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REVIEW

How Many Years Until Mangrove Ecosystems Recover from Catastrophic Oil Spills?

KATHRYN A. BURNS, STEPHEN D. GARRITY and SALLY C. LEVINGS
Address for reprints: S. Levings, Coastal Zone Analysis, Inc., P.O. Box 97, Sopchoppy, FL 32358-0097, USA

Kathryn A. Burns is a marine chemist at the Bermuda Biological Station for Research, 17 Biological Station Lane, Bermuda GE01. Stephen D. Garrity is a coastal marine ecologist and Sally C. Levings is a biologist with expertise in statistical analysis; both are with Coastal Zone Analysis, Inc., P.O. Box 97, Sopchoppy, Florida, FL 32358-0097, USA.

This short review article summarizes the results from long term assessment of an oil spill into a coastal fringe mangrove ecosystem in Panama. The study combined chemical and biological assessment methods to demonstrate that a time period of up to 20 years or longer is required for deep mud coastal habitats to recover from the toxic impact of catastrophic oil spills. This is due to the long term persistence of oil trapped in anoxic sediments and subsequent release into the water column.

Most oil spills are unplanned events involving tankers, oil rigs, storage tanks, pipelines, barges and other vessels (NRC, 1985). Between 1978–90, the largest sources of spilled oil worldwide were *1.* tankers and *2.* oil rigs or storage tanks (Welch *et al.*, 1991). In the tropics, there were at least 157 major (>1000 barrel) spills from ships and barges between January 1974 and 15 June 1990 (MMS, 1991). Of these, 26 occurred on the high seas, 99 in coastal or restricted waters, 12 in harbours and 20 at dock. A conservative estimate, made by adding spills in restricted waters involving groundings to those in harbours or at dock is that 83 (or 54%) of the major spills caused by shipping occurred near such potentially vulnerable ecosystems as coral reefs and reef flats, seagrass meadows, sand beaches, and mangrove forests.

Twenty-four of these spills occurred near coastlines in the Caribbean, four along peninsular Florida, two in the Bahamas and two off the east coast of Mexico (Fig. 1). There were also at least two spills in the open Caribbean, one in the open Atlantic, three each in the Orinoco and Amazon Rivers, and two at unspecified locales within the Caribbean region.

At least 19 refineries are located in coastal areas of the Caribbean and a pipeline for trans-shipment of Alaskan crude oil crosses Panama (Fig. 1) (Ward, 1990; Gundlach *et al.*, 1985). Recent spills have occurred at the Caribbean end of the pipeline and at two of the refineries (Cubit, pers. comm.; Bills & Whiting, 1991). Additionally, there are facilities in Bonaire, Curaçao, the Bahamas and Grand Cayman for transfer of Arabian crude oil from supertankers to smaller tankers (Fig. 1) (Cintron *et al.*, 1981).

Despite the frequency and continued probability of spills in nearshore, tropical waters, little work has been done on the effects of oiling on shoreline communities at low latitudes (Jacoby & Schaeffer-Novelli, 1990). In particular, although there is general agreement on the biological and economic value of mangrove forests, and on the vulnerability of mangroves and their biota to oiling, knowledge of the effects of oil on mangroves and associated species is extremely limited (Cairns & Buikema, 1984; Getter *et al.*, 1984; Gundlach & Hayes, 1978; Hatcher *et al.*, 1989; Lewis, 1983; Linden & Jernelov, 1980; Saenger *et al.*, 1983; Teas, 1983; Vandermuelen & Gilfillin, 1984).

In April 1986 more than 50 000 barrels of medium-weight crude oil spilled from a ruptured storage tank into mangroves, seagrasses, tidal flats and coral reefs on the Caribbean coast of Panama just east of the entrance

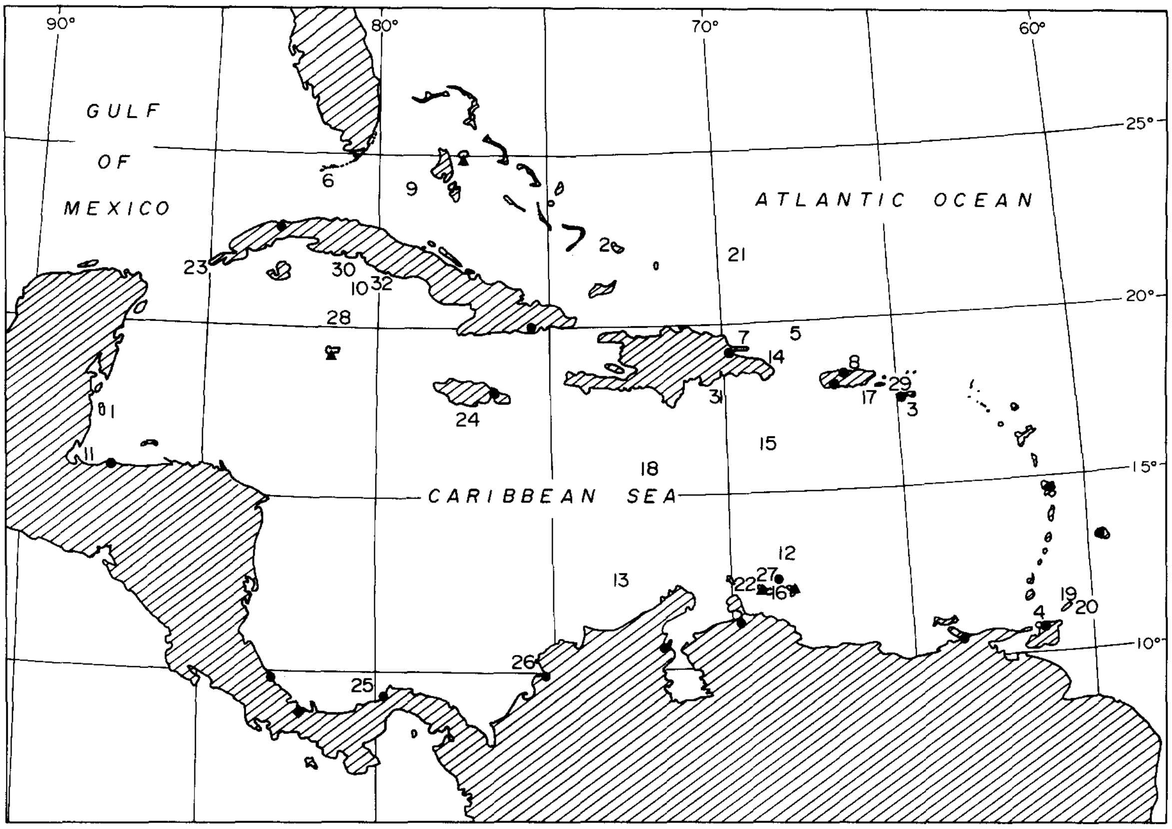

Fig. 1 Map of spill sites in the Caribbean, refineries and trans-shipment points for crude oil. Spills are numbered, refineries are circled, trans-shipment points are marked with triangles and the trans-Panama pipeline terminus is marked with a diamond.

