHs to the composition observed in SAV beds but this does not change the conclusion that DWH was present. (There were at least two other sources, a biogenic source that was present primarily in the Chandeleur Islands and a pyrogenic source that may represent urban soot, most prevalent in Pensacola.)

The geographic distribution and timing of PAHs sequestered in passive samplers was consistent with the distribution of DWH oil. In particular, the oil moved from south to north, and passive sampler data indicate the southern Chandeluer Islands were the most impacted area studied. (Total PAH concentrations in Pensacola were lower than those in the southern Chandeleur Islands when non-DWH components were discounted.) Observed slick movement was not uniform (Figs. 1 and 8), thus non-uniform oil distributions were anticipated, as also observed in SCAT beach surveys. The weight of evidence, therefore, suggests that the very large (779 million liters), long-lasting (April to July 2010) DWH oil blowout was the source of the oil detected. The DWH oil interacted with the study area from early May to late July (e.g., Figs. 1 and 8), was observed on adjacent beaches by SCAT survey, and was detected in proximal sediments (Scott Stout, personal communication). Thus, the likelihood that residual DWH oil was the source of PAHs sequestered in passive samplers deployed in SAV beds in September 2010 is substantial. Oil constituents from intertidal sources oiled by much smaller spills have been detectable with passive sampling techniques for years (Carls, unpublished *Selendang Ayu* data) to more than a decade (Carls et al., 2004; Springman et al., 2008).

Hydrocarbon composition, geographic distribution, correspondence with oiled beaches, and timing thus all provide evidence that the source of PAHs in passive samplers deployed in SAV beds along the Mississippi barrier islands originated from DWH oil. We expect that direct measures of sediment and oiled sediment completed by other investigators at about the same places and times will yield similar results and conclusions, corroborating the evidence that DWH oil was biologically available and detected in passive samplers. Multiple lines of evidence will strengthen these conclusions (or not) and we look forward to contributing to a synthesis publication.

Conclusion

Polynuclear aromatic hydrocarbons were detected in all passive samplers deployed in September 2011, thus providing evidence that PAHs were bioavailable to fauna in the SAV beds. All concentrations were low, but the greatest mean concentrations were detected in SAV beds in the southern Chandeleur Islands. Distributions and gradients of naphthalenes through phenanthrenes (and possibly chrysenes) were consistent with observed beach oiling and were likely caused by differential DWH oiling over a

period of time. However, assignment of DWH as the primary source oil is complicated because of composition differences among matrices (PAHs moved from oil to water to passive samplers), differential weathering, and by contributions from other PAH sources, hence will depend on verification from other samples such as sediment and vegetation collected at about the same places and times.

References

- Carls, M.G., Heintz, R.A., Marty, G.D., Rice, S.D., 2005. Cytochrome P4501A induction in oil-exposed pink salmon *Oncorhynchus gorbuscha* embryos predicts reduced survival potential. Marine Ecology-Progress Series 301, 253-265.
- Carls, M.G., Holland, L.G., Short, J.W., Heintz, R.A., Rice, S.D., 2004. Monitoring polynuclear aromatic hydrocarbons in aqueous environments with passive low-density polyethylene membrane devices. Environmental Toxicology and Chemistry 23, 1416-1424.
- Carls, M.G., Rice, S.D., Marty, G.D., Naydan, D.K., 2004. Pink salmon spawning habitat is recovering a decade after the *Exxon Valdez* oil spill. Transactions of the American Fisheries Society 133, 834-844.
- Neckles, H.A., Guntenspergen, G.R., Rizzo, W.M., 1997. Global change and submerged aquatic vegetation research. U.S. Geological Survey, National Wetlands Research Center.
- NOAA/NOS/OR&R. 2010. Trajectory forecast Mississippi Canyon 252. [Deepwater Horizon] trajectory products by date.
- Schmid, K., 2003. East Ship Island evolution, morphology, and hurricane response 1994 to 2001.

