Supporting Information

Silliman et al. 10.1073/pnas.1204922109

SI Methods

Oil and Plants. Plant oiling and ecological surveys were conducted at three reference and three impacted sites in October 2010 with follow-up surveys at the same locations occurring until April 2011. Five transects were surveyed at each site, and the percentage of live plant cover and the percentage of oil cover at 1, 2, 4, 8, 16, and 32 m along a line perpendicular to the marsh edge were recorded. Transects were spaced ~2 m apart. Belowground samples of ~20 cm depth and 15 cm diameter were also collected at each site at 3 m and 15 m distances from the marsh edge to quantify differences in proportion of live rhizomes. Samples were rinsed in a sieve, and rhizomes were sorted from debris. Rhizomes were then categorized as alive if white and turgid or dead if dark and flaccid by using established protocols.

Animals. We surveyed animals at our impacted and reference sites during October 2010, April 2011, and then again during January 2012. The densities of live *Littoraria irrorata*, *Geukensia demissa*, *Pagurus longicarpus*, and *Uca pugnax* burrows were collected with 50 × 50 cm quadrats ($n = 5$) at both 3 m and 15 m distances from the shoreline at all survey sites. However, here we only present data from 3-m samples where we documented consistent oiling, and we also exclude data for *G. demissa* because they were exceedingly rare in all samples. Indeed, animal counts were low at all sites for all species, but changes were detectable.

Polyaromatic Hydrocarbon (PAH) Concentrations. Soil samples were collected at each survey site during October 2010 at 3 m and 15 m from the marsh edge. For each sample, four cores, 5 cm in depth, were homogenized in the field, and subsamples were placed in precombusted glass jars (3 h, 450 °C) and then frozen the evening of collection. Frozen samples were maintained in a cooler until reaching the laboratory, where they were stored in a freezer set at −80 °C.

Aliquots of 0.05–2.5 g wet weight were placed in 16–30 g baked sodium sulfate before extraction. Samples were spiked with 100 μL of surrogate standard containing 120 μg/mL *n*-tetradecane d-30, 109.92 μg/mL *n*-tetracosane d-50, 40 μg/mL naphthalene d-8, and 42.48 μg/mL fluoranthene d-10. The samples were extracted three times by the accelerated solvent extractor by using a hexane and acetone mixture (50:50, vol/vol). After extraction and reduction of the sample to 1 mL in a TurboVap concentrator, extracts were back-extracted three times into a 50:50 (vol/vol) mixture of sodium chloride solution and hexane to remove the remaining water. Samples were purified and separated into alkane and PAH compound classes by using activated silica open-column chromatography. Deuterated internal standards were added to the samples before compound quantification with GC/MS via direct injection onto a 30 m × 0·32 mm i.d. DB-5, 0.25 μm, fused silica capillary column (J & W Scientific). Details of silica column, chromatograph, and use of standards for compound quantification can be found in ref. 1.

pH, Redox, and Salinity. On June 25, 2010, we collected data to test for differences in pH, redox, and soil salinity for two reference and two impacted sites. Measurements for all three parameters were taken at 3 m and 15 m. A minimum of four evenly spaced samples (~2 m apart) was quantified at each site. Redox and pH measurements were taken ~5 cm below the marsh surface with a Hanna Instruments HI98183-01 pH/ORP portable meter. Salinity measurements (parts per thousand) were taken from pore water extracted from the top 5 cm of sediment in each area with a refractometer.

Oiled Plant Survival and Oil-Addition Experiments. To determine the relationship between oil coverage and plant survival, we did a field survey and a manipulative experiment. For the field survey, we went to four oiled sites and took five random quadrat samples at each site. In each quadrat, we quantified the proportion of stems covered with oil and the proportion of the stem that remained green. For the manipulated field experiment, we selected a reference and a nearby impacted site for oil-addition manipulations. On October 18, 2010, we identified 18 live stems with similar heights, live cover, and stem width, and each was randomly assigned to one of three treatments: (*i*) control, (*ii*) 40% oil addition, or (*iii*) 80% oil addition ($n = 6$, 18 total). Weathered oil was collected from a pool found during surveys of impacted sites (Fig. 3*F*) and applied to the designated plants within 4 h. Plants assigned to oil-addition treatments were manually covered with a thick coating of weathered oil from their base to either 40% or 80% of their total height. Control plants were rubbed with a clean latex glove for the same amount of time and in a similar manner as a procedural control. On December 10, 2010, plant senescence was quantified by recording the number of leaves that remained alive at a point approximately halfway up the plant. On 80% oil-addition plants, this measurement was taken under the layer of oil by wiping away weathered oil. Although some staining of live plant tissue did occur under the oil, green color was still identifiable in live leaves and, as a conservative measure, only completely brown and brittle leaves were quantified as dead. At the end of the experiment, plants were removed and properly disposed of to prevent contamination of the area.

