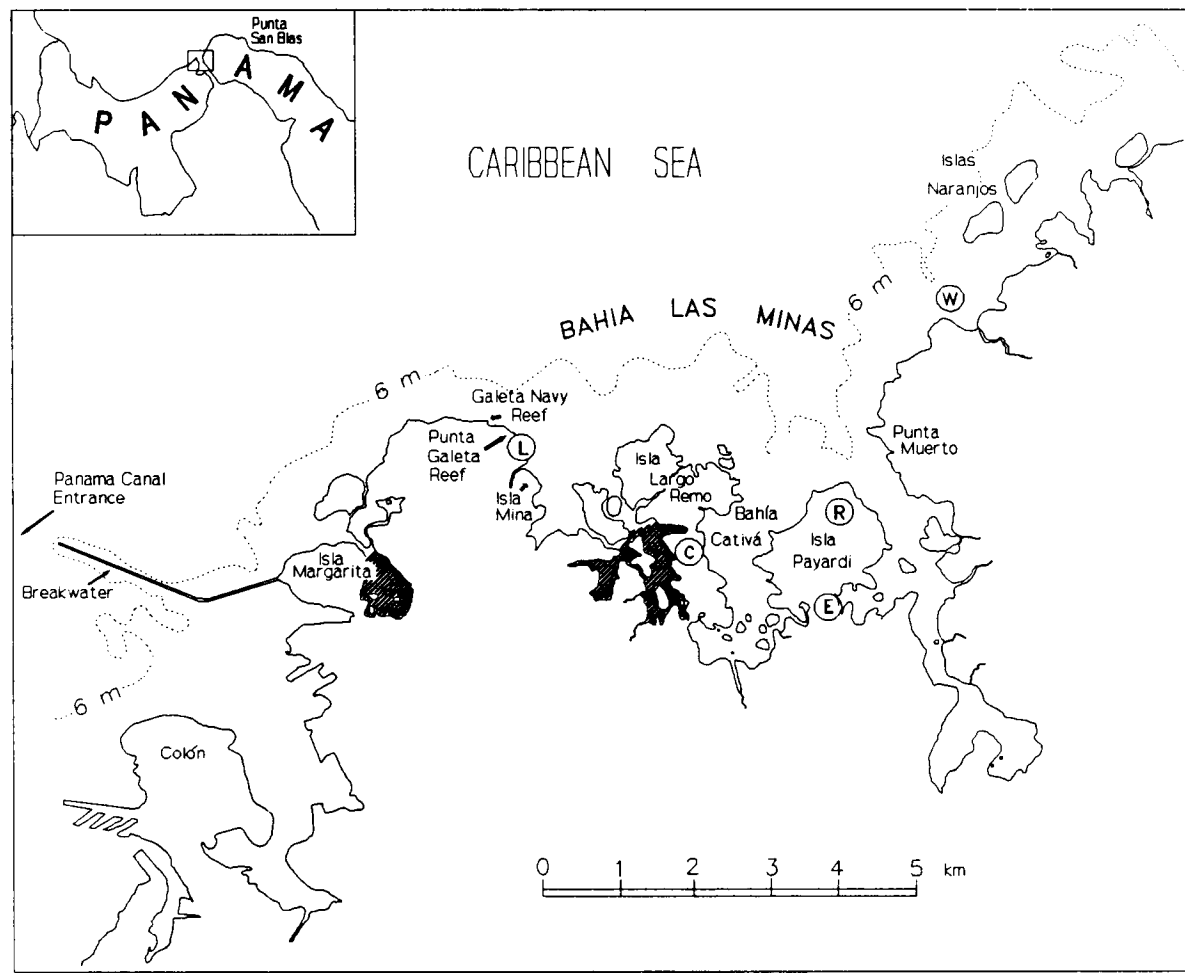


Long-term Assessment of the Oil Spill at Bahía Las Minas, Panama Interim Report

Volume II: Technical Report



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Editors

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List of Abbreviations and Acronyms

ANCOVA	analysis of covariance
ANOVA	analysis of variance
API	American Petroleum Institute
ASCII	American Standard Code for Information Interchange
BBSR	Bermuda Biological Station for Research, Inc.
BLM	Bahía Las Minas
BMDP	BMDP Statistical Software, Inc.
CID	collection identification
CPI	carbon preference index
EOM	extractable organic matter
ESP	Environmental Sciences Program (Smithsonian Institution)
ex/em	excitation/emission
FID	flame ionization detector
GC	gas chromatography
HPLC	high performance liquid chromatography
HWL	high water line
IAEA	International Atomic Energy Agency
IOC	Intergovernmental Oceanographic Commission
IRHE	Instituto de Recursos Hidráulicos y Electrificación
IS	internal standard
LMW	low molecular weight
MANOVA	multivariate analysis of variance
MMS	Minerals Management Service
MS	mass spectrometry
m/z	mass/charge
NRC	National Research Council
NSL	non-saponifiable lipid
PAH	polynuclear aromatic hydrocarbon
PVC	polyvinyl chloride
RF	response factor
RRI	relative retention index
SAS	SAS Institute, Inc.
SCUBA	self-contained underwater breathing apparatus
SI	Smithsonian Institution
SIM	selected ion monitoring
SL	saponifiable lipid
SPM	suspended particulate matter
SPSS	SPSS, Inc.
SQL	structured query language
SRB	Scientific Review Board
STRI	Smithsonian Tropical Research Institute
UNEP	United Nations Environmental Program

xxx

UNESCO	United Nations Educational, Scientific and Cultural Organization
URE	unresolved
UVF	ultraviolet fluorescence
VMIC	Venezuelan/Mexican Isthmian Crude, the type of oil spilled at Bahía Las Minas

List of Acronyms for Study Sites

Chapter 2. The Reef-Flat Sub-Project: Sessile Biota, Infauna, and Sea Urchins on Intertidal Flats

GAL	Galeta
LAR	Largo Remo
MSD	Maria Soto Abajo
MSU	Maria Soto Arriba

Chapter 3. Effects of an Oil Spill on the Gastropods of a Tropical Intertidal Reef Flat

GNRF	Galeta Navy Reef
MSR	Maria Soto Reef
TPR	Toro Point Reef

Chapter 5. Subtidal Reef Corals

DMA	Dos Marias
DONR	Doncella Reef
GALC	Galeta Channel
JUG	Juan Gallego
LRE1	Largo Remo East 1
LRE2	Largo Remo East 2
MAR3	Margarita 3
NARS	Naranjos South
PALW	Palina West
PAYN	Payardi North
PAYW	Payardi West
PM	Punta Muerto

Chapter 6. Mangrove Forests

CCPQ	(Central, Channel) Pequeña
CCSB	(Central, Channel) Samba Bonita
CODR	(Central, Open) Drogue
CRKU	(Central, River) Kuna
ECPY	(Eastern, Channel) Payardi
EOPM	(Eastern, Open) Punta Muerto
ERJA	(Eastern, River) Alejandro
ERPN	(Eastern, River) Puerto Norte
ERPS	(Eastern, River) Puerto Sur

ERUN	(Eastern, River) Unnamed
GOLI	(Isla Grande, Open) Lintón
GOMG	(Isla Grande, Open) Magote Sur
GOMS	(Isla Grande, Open) Maria Soto
GOPA	(Isla Grande, Open) Padre
GOPB	(Isla Grande, Open) Portobelo
MCNO	Margarita Channel Norte
MCSU	Margarita Channel Sur
NRME	Naranjos (River) Las Mercedes
WCLR	(Western, Channel) Largo Remo
WCLS	(Western, Channel) Largo Remo Sur
WCNO	(Western, Channel) Norte
WCSU	(Western, Channel) Sur
WOMI	(Western, Open) Mina
WOPG	(Western, Open) Peña Guapa
WRHD	(Western, River) Hidden
WRRB	(Western, River) Rabinowitz