 Mississippi Department of Environmental Quality, Office of Geology, Open-File Report 134.
- Short, J.W., Heintz, R.A., 1997. Identification of *Exxon Valdez* oil in sediments and tissues from Prince William Sound and the northwestern Gulf of Alaska based on a PAH weathering model. Environmental Science and Technology 31, 2375-2384.
- Short, J.W., Jackson, T.L., Larsen, M.L., Wade, T.L., 1996. Analytical methods used for the analysis of hydrocarbons in crude oil, tissues, sediments, and seawater collected for the natural resources damage assessment of the *Exxon Valdez* oil spill. American Fisheries Society Symposium 18, 140-148.
- Springman, K.R., Short, J.W., Lindeberg, M.R., Maselko, J.M., Kahn, C., Hodson, P.V., Rice, S.D., 2008. Semipermeable membrane devices link site-specific contaminants to effects: Part 1 Induction of CYP1A in rainbow trout from contaminants in Prince William Sound, Alaska. Marine Environmental Research 66, 477-486.

Table 1. Deuterated surrogate polynuclear aromatic hydrocarbon (PAH) standards and concentrations in hexane spike used for passive samplers.

(μg/ml)	Surrogate
0.5	naphthalene-d ₈
0.5	acenaphthene- d_{10}
0.5	phenanthrene-d ₁₀
0.5	chrysene-d ₁₂
0.5	perylene-d ₁₂
0.5	benzo[a]pyrene-d ₁₂

Fig. 1. Barrier island sample area in the north-central Gulf of Mexico. Deepwater Horizon (DWH) well and slick locations, Chandeleur Islands (C), Horn Island (H), Petit Bois Island (P), and Pensacola. NASA satellite photo, June 25, 2010 (http://earthobservatory.nasa.gov/NaturalHazards/view.php?id=44452).



Fig. 2. Total PAH distribution in SAV beds (method-detection limit corrected, without perylene). NOO is no observed oil. TPAHall.*mxd*

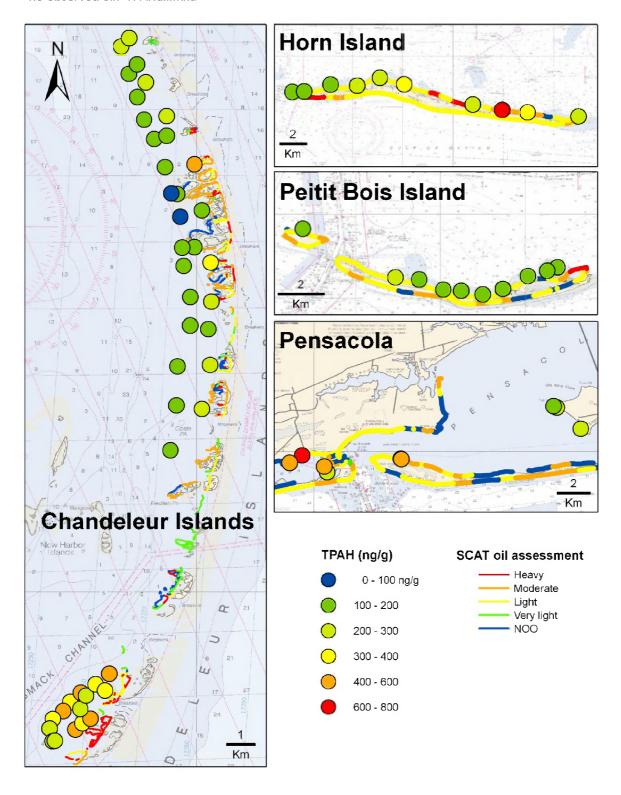


Fig. 3. Mean composition of PAHs in Mississippi barrier islands. Values within panels indicate percentages of naphthalenes, fluorenes, dibenzothiophenes, phenanthrenes, fluoranthenes/pyrenes, chrysenes, and higher molecular weight PAHs (left to right) excluding perylene. Vertical bars are ± 1 standard error. Mean total PAH concentrations (and range) are recorded at the upper right of each panel; the number of samples appears below. All values were corrected for method detection limits and do not include perylene. *allPAH.agr, SAV.dwg*