Transplant Experiment. In June 2011, we took 12 cores that were 10 cm diameter and 20 cm deep from the reference area of three impacted sites, standardized so that each consisted of 10 *Spartina* stems. We then replanted a third of the cores back into the reference area as a procedural control for transplant shock, a third into the impacted area, and a third in the eroded area and counted the number of stems present after 4 mo.

Shoreline Erosion. In October 2010, PVC poles were placed into the ground abutted to marsh peat edge and the open water at six monitoring points along the shoreline of each impacted site and at six points along the shoreline of each reference site. PVC markers were not placed at locations with extreme shoreline curvature. In April 2011, June 2011, October 2011, and again in January 2012, we quantified erosion at these sites by measuring the distance between the marsh edge and the PVC poles.

Shoreline slopes. To quantify erosion potential for our reference and impacted sites, we quantified shoreline slopes at each of three impacted and reference sites. We used the same three impacted sites as above; however, we identified one alternative reference site for quantifying shoreline slope, erosion rates (described above), and modeling erosion potential (below). We chose one alternate reference site for these measurements (erosion rates, slope, and modeled wave stress) because one of the reference sites that we monitored was located in a state park and protected by a bulkhead. Our alternate site was similar in characteristics and wave exposure to our other five sites (exact locations are in Table S1). At 20 randomly selected points at each site, we measured the change in substrate height that occurred from the edge of the shoreline to a distance of 3 m seaward.

Modeling methods. To test the alternative hypothesis to influence of oil on erosion rates, that variation in measured marsh erosion rates could be attributable to their position relative to wave energy, we used SWAN, a numerical model of wind-generated wave growth and propagation (2). A 3-arcsec computation grid of bathymetry, obtained from the National Oceanic and Atmospheric Administration/National Geophysical Data Center US Coastal Relief Model (http://www.ngdc.noaa.gov/mgg/coastal/crm.html), was used for the wave field calculation over the region from −90.05° to −89.75° longitude and 29.25° to 29.5° latitude. Wind speeds were set at 20 m/s at a reference height of 10 m above ground or water surface. Eight SWAN simulations were conducted to investigate the influence of wind direction. Wind directions were varied from 0° (northerly) by 45° intervals through 315° (northwesterly). Significant wave heights (the average of the highest third of waves over a time interval) were queried and reported from the model output for submerged sites chosen as the closest point along the 1-m isobaths to each of the six erosion observation sites.

Statistical Methods.

All statistics were performed in the R statistical programming environment. Model assumptions for all statistical (3) tests were evaluated visually and quantitatively with residual, quantile-quantile, and leverage plots.

To describe and compare oiling and marsh plant mortality as a function of distance from the shore at reference and impacted sites, we fit standard logistic curves. Model fits were performed via generalized nonlinear least squares, and hypothesis tests to determine whether there were differences among sites were inferred from likelihood ratio tests (LRT). To describe the relationship between proportions of plant stems covered with oil and proportion of unoiled plant stems that consisted of green tissue, we fit this logistic curve and used the delta method to obtain estimates of uncertainty around parameters estimates.

To determine whether there were differences in the amount of belowground plant material alive in these two kinds of sites, we compared the proportion of *Spartina* rhizomes alive at 3-m and 15-m locations in reference and impacted sites. Specifically, we tested for an effect of site type, distance from shore, and the interaction between these two factors on probability of rhizome mortality by using a generalized linear mixed-effects model with a binomial family error distribution. To account for the hierarchical structure, we modeled the effects of site type (reference or impacted; $n = 3$ each), transect ($n = 5$) nested within site type, and location on transect ($n =$ 2–3 and 15 m) as nested random effects.

To test whether the abundances of each of three common invertebrate species differed between reference and impacted sites, we used a generalized linear mixed-effects model with a zero-inflated negative binomial distribution with site type (reference or impacted) treated as a fixed effect and transect nested within site replicate treated as random effects.

To determine whether plant and rhizome loss was associated with accelerated rates of shoreline erosion over time, we used linear mixed-effects models to compare the distances between poles placed at the shoreline (six per site) immediately after the oil spill and the location of the shoreline in successive weeks at both reference and impacted sites. Because of losses of poles from shoreline clean-up efforts and stochastic events, we had different amounts of replication among sites and over time. Therefore, we treated sites nested in time as random effects in the model.