to the Panama Canal. This spill, the largest recorded into coastal waters of the tropical Americas, occurred close to the Smithsonian Tropical Research Institute's Galeta Marine Laboratory. Because prior studies of the area existed, the spill afforded a unique opportunity to assess the effects of oil on a complex tropical coastal ecosystem (Cubit *et al.*, 1987; Jackson *et al.*, 1989; Burns & Knap, 1989; Garrity & Levings, 1992a). Intertidal mangroves, seagrasses, algae and associated invertebrates were covered by oil and soon died. Subtidal corals and infauna of seagrass beds also died. Oil soaked into the organically-rich sediments of mangroves and seagrass beds. The saturated sediments then acted as long-term reservoirs of oil, and were a major factor in continued reoiling of the coast for at least 5 years. We here summarize evidence of the unexpectedly long-term (>5 yr) persistence of the full range of aromatic hydrocarbon residues in the organically-rich muds of fringing mangroves. These residues persisted in the anoxic muds despite the relatively warm temperatures of the tropics.

For biological assessment, we focused on the mangrove (*Rhizophora mangle* L.) and the plants and animals that grow on submerged prop roots. The submerged trunks and prop roots of red mangroves, particularly those along the outer fringe of such forests, serve as living substrates for a diverse and abundant group of plants and animals (Rützler & Feller, 1987). Such attached species, or epibiota, not only contribute to the high productivity of the mangrove swamps, but also support many mobile, higher trophic level organisms, like fish and crustaceans. Thus, the submerged maze of roots and epibiota of the mangrove fringe adds structural and trophic complexity and increased diversity to the mangrove forest, and to the nearshore ecosystem as a whole. A series of detailed papers describe the spill, initial effects, study design, changes in the structure of the habitat and patterns of epibiotic cover, and hydrocarbon chemistry of the sediments and bivalves (Burns *et al.*, 1992; Burns & Yelle, 1992; Garrity *et al.*, 1992; Garrity & Levings, 1992a,b; Levings *et al.*, 1992). In this review, we summarize these results from the perspective of previous oil spill studies. First, we provide evidence for chronic oiling using records of washouts of weathered oil and hydrocarbon concentrations in bivalves (Burns & Smith, 1981). Second, we examine chronic toxic effects of oiling by comparing selected epibiota at oiled and unoiled sites within three habitats. Third, we measure the amount of habitat lost 5 years after the spill. Our results emphasize *1.* the necessary complexity of these analyses, and *2.* the long-term persistence of oiling and its negative effects.

This summary is intended to alert the environmental community that toxic effects of hydrocarbons will probably persist for at least 20 years in deep mud tropical coastal habitats affected by catastrophic oil spills.

Methods

The chemistry programme was designed to provide detailed characterization over time of the amount and composition of oil in mangrove sediments and in the tissues of bivalves living attached to mangrove roots. A combination of chemical methods were used, ranging from ultraviolet fluorescence (UVF) which is selectively sensitive to aromatic hydrocarbons and derivatives, through the more quantitative, but less specific, method of flame ionization gas chromatography (GC), to the extremely sensitive and selective technique of selected ion monitoring gas chromatography/mass spectroscopy (SIM-GC/MS) (IOC/UNEP/IAEA, 1991).

At least four replicate study areas were selected in three biologically-distinct mangrove habitats in or within 25 km of Bahía las Minas. Each habitat had a characteristic epibiotic assemblage on submerged roots. Open coast sites fronted the ocean along the inner margins of fringing reef flats; no bivalve was sufficiently abundant to use as an indicator species. Along banks of more sheltered channels and lagoons, mangrove roots were encrusted by oysters, *Crassostrea viginica (rhizophorae* morph.). Drainage streams were more sheltered and less saline than the other habitats. Mangrove roots were covered by the false mussel *Mytilopsis sallei.* Sediments were cored in 1986, 1989 and 1990, corresponding to $\frac{1}{2}$, $2\frac{1}{2}$ and $3\frac{1}{2}$ years post-spill. Three replicate cores per site were taken and the three layers at 0–2, 8–10, and 18–20 cm depth were combined. Sediments were again cored in May 1991, and oil oozing into the holes after the core was removed was collected for analysis. After sectioning cores in the field laboratory, each was examined for the numbers of live and dead mangrove roots. All roots $\geq$5 mm diameter were counted.

Release of weathered oil was measured quarterly using artificial roots (hardwood dowels), placed in the field for approximately $2\frac{1}{2}$ months (February 1987–May 1991). Dowels were hung vertically from the edge of the mangrove fringe, with the top of the dowel at approximately mean high water (N=5 dowels/site/replicate). Oiling was measured as *1.* presence/absence and *2.* percent cover (explained below). We recorded any oil slicks at each visit to our sites when conditions were suitable to see slicks and noted approximate thickness (e.g. iridescent sheen, black fluid).

Dissolved and suspended hydrocarbons were measured using bivalves (Levings *et al.*, 1992) from channels (oysters) and drainage streams (false mussels). At each site, a composite sample was taken by collecting a few bivalves from at least 20 haphazardly-chosen roots spread throughout the site. All 26 sites were sampled in December 1988 and May 1991, while a subset of two randomly-selected sites were sampled each quarter from November 1989–December 1990. A comparative bioaccumulation experiment conducted in May 1991 indicated that oysters accumulated approximately half as much total oil in their tissues as false mussels, when transplanted to the same site, as measured by UVF and GC methods (Levings *et al.*, 1992) but accumulated the same levels of individual aromatic hydrocarbons as measured by SIM-GC/MS (Burns & Yelle, 1992).