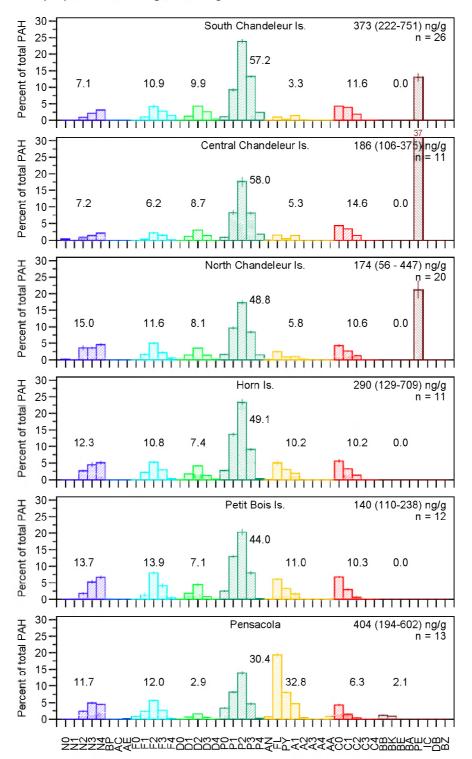


Fig. 4. Deepwater Horizon source oil, mousse, and tarball PAH composition. The bottom panel includes tarballs from an unknown source. *DWH.agr, SAV.dwg*

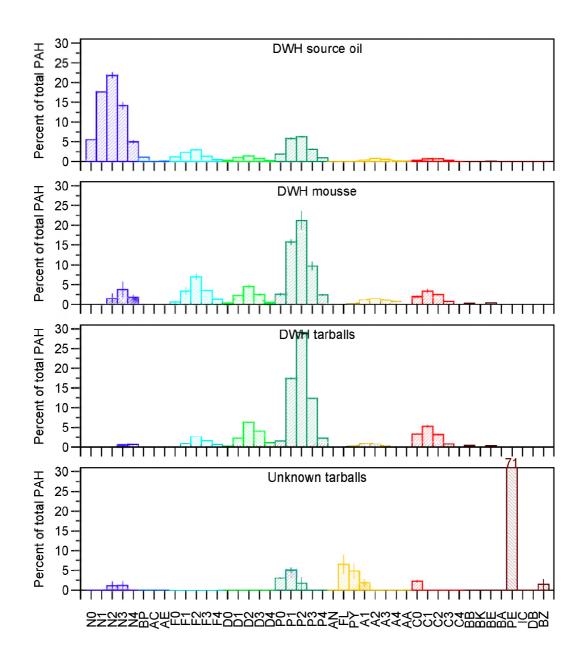


Fig. 5. Principal components analysis of normalized PAH composition in passive samplers deployed in SAV beds along Mississippi barrier islands. *Pca2a.agr*

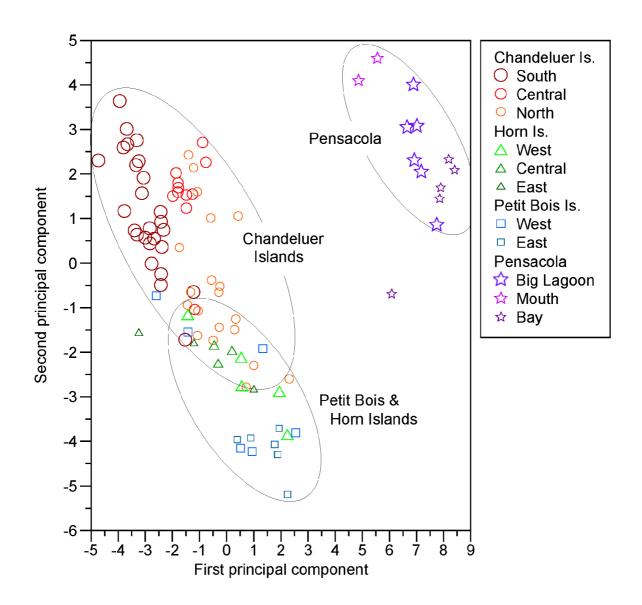


Fig. 6. Principal components analysis of normalized PAH composition in passive samplers deployed in SAV beds along Mississippi barrier islands. To focus on likely DWH oil content, composition was restricted to naphthalenes through phenanthrenes. *Pca5x.agr*