Chapter 7. Effects of the April 1986 Oil Spill at Isla Payardi on the Epibiota of Mangrove (*Rhizophora mangle* L.) Roots

ALER	Río Alejandro
CO3	Largo Remo Channel Site CO3
CSR	Río Coco Solo
DROM	Isla Droque Mangrove
GALM	Punta Galeta Mangrove
HIDC	Hidden Channel
HIDR	Hidden River
LINM	Isla Lintón Mangrove
LRCS	Largo Remo Channel South
LRCW	Largo Remo Channel West
LRRN	Largo Remo River North
LRRS	Largo Remo River South
MACN	Margarita Channel North
MACS	Margarita Channel South
MERR	Quebrada Las Mercedes
MINM	Isla Mina Mangrove
MSM	Maria Soto Mangrove
PADM	Isla del Padre Mangrove
PAYR	Payardi River
PBM	Portobelo Mangrove
PCE	Payardi Channel East
PCS	Payardi Channel South
PGC	Peña Guapa Channel

PGM	Peña Guapa Mangrove
PMM	Punta Muerto Mangrove
PMRE	Punta Muerto River East
PMRW	Punta Muerto River West
SBCE	Samba Bonita Channel East
SBCS	Samba Bonita Channel South
SBCW	Samba Bonita Channel West
UNR	Unnamed River

Chapter 8. Subtidal Seagrass Communities

BNV	Buenaventura
DONT	Doncella <i>Thalassia</i>
LINE	Lintón East
LREN	Largo Remo Entrance
LRL	Largo Remo Lagoon
LRN1	Largo Remo Norte 1
LRS	Largo Remo Sur
MAR1	Margarita 1
MAR2	Margarita 2
MINN	Mina Norte
MINS	Mina Sur
NARC	Naranjos Channel
NARE	Naranjos East
PALC	Palina Channel
PALN	Palina Norte
PGN	Peña Guapa Norte

Chapter 9. Hydrocarbon Analyses

DONR	Doncella Reef
DONT	Doncella <i>Thalassia</i>
GALC	Galeta Channel
GALS	Galeta Sand
GALT	Galeta <i>Thalassia</i>
HRB	Hidden River Bay
LRE	Largo Remo East
LREN	Largo Remo Entrance
LRLC	Largo Remo Lagoon Center
LRM	Largo Remo Mangrove
LRN2	Largo Remo North 2
LRN3	Largo Remo North 3
LRS	Largo Remo South
MARG	Margarita Grassbed

List of Acronyms for Study Sites (continued)

MARM	Margarita Mangrove
MARS	Margarita South
NARC	Naranjos Channel
NARM	Naranjos Mangrove
NARS	Naranjos South
PALN	Palina North
PALW	Palina West
PAYN	Payardi North
SBS	Samba Bonita South

Chapter 1 Introduction

The National Research Council (NRC 1985) classified tropical regions as "special problem areas" because of the limited knowledge regarding the effects of oil on tropical organisms. Available data indicated that coastal ecosystems in the tropics are at least as sensitive to oil as ecosystems along temperate coasts. Since there was so little information, the NRC report recommended further research, particularly on coral reefs, mangroves, and associated organisms.

In 1986 a major oil spill in the Republic of Panama polluted Caribbean coastal environments, including a biological preserve at a marine laboratory of the Smithsonian Tropical Research Institute (STRI). For the reef flat at this site, baseline biological and environmental data for some parameters extended back over 15 years. There had also been short-term studies on coral reefs, seagrass communities, and mangroves. These pre-spill studies provided a relatively comprehensive background against which to assess biological effects of the spill. Furthermore, observations of effects of the spill began as oil was washing ashore. Such promptness is important since many ecological changes start immediately after such acute pollution (e.g., Sanders *et al.* 1980).

The Minerals Management Service (MMS) of the U.S. Department of the Interior provided funding for the present study because of the unique opportunity for assessing the biological effects of a major oil spill in the Caribbean region. The objectives of the study are to monitor the long-term changes in the distribution and abundance of marine organisms that may result from the spill, and to understand the ecological processes causing any observed changes.

The study area, including oiled and unoled sites, extends along the Caribbean coast of Panama from Toro Point, just west of the entrance to the Panama Canal, to Isla Grande, a straight-line distance of approximately 55 km (Figure 1.1). Heavy oiling from the 1986 spill occurred in Bahía Las Minas.

1.1 The 1986 Oil Spill in Bahía Las Minas, Republic of Panama

The oil spill occurred on 27 April 1986 in Bahía Las Minas, Republic of Panama (Cubit *et al.* 1987; Jackson *et al.* 1989). Approximately 38.3 million liters (240,000 barrels) of medium-weight crude oil drained from a ruptured storage tank at a coastal refinery. The oil was 70% Venezuelan and 30% Mexican Isthmian, with a specific gravity of 27° at 15.6°C (American Petroleum Institute) or about 0.89 g/cc. About 22.3 million liters (140,000 barrels) flooded through the containment dike around the storage tank and overwhelmed separators and a retaining lagoon. In late May 1986, a refinery official reported 9.6 million liters (60,000 barrels) of oil had been recovered from the sea. This volume was somewhat more than the 8.0 million liters (50,000 barrels) reported earlier and published elsewhere (e.g., Jackson *et al.*