We used a generalized additive model with site type treated as a categorical fixed effect and degrees from North as a continuous fixed effect to test whether there were differences in wave heights (based on output from the SWAN numerical wave model) at our impacted and reference sites.

To test whether there were differences in the success of transplants in the reference, impacted, and eroded intertidal regions, we used a generalized linear model with quasi-Poisson family error distribution (to account for overdispersion) to test for differences in stem densities.

Finally, to test whether there were signs of recovery of salt-marsh grasses, we compared the proportion of plants alive from the shoreline to 15 m inland for four time points spanning a period of 15 mo by using linear mixed-effects models with site nested in time treated as a random effect.

To determine whether there were differences in the concentration of PAHs at reference and impacted sites, we compared sediment loads from 3-m and 15-m locations in reference and impacted sites. Using these data, we tested for an effect of site type, distance from shoreline, and the interaction between these two factors with a linear mixed-effects model on log-transformed concentrations. To account for the hierarchical structure of these data (i.e., locations are nested within replicate reference and impacted site), we modeled replicates of each site type (reference or impacted; $n = 3$ each) as a random effect.

To determine how oil cover affected the survival probability of cordgrass, we compared plant survival at 0.0%, 40%, or 80% oil coverage by using a generalized linear model with a quasibinomial error distribution. We used the quasibinomial error distribution to account for overdispersion in these data (dispersion parameter for the quasibinomial was 1.12).

1. Mitra S, Bianchi TS (2003) A preliminary assessment of polycyclic aromatic hydrocarbon distributions in the lower Mississippi River and Gulf of Mexico. *Mar Chem* 82(3-4):273–288.
2. Booij N, Ris R, Holthuijsen L (1999) A third-generation wave model for coastal regions. 1. Model description and validation. *J Geophys Res* 104(C4):7649–7666.
3. R Development Core Team (2011) *R: A Language and Environment for Statistical Computing* (R Foundation for Statistical Computing, Vienna, Austria).