Mangrove roots were sampled along the mangrove fringe by randomly selecting roots that were submerged at least 20 cm but not yet embedded in bottom sediments. The length and diameter of each root was measured. The cover of epibiota on the root was measured as percent of space between mean high water and the longest root tip occupied by individual species (e.g. *Mytilopsis sallei,* live or dead), mixed species groups (diatoms, blue-green algae), oil or bare space.

Evaluation of structural damage to the fringe was a multi-stage process. In May 1991, each site was visited and the condition of the mangrove fringe recorded for each metre of the shore along the fringe. We recorded if red mangroves were present, and if the trees were intact (healthy canopy, <10% defoliated), dead (totally defoliated, dead wood), damaged (live, but with more than 10% bare branches or dead wood), new (immature saplings or seedlings growing in the fringe) or disturbed by humans (trees or roots cut for access to timber). In shoreline without fringing mangroves, we looked for physical evidence that mangroves had been present before the spill (i.e. for decaying root channels or stumps) and compared our findings with phototransects and field notes from each site beginning 3 months after the spill. This enabled us to verify that areas without fringing mangroves in 1991 had (or had not) been occupied by mangroves before the spill (units of linear metres of shoreline fringed with red mangroves). At each site the density of roots along the fringe was calculated by counting the number of roots that met our monitoring criteria (above) in 10 randomly-selected 0.25 m^2 quadrats positioned where mangrove fringe survived (intact, damaged and new fringe included).

These data were combined with estimates of root surface area calculated from root length and diameter to generate an estimate of the percentage of the fringing mangrove habitat remaining 5 years after the spill. We assumed that the percent of the shoreline fringed with red mangroves at unoiled sites was the approximate amount of shore expected to be fringed with mangroves at oiled sites. We then estimated how many submerged prop roots that met our monitoring criteria were found within the fringe at oiled as compared to unoiled sites. Surface area/root was calculated assuming that roots were approximately cylindrical in shape. These values were combined and standardized to a 100 m length of shoreline to estimate *1.* the number of submerged roots and *2.* the number of m^2 of submerged root surface at oiled as compared with unoiled sites (units of numbers of roots/100 m shoreline or m^2 submerged root surface/100 m shoreline). This was an underestimate of the actual amount of habitat along the fringe (because not all roots met our monitoring criteria) but was not differentially biased among sites or habitats.

Repeated measures analysis of variance (ANOVAR) was applied to arcsine-transformed percent cover data (Green, 1979). Among-year comparisons were calculated using Fisher's least significant difference (LSD) methods (Milliken & Johnson, 1984). A special analysis of variance procedure was applied to bivalve hydrocarbon samples. Because composite samples were collected at each site (n=1 sample/site/quarter), we had no measure of variation within sites. We used the mean square for the site–date interaction as the error term to test for the main effects of site and date; data were weighted by the number of sites sampled/quarter (Proc GLM, SAS, 1988). This gave a conservative analysis of variation in tissue burdens among sites and times (R. Green, pers. comm.).

Results

Sediment hydrocarbon chemistry

Initial weathering processes removed labile components such as n-alkane hydrocarbons from oiled surface sediments within 6 months after the spill (Burns *et al.*, 1991). Degradation of oil to this stage in temperate salt marshes requires on the order of 2 years (Burns & Teal, 1979; Atlas *et al.*, 1981). Total oil concentrations remained high (up to 20% of dry wt) in mangrove surface sediments through at least 4 years after the spill and more oil was transported to deeper layers in later years (Burns & Yelle, 1992; Burns *et al.*, 1992). There was year to year variability at individual sites due to patchy distributions of oil residues within the sediment column. In most sediments, even in 1986, hydrocarbon patterns determined by GC contained up to 99% unresolved components, indicating microbial decomposition had removed resolved alkanes. Oil is composed of a mixture of alkyl substituted and parent hydrocarbons which is too complex to resolve into individual peaks by high resolution GC. Thus a characteristic unresolved mixture of hydrocarbons remains in degraded petroleum residues.

Residual pools of oil in mangrove sediments were fluid enough to flow out when sediments were cored or disturbed 5 years after the original spill. Most oil oozing into core holes in May 1991 was highly degraded, like residues extracted from sediment cores. However, one oiled stream contained a surprisingly fresh oil residue with the full suite of n-alkanes preserved (Fig. 2). The patterns of individual polynuclear aromatic hydrocarbons (PAHs) and triterpane biomarkers determined by SIM-GC/MS confirmed that this oil was the crude mixture spilled in 1986 (Burns *et al.*, 1992). Thus it must have been preserved relatively intact from the 1986 spill, probably in an anoxic layer of sediment.

Evidence for chronic reoiling of mangrove habitats

Oil was deposited on dowels each quarter from 1987–91, but the frequency and amount of oiling differed among habitats (Fig. 3). The frequency of reoiling on the open coast was affected by site location, with the two sites nearest the refinery approximately twice as likely to be oiled (some oil in 57% or more of samples) as the two more distant sites (some oil in <27% of samples). In contrast, secondary reoiling on the open coast did not involve large amounts of oil. All seven episodic oilings where mean percent cover was >10% at any one site occurred during rainy season, and probably resulted from washouts of oil from intertidal sediments.

Pulses of residual oil were frequent in channels and lagoons. One or more sites were oiled in 16 of 17 quarters (except November 1990). The greatest amount of oil was found from February–August 1989, at three of the five sites. This appeared to be related to the collapse and cutting of dead mangroves in the central and eastern wings of the bay and to replanting efforts by