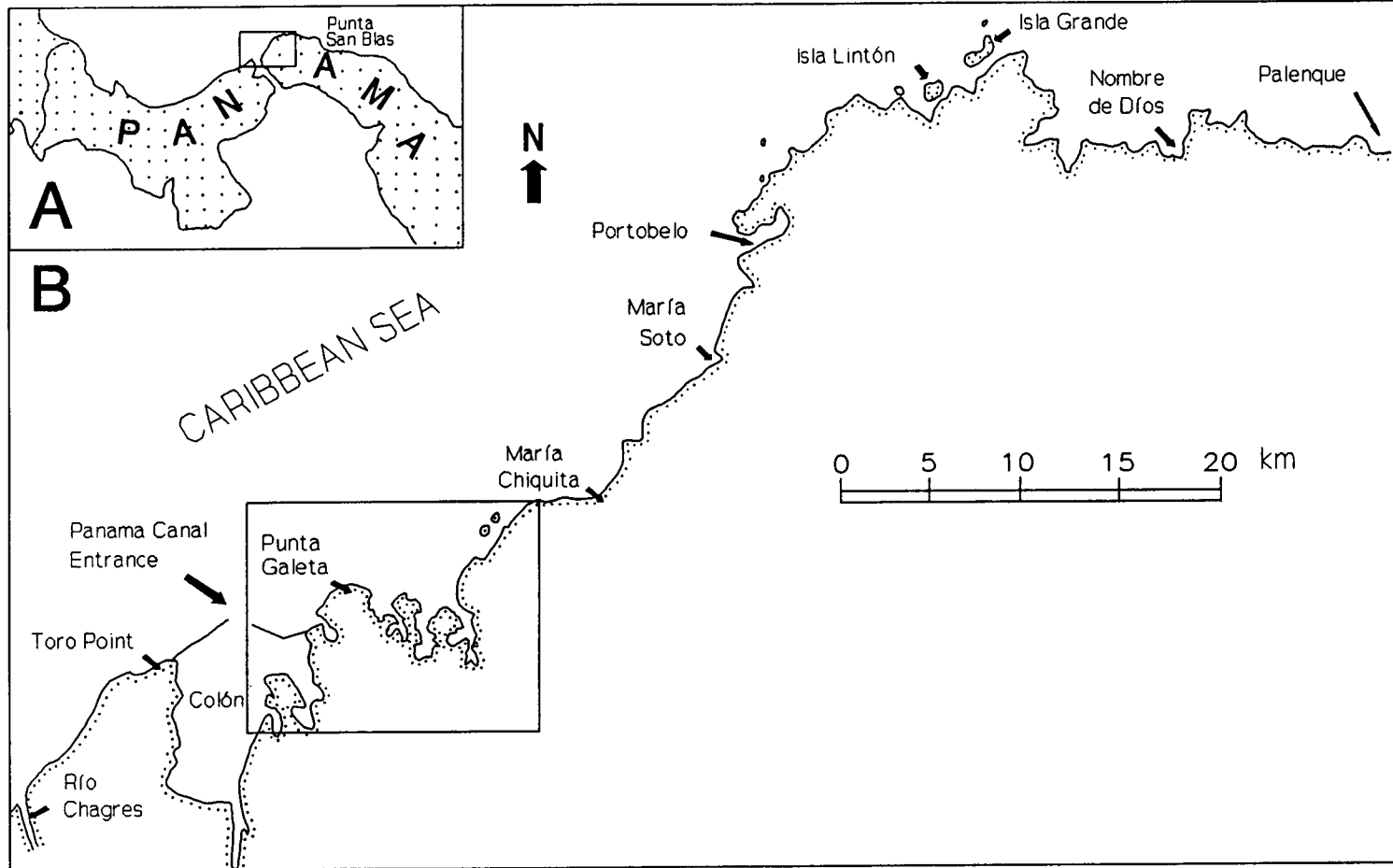


Figure 1.1 Region of the Republic of Panama affected by the 27 April 1986 oil spill, shown as increasing enlargements, (A)-(C). (A) Location within Panama, just east of the Caribbean entrance to the Panama Canal. (B) The boxed area includes the most heavily oiled coastal habitats. Punta Galeta, inside the boxed area, is 9°24'N, 79°52'W. Lightly oiled and unoiled study sites are at Toro Point and northeast of Bahía Las Minas. (C) Detail of Bahía Las Minas, the most heavily oiled area. The encircled "L" marks the marine laboratory of the Smithsonian Tropical Research Institute. Encircled "R" marks the refinery where the oil spill occurred. Horizontal hatching denotes embayments where little oil penetrated. The "C" marks the out-of-use cement plant. The "E" marks the electrical generating station. The "W" marks the location of the stern section of the wreck of the *Witwater*. The dashed line shows the 11-m depth contour.

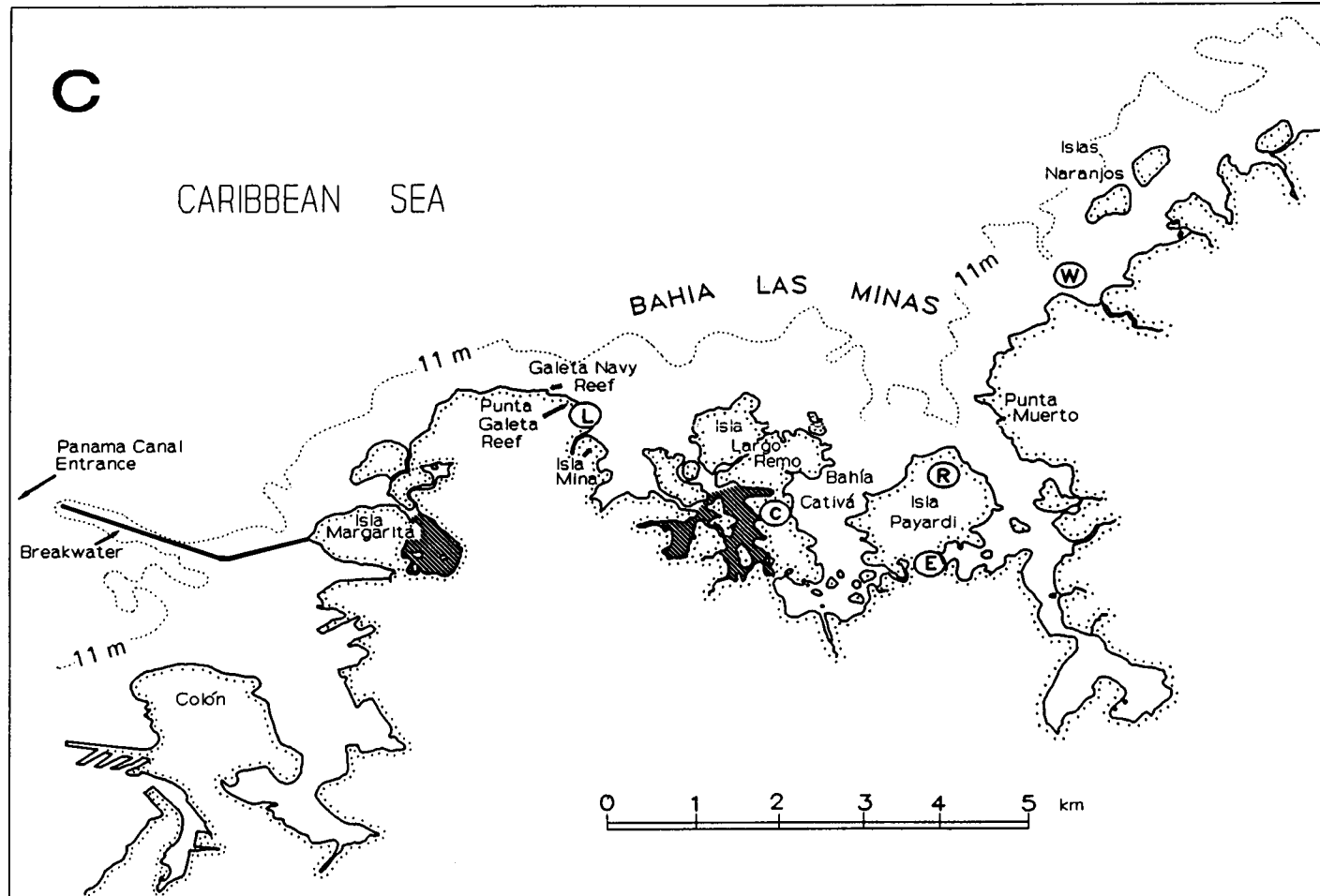


Figure 1.1 Region of the Republic of Panama affected by the 27 April 1986 oil spill, shown as increasing enlargements, (A)-(C) (continued).

1989). We do not know, however, how much oil was not recovered from the sea. It is possible that millions of liters still remain in coastal environments, particularly in mangrove sediments. In any event, even conservative estimates of the volume of this spill are greater than any other reported near coral reefs (Loya and Rinkevich 1980) and mangroves (Getter *et al.* 1981) in the tropical Americas.

For six days, onshore winds held the spilled oil in Bahía Cativá adjacent to the refinery (Figure 1.1). Then shifting winds and run-off from rains (Chapter 10) caused a large quantity of oil to float out to sea past a boom placed across the mouth of this embayment. Starting on 3 May 1986, aircraft sprayed approximately 21,000 liters of the dispersant "Corexit 9527" (Exxon Chemicals) onto oil slicks. A "C130" aircraft was observed spraying dispersant from a very low altitude (at or under tree level) in Bahía Cativá. A small "crop duster" aircraft was observed spraying dispersant on "mousse" near Islas Naranjos (Figure 1.1). Other such applications of dispersant were observed off the breakwater of the Panama Canal, offshore of Bahía Las Minas, and near Portobelo (Figure 1.1). No dispersant was sprayed near Galeta. The application of dispersant so many days after the spill and the calm sea conditions during the spraying appeared to render chemical dispersion ineffective. Although some coastal areas were exposed to the dispersant, particularly Bahía Cativá and near Islas Naranjos, only a relatively small amount was used and many areas, including Punta Galeta, were not directly contaminated by this compound.