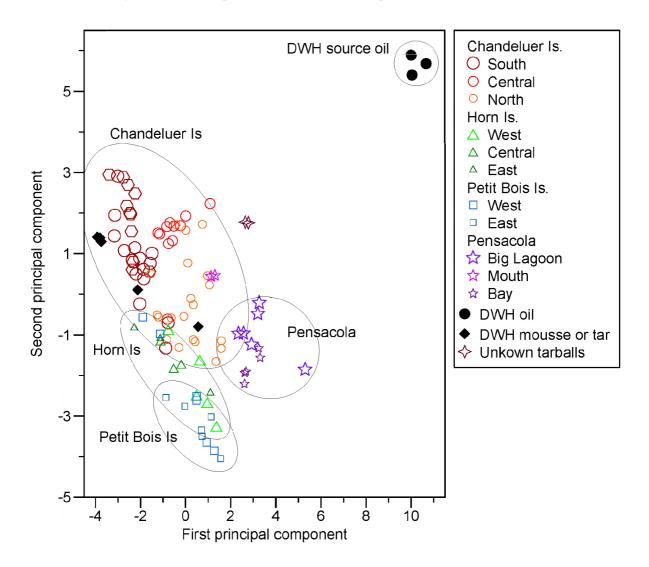


Fig. 7. Weathering, estimated by modeling (Short and Heintz 1997) in SAV beds. Weathering all.*jpg, weathering all.mxd*

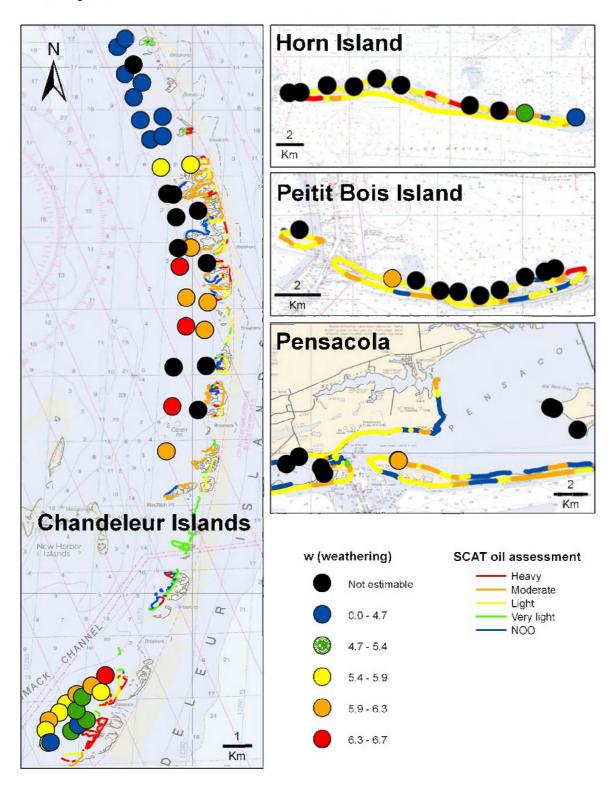


Fig. 8. Detailed example oiling of Horn and Petit Bois Islands. NASA satellite photo, June 26, 2010 (http://earthobservatory.nasa.gov/NaturalHazards/view.php?id=44466)



Appendix 1. Detailed biologically available PAH composition in SAV beds.