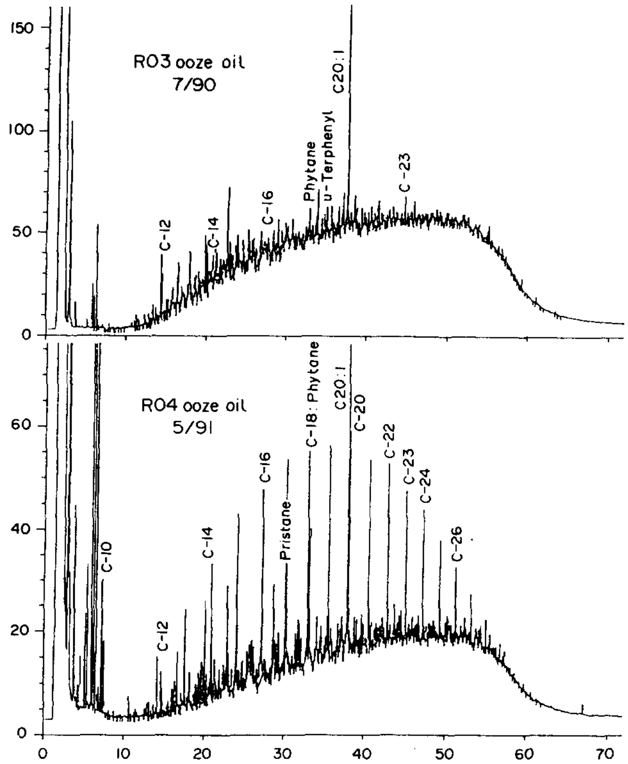

Fig. 2 Gas chromatogram traces of the hydrocarbon patterns in oil oozing from sites RO3 and RO4 in May 1991. Top trace is RO3 which shows a degraded oil pattern spanning the C12 to C32 n-alkane elution range, but most resolved alkane peaks have disappeared leaving the unresolved complex mixture of petroleum derived hydrocarbons. Bottom trace is from RO4 showing the same elution range but the preservation of marker alkanes. (Triterpane and aromatic hydrocarbon patterns determined by SIM-GC/MS confirmed these oils were related to the mixed Venezuelan Mexican Isthmus Crude spilled in 1986.) (See Burns *et al.*, 1992 for details.)

the refinery (Teas *et al.*, 1991; Garrity *et al.*, 1992), and subsequent washouts from oil-saturated sediments (Burns *et al.*, 1992).

In drainage streams, all dowels were oiled at three of the four sites until November 1990 (4.5 years post-spill). The proportion of dowels oiled was higher and less variable over time than in the other two habitats, except for one stream, where a few dowels were unoiled as early as May 1988. For all sites combined, percent cover on dowels was usually >50% through August 1989. Little oil was recorded on dowels in 1991.

The presence or absence of slicks off each study site was recorded at least once each quarter, weather and waves permitting. Slicks at oiled sites were observed in 92.7% of 316 observations (all habitats combined, August 1987–May 1991). Over time, slicks were found less frequently on the oiled open coast, and were characterized as iridescent or silver sheen rather than black oil. Slicks were seen in five of nine visits in May 1991. In channels and lagoons, the frequency of slicks did not decline (96% of 22 visits in May 1991), and some black oil was seen in May 1991. In drainage streams, oil slicks were seen on 128 of 129 visits between August 1987 and May 1991, and ranged from black oil to iridescent patches.

Bioaccumulation of oil leaching from sediments

Water soluble/suspended fractions of crude oil were

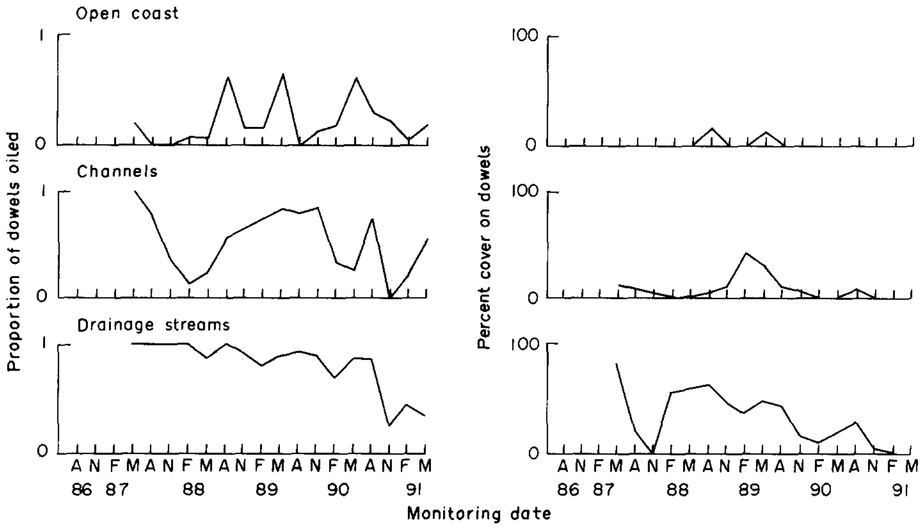

Fig. 3 Secondary oiling on dowels: February 1987–May 1991. Among-site mean proportion of dowels with at least some oil on their surfaces (left panels) and among-site mean percent cover of oil (right panels). Sample size = 1–5 sites/quarterly monitoring.

accumulated by bivalves between 1986–91 (Fig. 4). Tissue levels in both species remained high at oiled sites throughout the study, but highest concentrations were recorded in dry seasons (February). Oysters consistently had approximately half as much total oil in their tissues as false mussels when measured by UVF or GC methods. False mussels and oysters were transplanted from clean sites into the same oiled stream. Whether oil content was determined by GC or by UVF, oysters accumulated only half the amount of total oil that false mussels did under the same exposure conditions. Thus a bioconcentration factor (calculated as [bivalve]/[water]) for *Crassostrea* would be half of that calculated for *Mytilopsis* (Burns & Smith, 1981). This observation on the quantitative difference between the two species of indicator bivalves was critical for interpretation of the field data. There were no significant differences among streams or among channels in tissue burdens of oil averaged over 5 years (Table 1). However, differences among sites approached significance in streams. This was due to two periods when oil concentrations were >50 μg g^{-1} EOM (extractable organic matter) at two sites; these very high values were probably caused by particulate oil in gut contents rather than indicating a tissue equilibrium level (Burns & Smith, 1981). It is thus unlikely that there were any consistent differences in the amounts of dissolved/suspended oil over time in channels or streams.

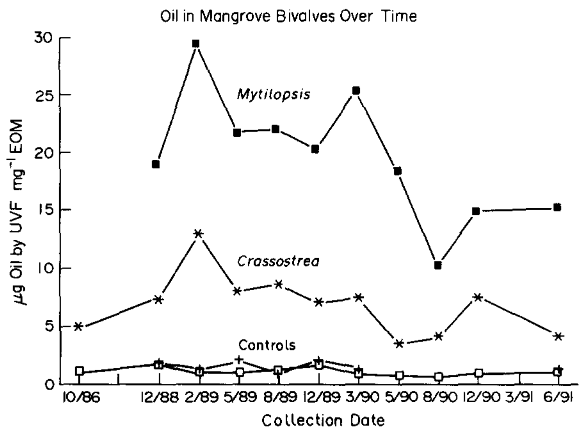

Fig. 4 Concentration of oil (determined by UVF analysis) in bivalves Sept. 1986–June 1991 (n=2–5 oiled sites per point). Units are μg oil/mg EOM. (See Levings *et al.*, 1992 for details.)