By 15 May, oil had spread along the coast and washed across fringing reefs into mangroves, small estuaries, and sand beaches within 10 km of the refinery. During the first two months after the spill, the distribution of oil was surveyed from low-flying aircraft between Río Chagres 27 km west of the refinery and Punta San Blas 98 km to the east (Figure 1.1). Surveys by foot and by boat were conducted from Río Chagres to Nombre de Dios. During these surveys, visual assessments were made of the degree of oiling (heavy, moderate, light, or absent), and of the habitats and types of organisms obviously oiled or affected by oiling. Heavy oiling was observed along much of the coast between Isla Margarita and Islas Naranjos, with the exception of two partially isolated lagoons (Figure 1.1, hatched areas). The length of this heavily oiled coastline is about 82 km (straight-line distance = 11 km), and includes approximately 16 km² of mangroves and 8 km² of intertidal reef flats and subtidal reefs (Cubit *et al.* 1985). Only a few patches of oil were observed east of María Chiquita and west of the entrance to the Panama Canal (Figure 1.1).

In similar habitats within the heavily polluted area, apparent degrees of oiling were highly variable. Probable causes of this heterogeneity include distance from the refinery, directions of movement of the spilled oil, and water depth. The greatest amounts of oil in mangroves, reef flats, and seagrass beds occurred within a few kilometers of the refinery. There was obviously less oil in these habitats at Islas Naranjos and Isla Margarita (Figure 1.1). Large differences in visible oiling also occurred on a much smaller scale of a few hundred meters, depending on the orientation of the coastline. Much of the oil that escaped from Bahía Cativá spread to the west. Accordingly, coasts that faced north to northeast were much more heavily oiled than coasts that faced west or south. Also, seasonal low tides (Cubit

et al. 1986, 1988b, 1989) occurred between 10 and 19 May 1986, causing oil to accumulate along the seaward margins of reef flats. As a result, visible oiling was heaviest in intertidal habitats just above mean low water, such as mangrove roots and associated sediments, reef-flat seagrass beds, coral rock, and beaches. Chemical analyses of petroleum hydrocarbons in surface sediments (Chapter 9) generally verified these visual assessments of variability. They also indicated considerable variability in concentrations of oil among replicates, confirming observations of small-scale patchiness as well.

Several different kinds of efforts were used to clean up the spilled oil. Some oil was removed from the sea using "skimmers" and shore-based pump trucks. As noted above, approximately 9.6 million liters (60,000 barrels) of oil were reported to be recovered this way. Channels were dug through mangroves, apparently to drain oil from these areas. However, these channels appeared instead to increase the movement of oil inshore, and disturbance from workers crushed windrows and may have increased subsequent erosion. In other areas, oiled rocks, rubble, and debris were physically removed and seawater was sprayed to "clean" sandy areas (Chapter 3). Skimming and pumping floating oil appeared by far to be the most effective way to recover oil from this spill. However, shallow water and mangroves impeded many of the kinds of clean-up operations deployed after major oil spills, perhaps for the better, since some of these procedures can be environmentally or biologically destructive.

Oil slicks have been regularly observed in Bahía Las Minas during the four years since the spill. The appearance of these slicks ranges from metallic sheens to brown patches, and slicks appear to come mainly from fringing mangroves, where much of the spilled oil washed ashore. Dead red mangrove trees (*Rhizophora mangle*) have been decaying and, as the wooden physical structure disappears, erosion of the associated oiled sediments has occurred. *Rhizophora* seedlings (survivors, recruits, and planted individuals) have apparently not prevented this erosion. Some slicks also appear to come from the oiled landfill under the refinery.

1.2 Historical Background of the Study Area

Much of the study area has a long history of exposure to various kinds and degrees of human disturbance and pollution. More than a century ago, construction of the Panama Canal and the city of Colón started, followed by decades of excavation, dredging, and landfill. There was extensive drainage of swamps and spraying for control of mosquitoes. On the mainland there has been extensive deforestation, resulting in increased erosion and subsequent deposition of terrigenous sediments in coastal environments.

Construction of the refinery started in 1956 and included excavating over four million cubic meters of coral reef for landfill. The refinery started operating in 1961. Other industrialization in Bahía Las Minas includes an electrical generating station and an out-of-use cement plant, which opened in 1967 and closed in 1975. The small port facility there is still used.

In 1968, the break-up of the tanker *Witwater* caused a major oil spill of approximately 3.2 million liters (20,000 barrels) of diesel oil and Bunker C in Bahía Las Minas (Rützler and Sterrer 1970). Observations on ecological effects of that spill were largely qualitative or conjectural; the Galeta Marine Laboratory was just being established. Since 1968, there have been many ecological and biological investigations in the area, resulting in over 130 publications (see bibliography in Cubit *et al.* 1988b). The Environmental Sciences Program (ESP) of the Smithsonian Institution has supported the collection of detailed time-series data on hydrographic and meteorological conditions (Cubit *et al.* 1988b, 1989) and on the biota of the reef flat at Punta Galeta (see Cubit *et al.* 1986). Such extensive pre-spill information is rarely available in studies of effects of oil spills.

Since the spill, there have been additional small spills in the study area. These include a May 1988 diesel spill near the Toro Point control site for the reef-flat gastropod study (Chapter 3) and fuel oil spills in December 1988 and June 1990 from an electrical generating station at the mainland connection of Isla Payardi (see Figure 1.1). There also are occasional small spills at the port facility of the refinery, and some oil slicks and tar balls wash ashore from offshore shipping. The chemical analyses being conducted should enable most of these sources of oil contamination in samples of biota and sediments to be differentiated.

1.3 Objectives of the Study and Organization of the Report

The study has two main objectives:

- To monitor the long-term changes that may occur in the distribution and abundance of marine organisms as a result of the 1986 oil spill at Bahía Las Minas.
- To understand the ecological processes causing any observed changes.

This report is organized into discrete sections, which include:

- Reef flat sessile biota, infauna, and sea urchins
- Reef flat gastropods
- Reef flat stomatopods
- Subtidal reef corals
- Mangrove forests
- Mangrove roots
- Subtidal seagrass communities
- Hydrocarbon analyses
- Physical environmental monitoring

Each chapter includes methods, results, and discussion. References cited are given at the end of the report. In addition, there are appendix sections detailing data and project management and providing additional data.

Chapter 2
The Reef-Flat Sub-Project:
Sessile Biota, Infauna, and Sea Urchins on Intertidal Flats

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Project Personnel:
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2.1 Introduction

Platforms of fringing reefs form broad shallow habitats along the Caribbean coast of Panama. These structures are covered with highly productive stands of algae and seagrasses (Griffith *et al.* 1987) and are common features of mature reefs throughout the tropics; however, little information exists regarding the effects of oil on these habitats. The *Oil in the Sea* report of the National Research Council (NRC 1985, p. 412) states:

"One area that has been totally neglected is that of spill impact on tropical or warm-water microphytes and macrophytes. Of particular concern in this respect are the giant algal flats that constitute important components of tropical trophic systems and barrier reef systems. These are highly vulnerable to oiling because of their shallowness. Except for a single follow-up study (Lopez, 1978), nothing is known of their recovery potential following oiling."