Biologically available polynuclear aromatic hydrocarbon (PAH) concentrations and composition were inferred from passive samplers (PEMDs, polyethylene membrane devices) deployed in submerged aquatic vegetation (SAV) beds after the Deepwater Horizon oil blowout in 2010. The illustration (Fig. A1.1) is intended to augment the summary figure provided in the main report.

Fig. A1.1. Mean composition of PAHs in Mississippi barrier islands. A) Chandeleur Islands. Values within panels indicate percentages of naphthalenes, fluorenes, dibenzothiophenes, phenanthrenes, fluoranthenes/pyrenes, chrysenes, and higher molecular weight PAHs (left to right). These estimates do not include perylene. Vertical bars are ± 1 standard error. Mean total PAH concentrations (and range) are recorded at the upper right of each panel; the number of samples appears below. All values were corrected for method detection limits and do not include perylene. *Chan.agr, SAV.dwg*

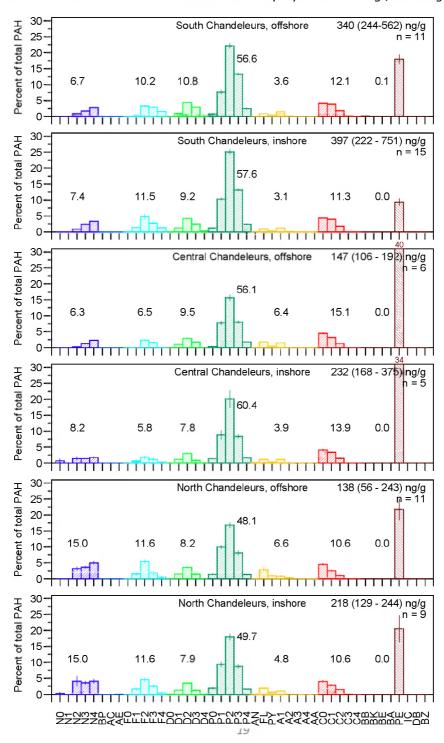


Fig.A1.1, continued. B) Horn and Petit Bois Islands. Horn.agr, SAV.dwg

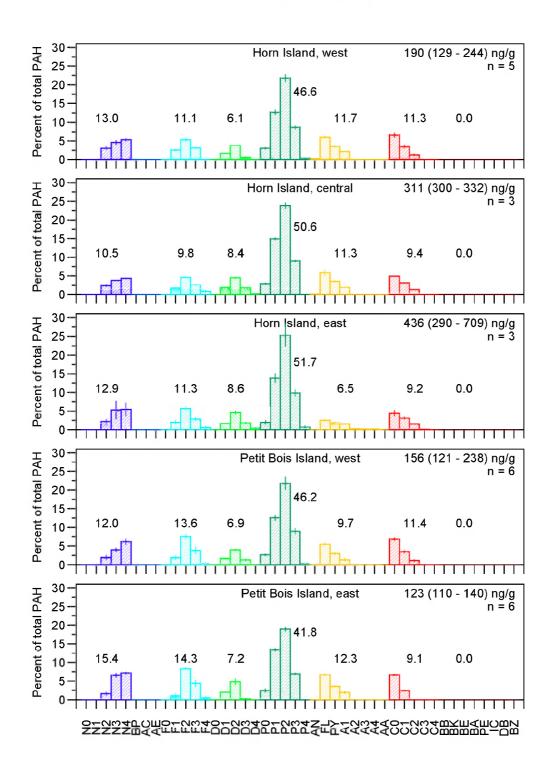
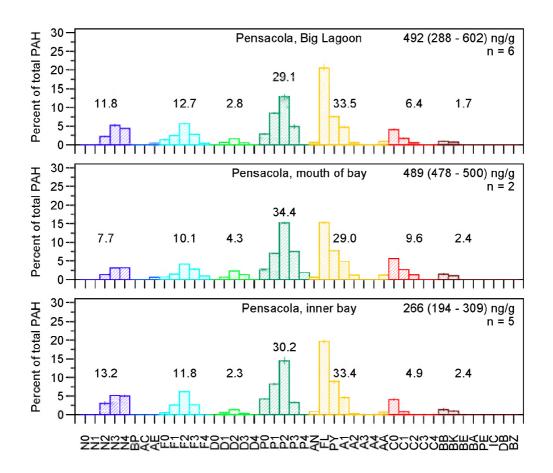