TABLE 1

Summary ANOVA on levels of hydrocarbons in bivalves.

Source	Channels—oysters DF	F value	Probability	Drainage streams—false mussels DF	F value	Probability
Site	4	1.25	0.3129	3	2.91	0.0584
Date	9	2.06	0.0721	9	0.85	0.5787
Error (site-date)	26			21		

See text for details of analytical procedure.

The time series data in Fig. 4 suggest roughly similar amounts of soluble oil fractions were present over time in oiled channels and streams. Significant levels of potentially toxic hydrocarbons and derivatives were bioavailable through at least the fifth year after the oil spill.

Polyaromatic hydrocarbons (PAHs) accumulated by bivalves were similar in composition to those in oil leaching from or resuspended from sediments (Fig. 5). Oysters and false mussels accumulated the whole suite of parent and substituted PAHs in the naphthalene through chrysene elution range. The most abundant PAHs in bivalve tissues were generally the dibenzothiophene series, followed by the phenanthrenes and chrysenes. This agrees with the UVF spectra from bivalve extracts which show maximum fluorescence intensity at wavelengths characteristic of three and four ringed aromatics. Oysters at site CO2 contained more C1 and C2 phenanthrenes than sediments, suggesting the more soluble C1 and C2 compounds were being depleted in sediments, leached into the water and accumulated by bivalves. The most residual PAHs in sediments were the higher alkyl substituted DBTs, phenanthrene/anthracene and the pyrene/fluoranthene series.

Reductions in epibiota

Each of the three habitats studied had a different

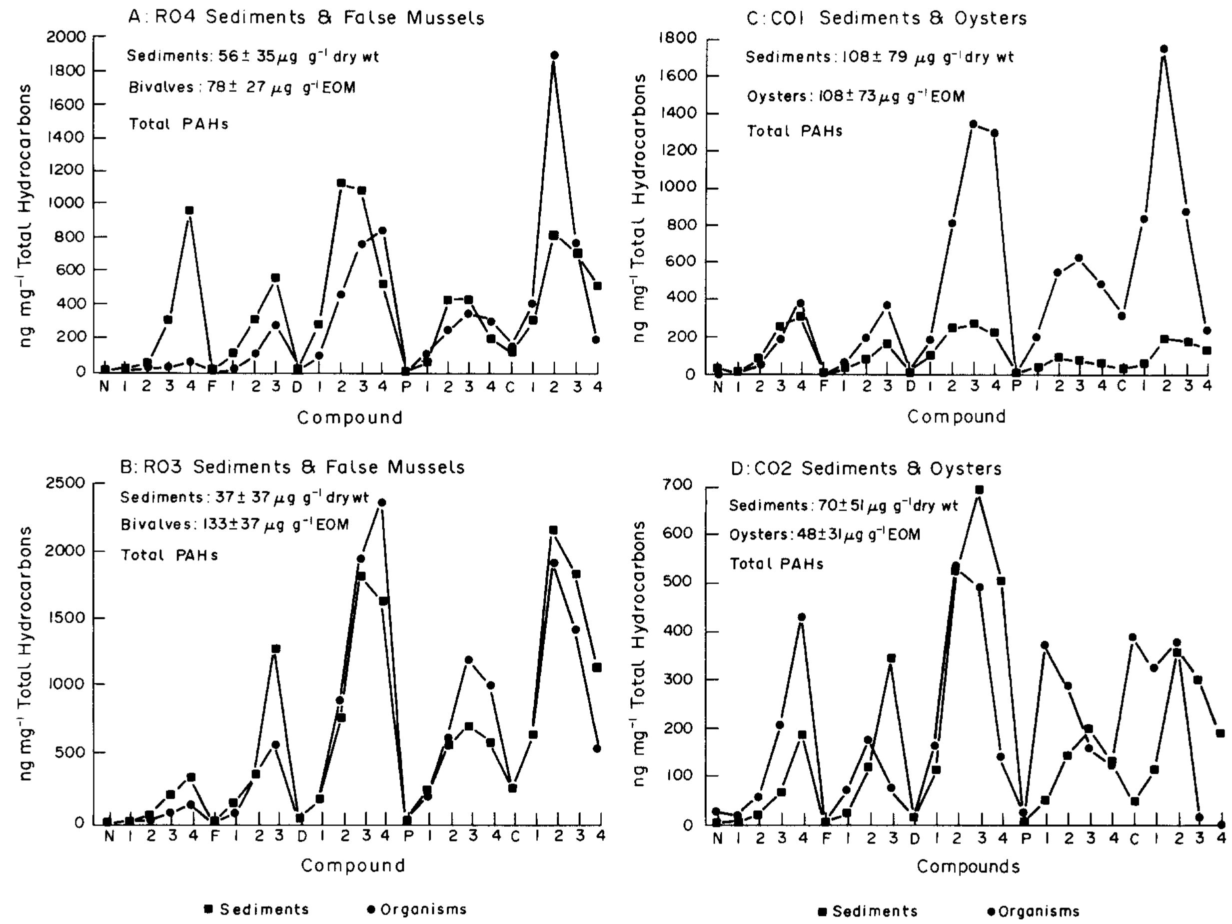

Fig. 5 Sample plots of the relative composition of individual aromatic hydrocarbons in two of the most heavily oiled CO and RO sites, in ng normalized to total mg of oil estimated by GC. The hydrocarbons are grouped to ring structure for Naphthalenes (N), Fluorenes (F), Dibenzothiophenes (D), Phenanthrenes/ Anthracenes (P), Chrysenes/Benzanthracenes (C). Numbers following letters are for the sum of isomers with C1–C4 alkyl substitutions. Sediments averaged over the 0–2 and 8–10 cm layers (data for 1989 and 1990 combined) compared with average concentration in bivalve tissues over the time period 12/88 to 3/90. Averages plus standard deviations of n = 4 samples for total PAHs in µg g^{-1} dry wt for sediments and µg g^{-1} EOM for bivalves indicated in each panel. (See Burns & Yelle, 1992 for details.)