Data from the long-term environmental monitoring program at Punta Galeta Reef allowed us to compare abundances of algae, seagrasses, and invertebrates on the reef flat before and after the oil spill. Gross variations in the spatial pattern of oiling on the reef flat also allowed us to compare changes in the biota accordingly. After the oil spill, we also began monitoring additional oiled and unoled sites to measure the extent to which the longer-term post-spill events at Punta Galeta Reef represented general phenomena. The organisms quantified in surveys of biotic coverage and echinoid populations on the reef flat represent more than 90% of the living biomass, sessile organisms, primary producers, structuring species, and reef builders in this habitat. These groups, however, do not represent an equivalent percentage of the diversity (i.e., total number of species) on the reef flat. To obtain

a better assessment of diversity, we added the surveys of the species-rich infauna found in turfs of *Laurencia papillosa*, the predominant alga of the reef flat.

2.2 Methods and Rationale for Studies

2.2.1 Selection of Additional Oiled and Unoiled Sites

From previous studies we know that the biota on the Caribbean coast of Panama can change dramatically for reasons other than the effects of oil spills. For example, changes in the abundances of sea urchins and the zonation of algae are associated with year-to-year variations in mean sea levels (Cubit 1985, Cubit *et al.* 1986). In the 1980's alone, we have seen mass mortalities of the sea fan *Gorgonia ventalina* (Guzmán and Cortes 1984; J. Cubit, pers. obs.) and the sea urchin *Diadema antillarum* (Lessios *et al.* 1984a,b). In addition, bleaching and other signs of stress have been observed for corals and other cnidarians (Glynn 1984). Such phenomena occurred over wide areas of the Caribbean Sea. In considering the long-term data from Punta Galeta, it would be easy to attribute such sudden changes occurring now to effects of the 1986 oil spill. Thus, the primary purpose for adding the unoiled sites was to detect pronounced widespread phenomena that would appear in our before-and-after studies at Punta Galeta, but which were not caused by the oil spill at Bahía Las Minas.

Since the precise abundances of the biota at any one time vary naturally from reef flat to reef flat, the added sites serve mainly as comparisons for change of state, rather than absolute state. The principal habitats of interest are those for which we had been making pre- and post-spill comparisons at Punta Galeta. Therefore, in choosing additional sites, our principal criteria were to find sites with habitats comparable to those at Punta Galeta: i.e., sites with a fore-reef slope, emergent reef flat, and inner reef flat habitats with coral rubble and seagrasses. To fit our time and logistical constraints, we selected one additional oiled site and two unoiled sites.

To find such sites, we surveyed the coastline from the Río Chagres to Palenque, a distance of more than 100 km (Figure 2.1). Initial surveys were made by boat, on foot, and from low altitude (<150 m) overflights. During these flights, we also took aerial photos for later reference. Final selection of sites was made on foot. Our selection proceeded as follows:

1. The coastline between the Río Chagres and the city of Colón was eliminated because the reef flats did not comprise the same complement of habitats found at Punta Galeta. Most of these were more elevated than the Punta Galeta reef flat (based on aerial surveys followed up by surveys on foot).
2. The reef flat at Isla Margarita was rejected because dredging projects had changed the reef flat structure (based on surveys on foot).

3. The coastline between Isla Grande and Palenque was eliminated because extensive deforestation had resulted in heavy depositions of terrigenous sediments on the reef flats (based on overflights).
4. The coastline between Portobelo and Isla Grande was eliminated because the reef flats were much more protected from wave action and lacked a forereef slope (determined from overflights).
5. The reef flats between Buenaventura and Portobelo were eliminated because of presence of terrigenous sediments or lack of suitable reef flat structure (surveyed on foot).
6. The unoiled sites were thus narrowed to the coastline between Buenaventura and María Chiquita, and the oiled sites between the Islas Naranjos and Punta Galeta (as shown in Figure 2.1) This coastline was surveyed on foot to make the following selections:
 - a. The unoiled sites: both sites were in the María Soto region, where the reef flats most closely matched those at Punta Galeta. These reef flats are approximately 23 km east of the site of the oil spill.
 - b. The additional oiled site: the seaward (northern) reef flat of Largo Remo Island most closely matched Punta Galeta's. This reef flat borders Bahía Cativá to the west, where the initial oil spill was concentrated. It probably received much more oil than Punta Galeta during the spill. In addition, after the initial spill, it continued to receive more light oiling from the chronic seepage at the refinery and adjacent mangroves. Thus, any effects of oil should be more pronounced at this site than at Punta Galeta.
7. For permanency, all transects were marked with steel stakes driven deep into the reef flat. Additional back-up stakes were set at fixed distances from the primary stakes. The positions of the stakes have been mapped by measuring triangulation distances among stakes and by establishment of lines-of-sight to local landmarks.

2.2.2 Spatial Coverage of Sessile Organisms

As part of a long-term program to monitor changes in zonation at the seaward edge of the reef flat at Punta Galeta, we measured the percent cover of the sessile biota and the thickness of the algal mat in ten permanent transects. The transects were oriented perpendicular to the shoreline and placed randomly within 20 m intervals along the shore. We used a linear point-sample method, recording the species present under exact points (not areas) along the transect line. The thickness of the *Laurencia papillosa* mat was also measured at these points. *L. papillosa* is the