Fig. A1.1 continued. C) Pensacola. Pensa.agr, SAV.dwg



Appendix 2. Source modeling

Source modeling indicated PAHs in passive samplers deployed in SAV beds were consistent with oil, corroborating inferences drawn from the previously presented chemical evidence. Modeling indicated oil in nearly all Chandeluer Island samples, regardless of geographic position (Fig. A2.1). Source models also indicated oil in samples along Horn Island and most of those along Petit Bois Island (Fig. A2.1). Results of this source model (Carls 2006) generally did not support the presence of oil in Pensacola (Fig. A2.1), but results of a more recent model (Carls, unpublished) suggested that oil was plausible in the Pensacola area (Fig. A2.2). Failure to clearly identify oil in Pensacola samples was likely due to the more pyrogenic patterns evident in homologs heavier than the phenanthrenes. Interpretation of Pensacola samples merits further discussion and comparison to sediment results; reasons could be 1) non-DWH oil, 2) a mixture of oil, pyrogenic, and/or natural sources, or 3) sampling artifacts caused by dissolution of PAHs from oil and their recapture by passive samplers. However, because of their relative quantity, the fluoranthene/pyrenes in Pensacola samples most likely represent an independent hydrocarbon source and are probably an urban signature.

Fig. A2.1. Oil source modeling (Carls 2006) in SAV beds indicated oil was frequently present in Chandeleur, Horn, and Petit Bois samples. Model values ≥4 provide strong evidence for oil. *PetPyr all.mxd*

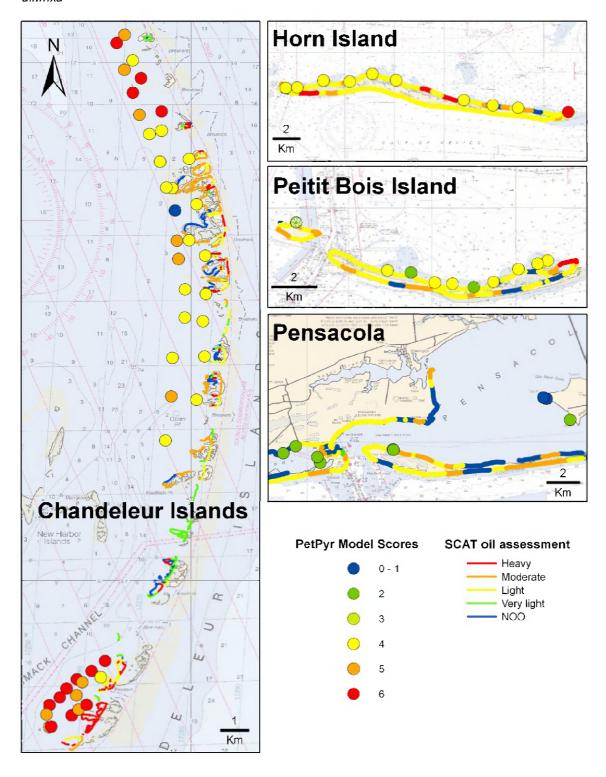
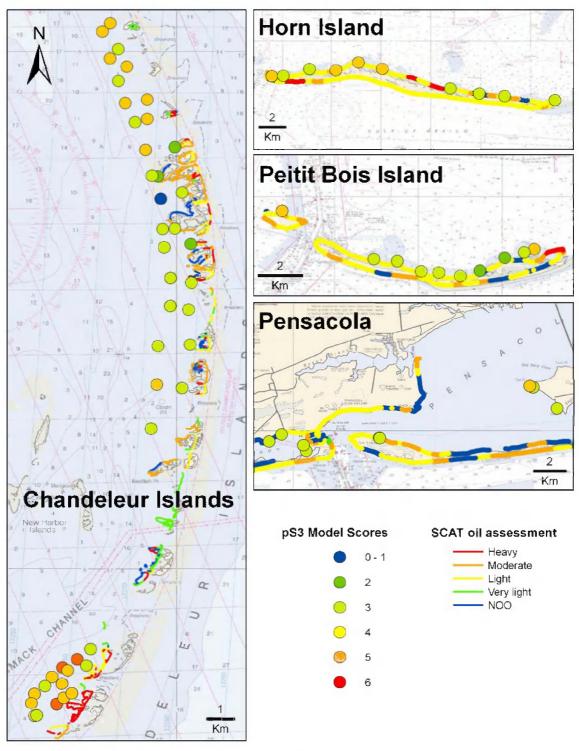