assemblage of plants and animals living attached to submerged prop roots. On the open coast, sessile invertebrates that grew on submerged prop roots included sponges, corals, anemones, tunicates, bryozoans, vermetids and hydroids (barnacles and bivalves excluded from this analysis). After the spill, roots on the open coast were coated with oil. Subsequently, cover dropped to <5% in the first year after oiling, with almost complete disappearance of most groups except aborescent hydroids and bryozoans (Fig. 6). Cover varied at unoiled sites, but averaged ~10% from 1986–91. There was a significant main effect of oiling (Table 2). Differences between oiled and unoiled sites were significant for years 2–4 after the spill. Cover at oiled sites increased over time, and oiled and unoiled sites did not differ significantly in sessile cover between August 1990–May 1991 (year 5, ~7% cover oiled vs. 12% unoiled, Fisher LSD tests). All groups were present in 1991, but corals, anemones and tunicates were still rarer at oiled than unoiled sites. Only arborescent hydroids and bryozoans approached levels characteristic of unoiled sites.

In August 1986, intertidal portions of prop roots along oiled channels within Bahia las Minas were heavily coated with oil. Oysters were severely affected (Fig. 6). In August 1986, mean abundance was 27% cover, and dead oysters averaged 22% cover on roots. In the next 9 months, oyster abundance at oiled sites plummeted to 6% cover. The cover of dead individuals decreased to ~11% cover as valves detached from roots or were covered by oil or other organisms. At two unoiled sites, mean abundance of oysters averaged 43% cover, and dead oysters covered ~ 3% of root surfaces. From the second to the fifth year after the spill, oyster cover increased slowly at oiled sites, fluctuating at ~15% cover from 1988–91. At unoiled sites, cover of oysters varied between 14–24% of root surfaces over the same time period. Differences between oiled and unoiled sites were significant through year 5 post-spill (Table 2, two ANOVARs shown, due to site changes in 1988, see Levings *et al.*, 1992 for details).

In drainage streams, roots were thickly and repeatedly coated with oil. False mussels disappeared from oiled streams within 3 months after oiling and few had returned 5 years later (Fig. 6). False mussels were significantly less abundant at oiled than unoiled streams (Table 2, LSD tests significant, 1987–91). There were slight increases in mussel abundance in two oiled streams in 1991. False mussels (and other organisms) recruited onto some roots that had entered the water

TABLE 2
Summary ANOVAR results for major groups of epibiota by habitat.

	Open coast—sessile invertebrates	Channels—oysters, years 2-5 post-spill	Channels—oysters, years 3-5 post-spill	Drainage streams false mussels
Between:				
Oil	0.0106	0.0116	0.0038	0.0005
Within:				
Year	0.0001	0.0384	0.0191	0.0001
Year-oil	0.0144	0.0271	0.5660	0.0008

Probability levels for ANOVAR, arcsine-transformed, percent cover data, years 2-5 post-spill. For oysters, two analyses are presented. Year: 2-5 post-spill, n = 2 unoiled, 5 oiled sites. Years 3-5 post-spill, n = 3 unoiled, 5 oiled sites. See Levings *et al.* (1992) for details.

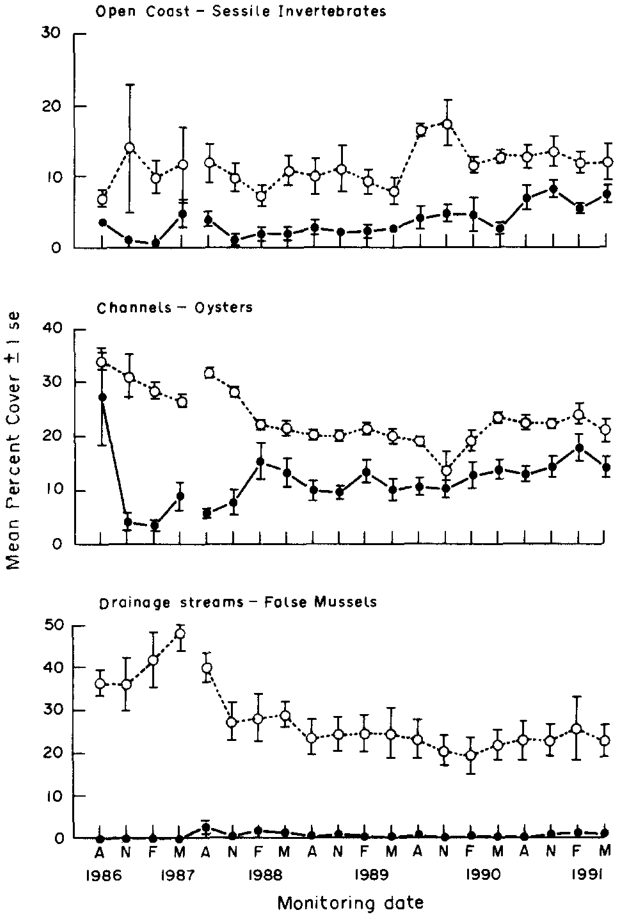

Fig. 6 Mean percent cover of major sessile animals by habitat. Among-site means ± one standard error. Open symbols = unoiled sites, filled symbols = oiled sites. Quarterly monitoring began in August 1986, 3 months after the spill. Lines are broken where site locations were changed 1 year after monitoring began. Note that scales differ among panels.

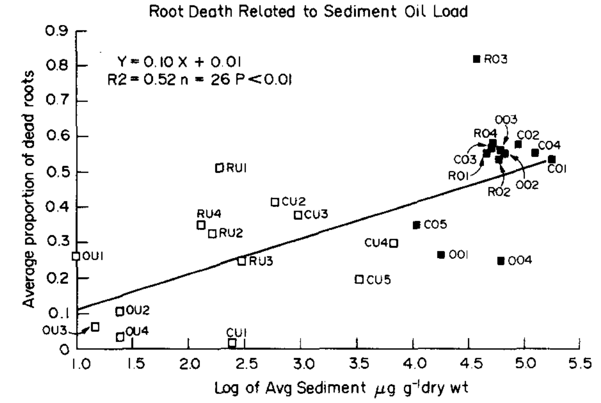

Fig. 7 Scatter plot and regression line of the log of average concentration of oil in sediments vs. the average proportion of dead roots in sediment cores. Dark squares were sites heavily oiled in 1986; open squares are sites that appeared to be unoiled in 1986 but which could have been subject to prior spills. Sites are coded by number, habitat and oiling condition. 'OO' open coast, oiled; 'OU' open coast, unoiled; 'CO' channel, oiled; 'CU' Channel, unoiled; 'RO' stream, oiled; 'RU' stream, unoiled. (See Burns *et al.*, 1992 for details.)

that year. This was the first indication of recovery in 5 years of study. However, long term survival of mussels settling at these sites has yet to be demonstrated.