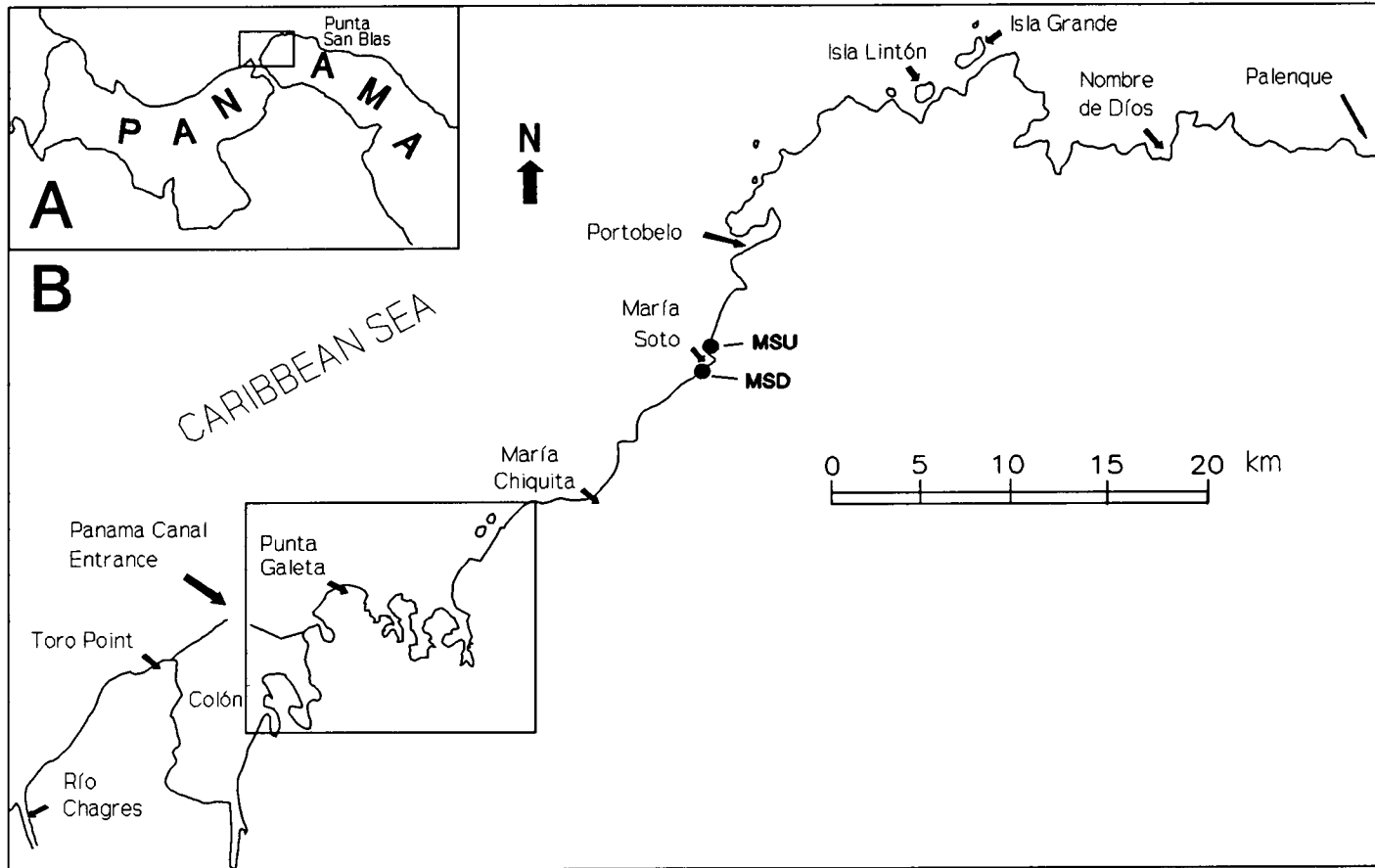


Figure 2.1 Map of areas surveyed for potential study sites, shown as increasing enlargements (A-C). The oiled sites chosen were Punta Galeta (GAL) and Isla Largo Remo (LAR); the unoiled sites were María Soto Arriba (MSU) and María Soto Abajo (MSD). See Figure 1.1 for further details.

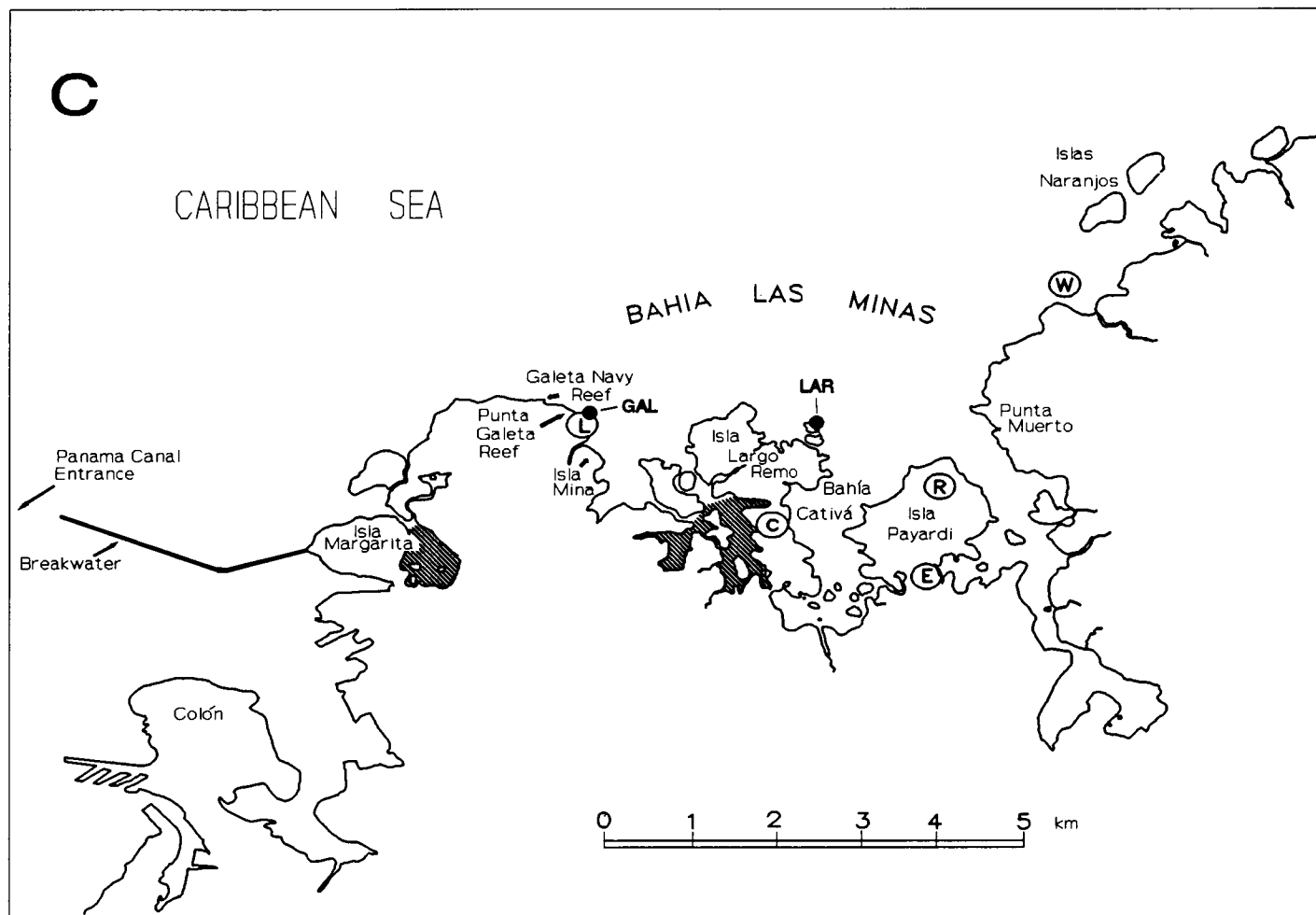


Figure 2.1 Map of areas surveyed for potential study sites, shown as increasing enlargements (A-C) (continued).

principal species at the seaward edge of the reef flat, forming a broad, thick mat paralleling the shoreline. Organisms attached to other organisms were distinguished from those attached directly to the primary substratum (i.e., carbonate rock or sand). Following standard usage, the former are referred to as "secondary cover," the latter as "primary cover." The points were 10 per meter in a stratified-random array: using a random numbers table, five points were randomly spaced within 0.5 m intervals. To span the original area of interest, the *Laurencia papillosa* zone (see Cubit 1985), the transects ranged in length from 9 to 21 m (mean = 15.6 m). The seaward, or zero, end of these transects was located approximately at the lowest low water line on the foreslope of the reef (see Figure 2.2 and figure of reef flat in Cubit and Williams 1983). Sixteen surveys were conducted in the pre-spill period of March 1983 to December 1984. The first post-spill survey was conducted in June 1986 and repeated at approximately three-month intervals thereafter.

At each of the three additional sites, we established five matching transects, which were spaced roughly equidistantly along the portion of the shoreline that most closely matched the seaward edge of the Punta Galeta reef flat. The sections of shoreline surveyed at all four study sites are approximately 100 to 300 m between the first and last transects.

2.2.3 Populations of Sea Urchins

Censuses of sea urchin populations are another part of the long-term monitoring program at Punta Galeta. The urchins are counted in permanent transects and plots. The transects are 1 x 20 m, and the plots, which are approximately square, range in area from 2 to 6 m². Counts are made by species in a square meter quadrat moved over the census area. To maintain continuity with the pre-spill surveys, censuses are made once per month at Punta Galeta, water conditions permitting. The sampling schedule was modified slightly during the oil spill. The first large slicks arrived at Punta Galeta in the afternoon of 9 May. Forewarned, we immediately censused the sea urchin populations. The first post-spill censuses of the transects were made on 31 May and 3 June 1986, 23-26 days after the first oil slicks arrived at Punta Galeta. The censuses were repeated on 19-20 June, 43 days after the first slicks arrived at this site. So, in effect there is an extra census of the transects, but not the plots, between May and June. The censuses made between 31 May and 3 June at Punta Galeta are treated as June data in the analyses.