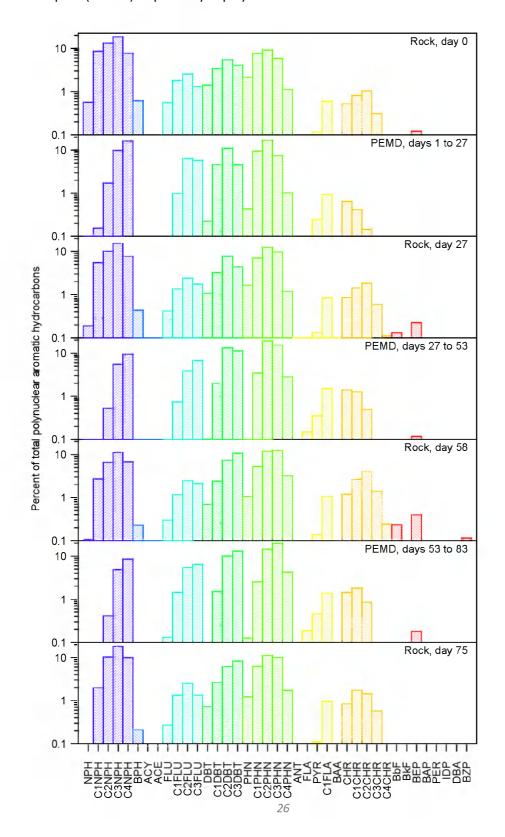
Fig. A2.2. Alternative **o**il source modeling (Carls, unpublished) in SAV beds frequently indicated oil was present. In particular, results for Pensacola samples were more similar to those elsewhere than determined by the model illustrated in Fig. A2.1. pS3 *all.mxd*



Appendix 3. Differences in PAH composition in source oil and passive samplers

Sources of PAHs (petrogenic and pyrogenic) are not always distinguishable in passive samplers (PEMDs, polyethylene membrane devices) due to chemical changes resultant from transfer from source oil to water to sampler. In the example here, rock was coated with weathered Alaska North Slope crude oil. The oil film was allowed to dry on the rock, rock was added to polyvinyl chloride cylinders, and water was passed through them (Carls et al., 2005). The PEMDs were deployed in effluent water, thus PAHs that left the rock and were sequestered in the passive samplers were mediated by aqueous transfer. In general, the smaller parent homologs were below method detection limits or present in relatively smaller quantities in PEMDs than in rock collected before and after exposure (NO, N1, FO, DO, PO). Conversely, relatively more N4, F3, C0, and C1 were present. Changes in chrysene (C) composition were often sufficient to change the pattern from petrogenic (as illustrated in the rock samples) to pyrogenic (declining from C0 to C4 as illustrated in PEMDs). However, this apparent change in source (from petrogenic to pyrogenic) was an artifact caused by composition change from rock to water and water to PEMDs.

Fig. A.3. Relative polynuclear aromatic hydrocarbon (PAH) composition in oiled rock columns and passive samplers (PEMDs) sequentially deployed in the effluent.





Submerged Aquatic Vegetation Exposure to Deepwater Horizon Spill

Appendix C - Synthetic Aperture Radar Satellite Imagery Footprints