Structural changes to the mangrove fringe after oiling

Structural changes to the fringe are discussed in detail in Garrity *et al.* (1992); we briefly summarize results here. On the open coast, an average of 13.3% less shoreline was fringed with mangroves at oiled compared with unoiled sites and there were 24.3% fewer submerged roots where mangroves survived (estimated unoiled total: 1534 roots 100 m^{-1} shore, oiled total: 1075 roots 100 m^{-1}). When average decreases in the submerged area/root (22 cm^2 less area/root at oiled sites) were added to this estimate, there was 65.8 m^2 of submerged root area at unoiled sites as compared with 43.8 m^2 at oiled sites (66.6% of that at unoiled sites).

In channels and lagoons, approximately 23.5% of the fringe was lost and root density was 20% lower at oiled than unoiled sites (estimated unoiled total: 1840 roots 100 m^{-1} shore, oiled total: 1132 roots 100 m^{-1}). There were no significant differences in root size. Total surface area 100 m^{-1} shore was estimated at 42.1 m^2 in oiled channels and 67.7 m^2 in unoiled channels (62.1% of that at unoiled sites). Results were most striking in drainage streams, where an average of 56% of the fringe was lost and roots were 16.2% less abundant at oiled than unoiled sites (estimated unoiled total: 1445 roots 100 m^{-1} shore, oiled total: 602 roots 100 m^{-1}). Roots had smaller surface areas at oiled than unoiled sites (379 cm^2 vs. 596 cm^2/root). We estimated that unoiled sites had 86.2 m^2 100 m^{-1} shore while only 22.8 m^2 100 m^{-1} shore was submerged at oiled sites. This translates to a 74% reduction in the fringe habitat available for epibiotic settlement.

Effects of oiling on mangrove roots

Evidence for the prolonged effect of sediment oil on mangrove trees comes from data on the proportion of live vs. dead roots embedded in sediment cores. Oil content was patchy both horizontally and vertically within sites. Because only a few cores had high concentrations at 18–20 cm depth, sediment concentrations of oil in UVF units in the 0–2 and 8–10 cm layers were averaged over 1989 and 1990 and plotted (log transformed) against the average proportion of dead roots in 1989 and 1990 (Fig. 7). The relationship showed the clear distinction between heavily oiled sites and the cleanest controls. However, there also appeared to be an intermediate concentration range showing possible impact. The unoiled streams and channels (RU1-4, CU3-5) were all within the Bahía las Minas area. Not only was this area affected by the 1986 spill of crude oil, but the wreck of the barge *Witwater* in 1968, carrying heavy fuel oil and diesel, killed extensive areas of mangroves nearly 20 years earlier (Rützler & Sterrer, 1970; Duke & Pinzón, 1992). Traces of oil were found in the deep layers of the core from CU3 in 1986; this site had been oiled by the *Witwater* spill. Also, CU2, which is in Margarita Lagoon, outside of Bahía las Minas, showed relatively high levels of light fuel oil in 1986; this oil must have come from a separate, isolated spill. One result of the history of repeated oil pollution events along this coast appears to be an effect on the condition of mangrove roots related to contaminant loading and detectable up to 20 years after major spills.

Discussion

The Galeta oil spill was a single, very large spill from a ruptured storage tank in a refinery complex. Oil from the spill was trapped in the deep muddy habitats of the contiguous mangrove ecosystem. This trapped oil was resuspended and circulated throughout Bahía las Minas for at least 5 years after oiling. Thus a point-source spill became a source of chronic oiling. Likely sources of the oil we recorded included oil gradually released from pools of oil in fill under and around the collapsed storage tank in addition to oil leaching from the intertidal sediments.

Artificial roots and records of slicks measured the amount of visible, weathered oil floating through a habitat; bivalves accumulated dissolved/suspended oil. Patterns of resuspension determined by these two methods differed. Weathered oil was deposited episodically on dowels; amounts of oil on dowels decreased over time, with little deposited in any habitat in 1991. Similarly the number and thickness of slicks declined over time, especially on the open coast, but less so in channels or streams. This suggests that *1.* seasonal variation in weather, water levels and tidal flushing controlled the amount of oil released, and *2.* the amount of weathered oil being released may finally have been declining 5 years after the spill. It is possible that erosion and subsequent release of trapped oil were slowing as mangroves became reestablished at oiled sites and developed a root mat that could stabilize the sediments.

In contrast, oil content in bivalve tissues in channels and streams was high and variable through 5 years post spill. There were no differences over time in average oil content of bivalves among oiled streams or among oiled channels. Accounting for the difference in bioaccumulation patterns between the two indicator species, our data suggest that *1.* roughly similar amounts of dissolved/suspended oil circulated in streams and channels at any one time, and *2.* variations in amounts of dissolved/suspended oil may have been related in both habitats. That is, factors which increased tissue burdens in streams also acted in the same way in channels. Unfortunately, no bivalve was abundant enough to sample on the open coast, so we have no estimate of dissolved/suspended oil concentrations there. The number and thickness of slicks declined relatively quickly on the open coast as compared with the more sheltered habitats and little oil was recorded on dowels.

Based on bivalve tissue concentrations, the amounts of dissolved/suspended oil may be declining more slowly, or not at all, when compared with visible tarry residues recorded on dowels or in slicks. The different patterns of decline in visible oil residues as compared with tissue content suggest that the processes which release the two types of residues are partially uncoupled. Erosion was probably the main factor releasing tarry oils trapped in sediments; both erosion and diffusion processes would release dissolved/suspended oils from sediments. Thus visible oil may disappear long before sediments are depleted of toxic hydrocarbon compounds (Burns & Yelle, 1992). Our data suggest that levels of suspended oil 5 years post spill would be high enough to reduce the growth and reproductive rate of the bivalves (Bayne *et al.*, 1982; Widdows *et al.*, 1990).