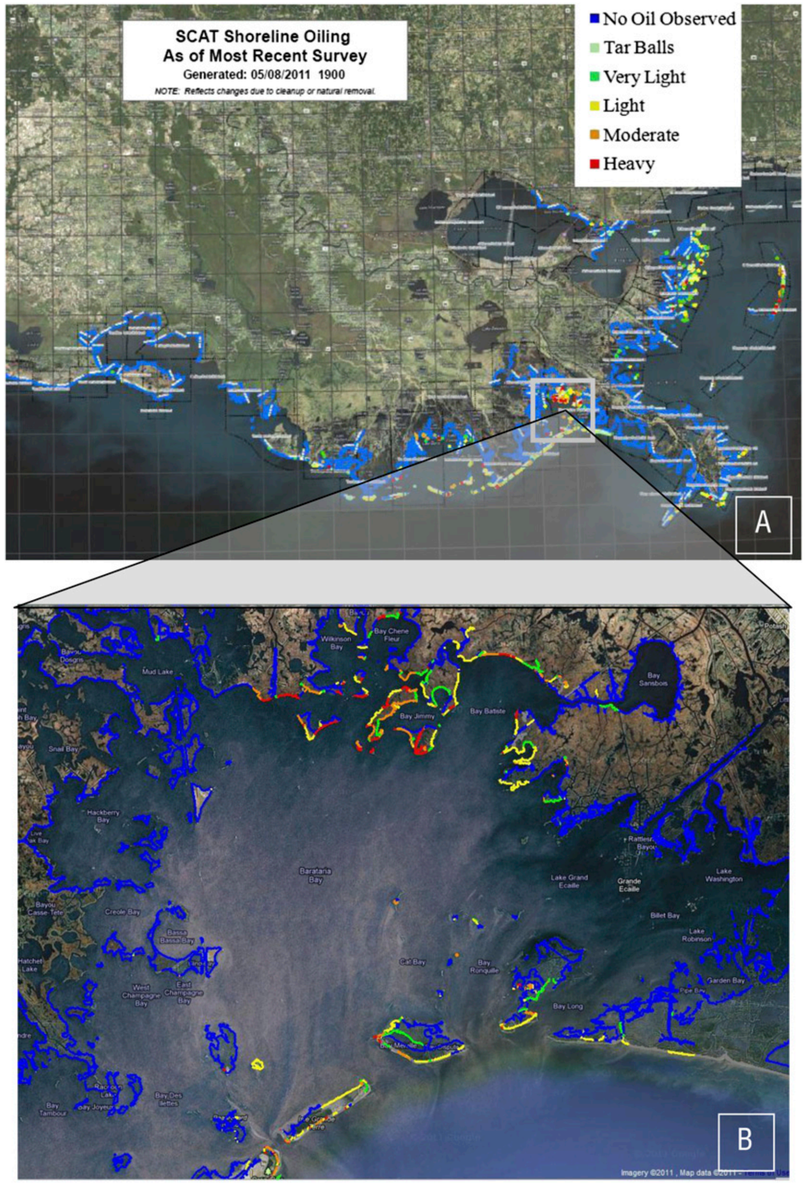

Fig. S1. Map of oil coverage along Louisiana coastline (*A*) and Barataria Bay (*B*) as of May 2011. Warmer colors represent more intense oil coverage. Map was accessed at http://www.noaa.gov/deepwaterhorizon/maps/ on May 8, 2011.

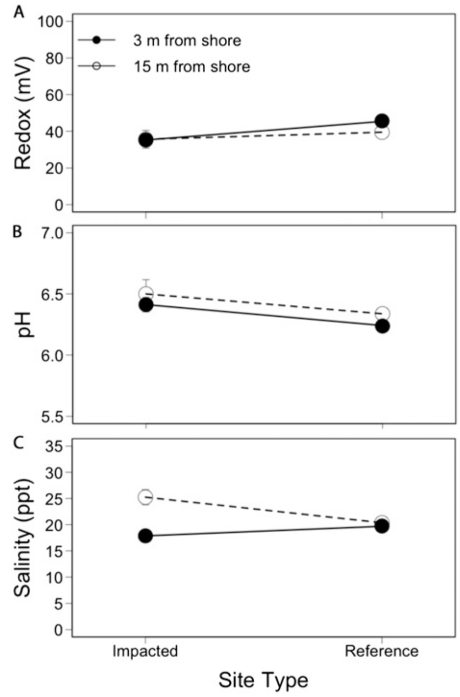

Fig. S2. Results from comparisons of abiotic characteristics often associated with die-offs. (*A*) There were no differences in the levels of redox at impacted and reference sites (LRT, $P = 0.12$) or at different distances from the shore (LRT, $P = 0.65$). (*B*) There were also no detectable differences in soil pH between site types (LRT, $P = 0.14$) or distances (LRT, $P = 0.49$). (*C*) There was a significant interaction effect between site type and distance from shoreline on measures of soil salinity (LRT, $P = 0.002$), with impacted sites 15 m from the shoreline having higher salinities than any other sites. This elevated salinity is unlikely related to observed die-off because die-off occurred predominantly in plants located <10 m from the shoreline (Fig. 1*C*).

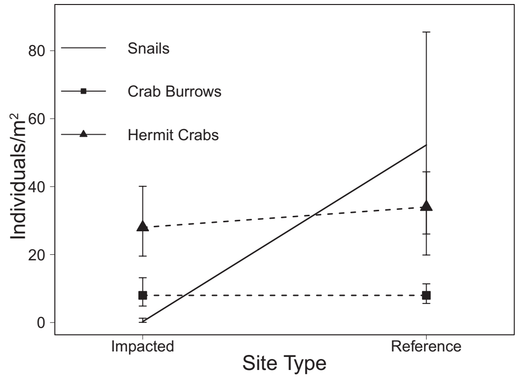

Fig. S3. Densities of the three common macroinvertebrate animals from surveys in reference and impacted sites in October 2010 at 3 m from the shoreline. There were significantly fewer snails at impacted sites (LRT, $P = 0.005$); however, there were no differences in the numbers of hermit crabs or crab burrows (used as a proxy for fiddler crab density). Data were analyzed with a generalized linear model with zero-inflated negative binomial error distribution. Data were collected from surveys at three reference and three impacted sites ($n = 5$) ($n = 15$ total). Means and SE bars are extracted from model fits.