Matching transects were established at the three additional sites. These are monitored every three months, water conditions permitting; a minimum of three surveys are made per year.

2.2.4 Core Samples of the *Laurencia papillosa* Turf

The core samples are taken in mats of the alga *Laurencia papillosa*, the cover

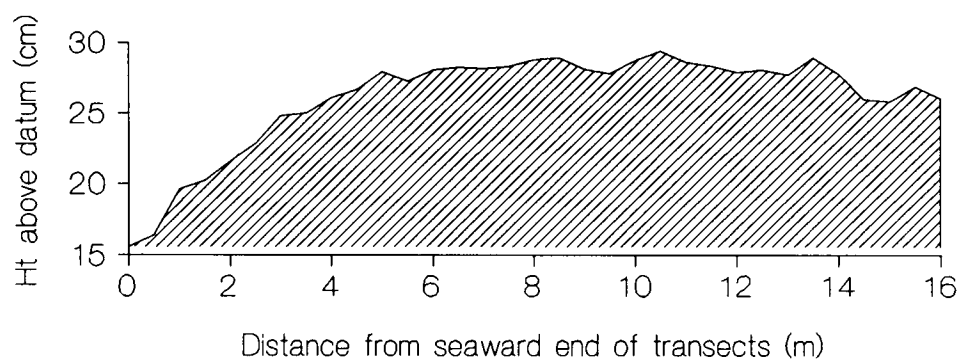


Figure 2.2 Topographic profile of the seaward edge of the reef flat at Punta Galeta. Elevations are above Galeta datum, which is approximately 30 cm below mean low water (Cubit *et al.* 1986, 1988, 1989). The profile is the average of elevations measured along the ten transects used for measuring coverage of sessile biota, and the meter intervals correspond to those given in the transect data.

and thickness of which are also measured in surveys of spatial cover on the reef flat (Chapter 2.2.2). This habitat is near the seaward edge of the reef flat and was strongly affected by the oil spill; most of the algal mat died back to basal tissue. The data from the core samples yielded information regarding recruitment, population dynamics, demography, and abundances for a variety of invertebrates, including those of economic importance. Studies in Florida have shown that the infauna in beds of *Laurencia* spp. are a major source of forage for palinurid lobsters, as well as a refuge and the preferred settlement habitat for juvenile lobsters (Marx and Herrnkind 1985a,b; Herrnkind and Butler 1986). Studies at Punta Galeta (Richards, unpubl.) have also shown that palinurid lobsters forage extensively in *Laurencia papillosa* beds. During more than 200 hours of night-time observations and more than 1000 hours of daytime observations over a ten-year period, other economically and ecologically important species of crabs, shrimp, octopus, and fish have also been observed to forage on the infauna or take refuge in this habitat (J. Cubit, pers. obs.).

To compare the infauna in the *Laurencia* beds at oiled and unoiled sites, 60 core samples per site are taken from *Laurencia* beds every six months. That is, for the two oiled and two unoiled sites, a total 240 samples are taken each sampling period. The core samples are taken using a hand sledge to drive a stainless steel pipe 5.2 cm in diameter (21.2-cm² area) through the algal mat into the hard substratum of the reef flat. To assure a complete core of the algal mat, cores are rejected if a plug of hard substratum is not removed at the base of the core. The cores are taken along transect lines perpendicular to the shoreline. The transects are randomly placed for each sampling. The positions of all samples are recorded to avoid repeated sampling at the same location later. Samples are taken in clusters of three at four positions along the transect lines. The spacing is 2 m between clusters. The first cluster on each transect is taken at the seaward edge of the *L. papillosa* zone. The remainder are taken 2, 4, and 6 m from the seaward edge of the *L. papillosa* zone. Each sample of a cluster is taken in the densest *L. papillosa* mat within the same 50 x 50 cm quadrat, maximizing the distance among samples. If the algal mat does not exist within the designated location, the position is moved laterally (parallel to the shoreline) to the nearest area of algal mat. Where the algal zone is less than 6 m wide, a landward set of samples is not taken. To avoid interference, no samples are taken in the survey areas of the permanent transects used to measure spatial coverage and urchin abundances. The samples are labeled showing the position, site, and date where collected and are preserved on the day of collection.

The samples are stained with rose bengal to assist in distinguishing animals from the debris. The algae, substratum, and infauna are separated. The type of substratum and species of predominant macroalgae in the mat are recorded and the materials are separated, dried, and weighed. At this first level of processing, the infauna are separated into major taxonomic groups (usually order). Except for polychaetes, all invertebrates are counted and their length measured. Because the polychaetes tend to be in fragments and are elastic, no linear measurement has proved satisfactory to quantify their abundance. Methods to quantify polychaete abundance are now under study.

2.2.5 Qualitative Sampling Immediately after the Oil Spill at Punta Galeta

7 June 1986: To examine the developing flora and fauna at Punta Galeta after the oil spill, samples of the hard carbonate substratum were collected from the seawardmost portion of the reef flat surveyed in the transects for measuring spatial coverage. Six pieces of substratum approximately 2 to 3 cm² in surface area were wrapped in porous waxed paper and placed in paper envelopes to permit gas exchange while retaining moisture. Samples were microscopically examined while fresh and then preserved by desiccation at room temperature.

29 June 1986: 18 samples of algae were taken adjacent to nine of the 10 transect lines. The samples were taken 10 cm away from the line. Nine samples were taken 10 cm landward of the zero marker for the transects. The other nine samples were taken 2 m landward of this marker. The samples were processed as on 7 June 1986.

2.2.6 Growth of Sea Urchins

To complement the on-going censuses of sea urchin populations, pilot studies for examining the growth of sea urchins were conducted. Two methods were investigated for measuring growth rates of the principal sea urchin of the reef flat, *Echinometra lucunter*. In both methods, a range of sizes of urchins found at the study sites (approximately 1 to 4 cm test diameter) were used. The first method examined the plates of the test for growth rings. The animals were cleaned in sodium hypochlorite ("Clorox") and plates from various parts of the test examined microscopically. The plates were ground with fine abrasives and were also treated with combinations of heat, xylene, and immersion oil to enhance the contrast of the growth rings. The second method used tetracycline markers following procedures reported in the literature (reviewed in Ebert 1982, H. Lessios, pers. comm.). The tetracycline was administered in two ways: by the reported procedure of injection (Ebert 1982) and by immersion in a solution of tetracycline in seawater. To examine the efficacy of this staining method, the urchins were frozen 1-2 days after treatment. After cleaning, the jaws of the urchins were examined under a microscope with epifluorescent illumination.

2.2.7 Database Development

The following set of databases from monitoring studies at Punta Galeta is being organized to determine the effects of the 1986 oil spill:

1. ZONE (1971-1977): Percent cover of sessile organisms in various zones on the reef flat.
2. BIOMASS (1979-1980): Dry weights per unit area of sessile organisms on the reef flat.