Evidence that the composition of residues leaching from the sediments and into the water column had begun to change detectably in year five came from dissection of the regression correlation between oil content in bivalve tissue estimated by UVF and GC analysis. We showed a highly significant correlation between these estimates for the entire data set spanning December 1988 through June 1991. Table 3A shows that the slopes of the individual regression correlations between UVF and GC oil determinations were similar until August 1990. Samples collected November 1990–June 1991 showed a change in slope in the regression line. To test the statistical consistency of the relationships, we used Analysis of Covariance (ANCOVA), testing first for the equality of slopes among years. There was significant heterogeneity of slopes (Table 3B). However, the scatter was great enough that there were no significant differences among any pair of years using the Tukey-Kramer method for unplanned comparisons (Sokal & Rohlf, 1981). Despite this lack of resolution between years, it appears there was a change in the relationship between the oil estimates based on UVF and GC analysis in the last group of samples when the regression slopes changed from the range of 0.727 to 0.745 and dropped to 0.454. This means that samples in the last group were relatively more fluorescent and indicates a change in the composition of oil accumulated by the bivalves in year five. This change could be from a difference in the composition of PAHs accumulated or from a larger percentage of the UVF signal due to fluorescent derivatives rather than parent hydrocarbons. We could demonstrate no difference in PAH pattern significant enough over time to cause this

TABLE 3A

Regression equations and estimates of standard error of slopes for the UVF (X) and GC (Y) determinations of oil residues in bivalve tissue showing change in slope over time.

Sampling interval	Regression equation	Std error of slope	N	R^2	P value
December 1988 to February 1989	$Y = 0.7446 X + 0.16$	0.0902	32	0.82	<0.001
May 1989 to December 1989	$Y = 0.7275 X + 0.02$	0.0914	33	0.63	<0.001
March 1990 to August 1990	$Y = 0.7378 X + 0.02$	0.0748	41	0.68	<0.001
November 1990 to June 1991	$Y = 0.4541 X + 0.07$	0.0842	44	0.45	<0.01

Concentrations are transformed to Log of μg oil/mg EOM. N is the number of samples, R^2 is the correlation coefficient, P is the level of significance.

TABLE 3B

Analysis of covariance: Log UVF vs. Log GC estimates over time using the four groups above.

Source	DF	Sum of squares	F value	Probability
Log UVF	1	19.168	252.69	0.0001
Group	3	1.347	5.92	0.0008
Log UVF–group	3	0.642	2.82	0.0410
Error	141	31.852		

divergence. This indirect evidence for a greater percentage of bioaccumulated residues due to hydrocarbon oxidation products over time is consistent with more direct evidence for this process obtained in other spill studies (Burns, 1992).

There were persistent differences in bivalve cover in oiled and unoiled channels and streams 5 years after the spill. Only on the open coast had cover of the major animal group, sessile invertebrates, returned to approximately the same level at oiled and unoiled sites (differences significant for first 4 years), but some groups were still rare. Chronic effects of the spill were probably related to *1.* changes in the physical structure of the mangrove fringe, *2.* the continued presence of toxic hydrocarbon compounds in the environment, and *3.* other, yet unstudied, ecological interactions (i.e. changes in the density or activity of predators). The cover of oysters in oiled channels increased over time, but in contrast, few false mussels were found in any oiled stream 5 years after the spill. False mussels may have been slower in returning, in part, because they accumulated more total oil compounds and may have had a correspondingly reduced physiological ability to grow and reproduce.

Finally, reductions of the number and size of submerged mangrove roots reduced the area that could serve as settlement surfaces for epibiota. Since productivity is positively related to the area of hard substrate for settlement, there was a direct reduction in the productivity of the mangrove fringe, independent of any persistent toxic effects of dissolved oil compounds. These findings emphasize the complexity of the analyses necessary to follow damage and recovery from this spill and similar ones into biogenically structured habitats.

Summary

The Galeta oil spill was catastrophic. Five years after the spill the effects of oiling were visible both as *1.* reduction of the area of substrate in the mangrove fringe in the three habitats examined and *2.* reductions of the cover of attached animals in two of three habitats. Organisms within the bay were still exposed to high levels of dissolved/suspended oil compounds, although visible tarry residues were declining.

Hydrocarbon chemistry confirmed the long-term persistence of crude oil residues in the deep muds of mangrove ecosystems. The initially-rapid degradation of alkane fractions observed in 1986 was not followed by high degradation rates of the more residual aromatic hydrocarbons. Pools of trapped oil maintained surprising consistency in composition measured by UVF, GC and SIM-GC/MS analyses. Trapped oil continually leached from mangrove sediments into coastal waters and was bioaccumulated by encrusting bivalves for 5 years after the spill, when our observations ended. Reoiling associated with erosion of heavily oiled sediments caused chronic oil pollution in the coastal waters. The most residual aromatic fractions appear to be the dibenzothiophene, chrysene and phenanthrene series. Continued high concentrations of these relatively toxic fractions, even in sediment residues that appear highly weathered by GC, indicates their life span in mangroves is much longer than 5 years.

Even with the large database we have acquired, many questions remain unanswered, and ecosystem recovery is far from complete. We know nothing about transmission of possible long term effects up the food chain or on local fisheries dependent in part on the species of the mangrove fringe community. The interactions between the effects of structural changes to the habitat and continued toxic effects of hydrocarbons and related oxidation products will be critical in understanding recovery processes over the long term.

The impact of this oil spill was clearly discernable despite the history of repeated oil pollution events along this coast. Medium weight hydrocarbons persisted for over 20 years in sediments heavily oiled by the West Falmouth spill (Teal *et al.*, 1992) and oil hydrocarbons were detectable in mangrove sediments in Puerto Rico for at least 20 years after a tanker grounding (Corredor *et al.*, 1990). In previously reported studies of oil spills, biological and chemical measurements were not made more than 5–7 years post-spill. The time period between 5 and 20 years post-spill is when the effects of oiling in muddy coastal habitats grade from acute and severe toxicity into sublethal stress and eventually into nearly non-detectable effects. The long time scale of these impacts accentuates the need for careful protection of these

important habitats. Study of the Bahía las Minas spill still has the potential to answer many remaining questions regarding time scales and processes involved in environmental recovery from catastrophic oil spills.

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