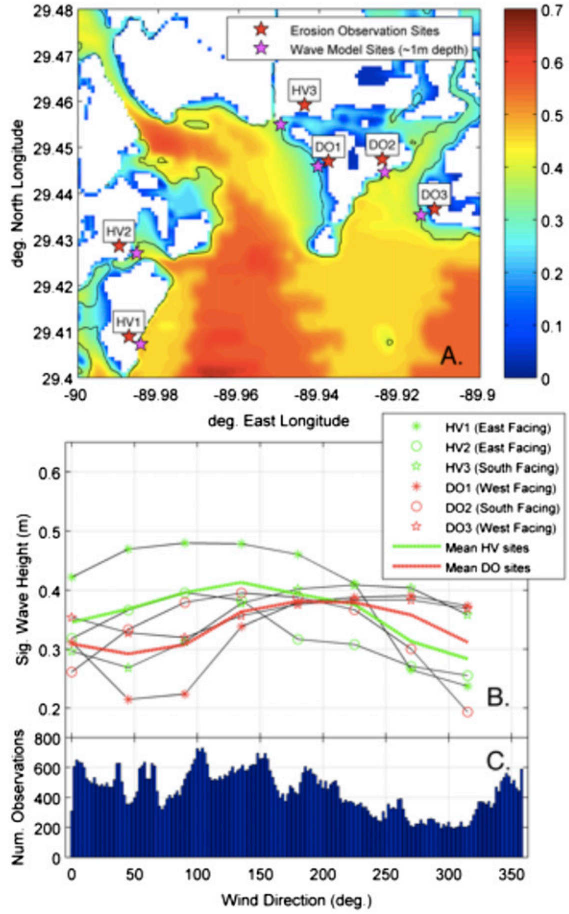

Fig. S4. Results of SWAN numerical modeling of wave height distributions within northern Barataria Bay region. (*A*) Color map showing spatial distribution of modeled significant wave height (color bar scale in units of meters) for a 20 m/s easterly wind field. Locations of erosion observation sites and adjacent wave model query sites are shown and labeled. Black line shows the approximate location of the 1-m bathymetric contour. (*B*) Line plot of modeled significant wave heights at each erosion observation site as a function of wind direction for a wind speed of 20 m/s from the east (90°). Red data markers represent sites that experienced significant vegetation die-off, and green data markers represents sites where vegetation was healthy. Also shown are the mean wave heights for all impacted and reference vegetation sites over the range of wind directions. (*C*) Histogram of observed wind directions observed at the National Oceanic and Atmospheric Administration/National Geophysical Data Center station GISL1, located along the southern boundary of Barataria Bay, for the 14-mo interval from October 2010 through November 2011. Data for the intervals April 3 to May 4 and August 7 to September 17, 2011, were unavailable. Waves appear to be influenced by orientation, fetch, and bathymetry, with larger waves occupying at east-facing shoreline sites. Wind fields oriented from northerly, easterly, and southerly directions result in larger wave heights at reference sites than at impacted sites. Because the majority of wind observations are from northerly, easterly, and southerly directions, it would be expected that the larger waves assailing east-facing sites should drive more rapid marsh shoreline retreat rates than witnessed at west-facing sites; this is not the case, however. In fact, wave heights were significantly higher on average at reference sites (df = 1, F= 5.32, P = 0.026) than at oiled sites, which casts doubt on the possibility that the higher marsh erosion rates documented at oiled sites were predisposed to rapid erosion as a consequence of their position in the landscape.

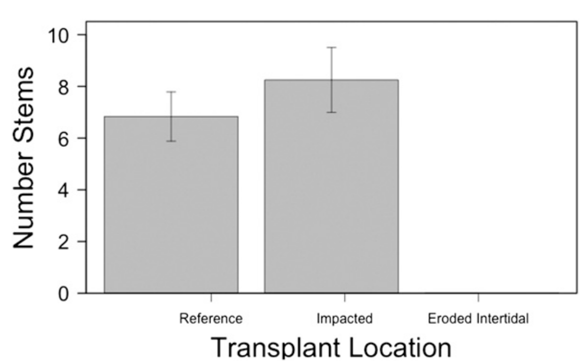

Fig. S5. Results from *Spartina alterniflora* transplant experiment in which healthy *Spartina* stems were planted in reference, impacted, and eroded intertidal regions of impacted sites. There was no difference in survival among the reference and impacted areas at these sites, but plants at these sites performed significantly better than plants in the eroded intertidal did (F = 57.26, P < 0.0001), with no stems persisting in the eroded intertidal zone.

Table S1. Coordinates of reference and impacted sites

Sites	Latitude	Longitude
Reference	29°24′535″N	89°59′239″W
	29°25′717″N	89°59′388″W
	29°13′402″N	90°00′443″W
	29°27′553″N	89°56′619″W
Impacted	29°26′819″N	89°56′264″W
	29°26′847″N	89°55′460″W
	29°26′196″N	89°54′681″W