3. CONSURV (1981-1982, plus two post-spill surveys in 1986): Percent cover of sessile organisms on the reef flat.
4. REDGE (1983-1984, 1986-present): Percent cover of sessile organisms and volume of *Laurencia papillosa* bed at the seaward section of the reef flat.
5. CORES (1987-present): Infauna of core samples from the *Laurencia papillosa* bed.
6. URCHINS (1971-present, 1978-present; two sets of plots): sea urchin populations in permanent plots on the reef flat.
7. HYDMET (1974-present): solar radiation, rainfall, wind, air temperature, water temperature, salinity, water level at Punta Galeta. (Managed By Karl Kaufmann, Ricardo Thompson and Carlos Guevara.)

The cover, biomass, and urchin measurements include more than 90% of the total biomass, biotic coverage, primary producers, structural components, and builders of the reef flat. The CORES data include much of the species diversity on the reef flat. All census information includes map coordinates, which allows data to be related to patterns of oiling and among studies.

2.3 Results

2.3.1 Field Observations During the First Month of Oiling at Punta Galeta

The first large masses of oil arrived at Punta Galeta on 9 May 1986, 12 days after the spill at the refinery. Although the oil had weathered during this time, it was still liquid (not tarry) and had the appearance of thick, used crankcase oil. During the low tides between 10 and 19 May 1986, the oil accumulated along the seaward side of the reef flat (Figure 2.3). Organisms in this zone (approximately meters 0-6 in Figure 2.2) were directly immersed in oil, and the surface of the substratum was obscured by oil. By 17 May 1986, the smell of rotting invertebrates and the appearance of numerous sea urchin tests indicated massive mortality beneath this layer of oil. By 24 May 1986 enough oil had worn away to reveal a 1-3 m wide band of white carbonate substratum, which included bleached corals and coralline algae. The band paralleled the shore at about the lowest low water line, marking a zone previously occupied by a mixture of zoanths, corals, and a mixture of calcareous and fleshy algae. By systematic visual estimation, less than 10% of the original sessile community still remained. A translucent fuzz of fine filamentous algae had started growing over the white carbonate substrate.

By search, we could find none of the crabs (e.g., species of *Microphrys*, *Pachygrapsus*, and *Grapsus*) normally present in this zone. Live *Grapsus grapsus*, however, were present on nearby emergent habitats (logs and coral rubble). These usually fast-running crabs were easily caught by hand, and appeared to be blind.



Figure 2.3 Aerial photograph of Punta Galeta during the oil spill, showing the accumulation of oil along the seaward edge of the reef flat.

On 1 June 1986, patches of oil still adhered to the reef flat. The fleshy, turf-forming, algae *Laurencia papillosa* and *Gelidiella acerosa* showed no visible damage in deeper tide pools and shallow subtidal areas, but were much less abundant than usual on the emergent substrata of the reef flat. The thalli of these algae dried at low tide, allowing the oil to adhere. Afterwards the fronds gradually died back to the basal tissue.

By 7 June 1986 the translucent, golden-green fuzz of microalgae was approximately 0.1 - 0.5 cm thick and covered a visually estimated 75% of the seaward edge of the reef flat. This is the zone where the spatial coverage of sessile biota and the "reef-edge" populations of sea urchins are censused; see Figure 2.2. The mat of microalgae was absent in the following areas: (1) in pits where sand had accumulated; (2) in pits where coralline algae were still present; (3) in patches ~2-10 cm diameter around holes (~1 cm diameter) occupied by small crabs (*Pachygrapsus* sp., ~0.5 cm carapace width), which had recently colonized these holes. Similar holes without crabs were not surrounded by cleared patches. In microscopic examination of the samples taken on this date, the algal fuzz consisted of the following (in order of abundance): the filamentous green alga *Cladophora* sp., the cylindrical green alga *Enteromorpha* sp., and the filamentous red alga *Centroceras* sp. At least seven species of pennate and centric diatoms overgrew the substratum and other algae. In addition, ciliated protozoans, nematodes, and harpacticoid copepods were abundant among the algal filaments. Small globules of oil were also found in the algal samples. Although blue-green algae were abundant in nearby areas, few blue-green algae were found in these samples.

2.3.2 Spatial Coverage of Sessile Organisms

Since the oil spill occurred only a few years ago, the long-term, pre-spill data from Punta Galeta, rather than the short-term, post-spill data from all sites, will be emphasized to examine the effects of oiling. As defined earlier, the purpose of the data from the other sites is to compare changes of state, rather than any differences in absolute state. The temporal comparisons among sites are of limited value when based on shorter-term data. There are two aspects of the surveys from Punta Galeta that can be used to infer the effects of the oil spill from within-site comparisons: temporal changes (pre-spill vs. post-spill) and spatial changes (comparing the heavily oiled seaward edge of the reef flat to the rest of the surveyed area). In the following, three levels of data organization are used to make these comparisons:

1. Temporal variation of the principal categories of spatial data: (a) macrospecies (macroalgae and invertebrates); (b) microalgae; and (c) bare substratum (sand, rock, and coral rubble).
2. Temporal variation of finer divisions of the first set of categories.
3. Temporal and spatial variation of the finer divisions of spatial categories.

The first post-spill measurements of spatial coverage at Punta Galeta were made on 25-26 June 1986. In this survey the overall coverage of macroalgae and invertebrates was lower than in any pre-spill survey (Figure 2.4). The proportional reduction was greater for invertebrates, as a group, than for macroalgae (Figure 2.5). The groups of invertebrates showing the greatest percent reduction were the stony corals (mostly *Millepora* spp. and *Porites* spp.) ($p \leq .01$, paired t-test) and zoanthids in the genus *Palythoa* ($p \leq .01$, paired t-test) (Figure 2.5). The overall abundance of zoanthids in the genus *Zoanthus* was less affected ($p > .05$, paired t-test) (Figure 2.5).

At the time of the first post-spill census, the reductions in abundance of all macrospecies were greatest at the seaward ends of the transects, where the oil had accumulated during the low tides (Figure 2.6). Differences in zonation patterns account for many of the differences in proportional mortality among the various groups of organisms. For example, in the pre-spill surveys, the highest densities of cnidarians were at the seaward edge of the reef flat, where the coverage of all macrospecies had been reduced to near zero in the first post-spill survey. Populations of the cnidarians *Palythoa* spp. and the stony corals, which had been almost entirely restricted to this zone, were therefore almost eliminated by the oil spill. Although the cnidarian *Zoanthus* also suffered heavy mortality in this seaward zone, survival of the *Zoanthus* population in the landward portions of the transects maintained a substantial population through the oil spill.

As noted in the field observations (Chapter 2.3.1), microalgae immediately colonized much of the substratum vacated by mortality of macroalgae and invertebrates, so little open substratum existed after the oil spill (Figures 2.4, 2.5, 2.6). By the time of the first post-spill census, the proliferation of microalgae at the edge of the reef flat had developed into an opaque, dark, golden-brown felt that covered nearly all hard substrata (Figures 2.4 and 2.5; Appendices C.2 and C.3). This mat was composed of the same species listed above, and the diatoms were a principal component, overgrowing both the substratum and other types of algae. In more sheltered areas, blue-green algae, such as *Calothrix*, *Lyngbya*, and *Oscillatoria*, also formed thick mats, but were absent from the edge of the reef flat. The mean percent cover of microalgae in the first post-spill survey was nearly double the highest coverage measured in any of the 16 pre-spill surveys. The secondary cover of microalgae (i.e., epiphytic cover) was also highest on this date (Appendix C.4). As shown in Figure 2.6, the microalgae were more abundant nearer the seaward edge of the reef flat, which was the reverse of the pattern of zonation for microalgae before the oil spill.

Recovery patterns after the first post-spill survey were dependent on recruitment events, growth, and the most severe exposures of the reef flat above water level in 15 years of records (Figure 2.7). The extreme intertidal exposures of

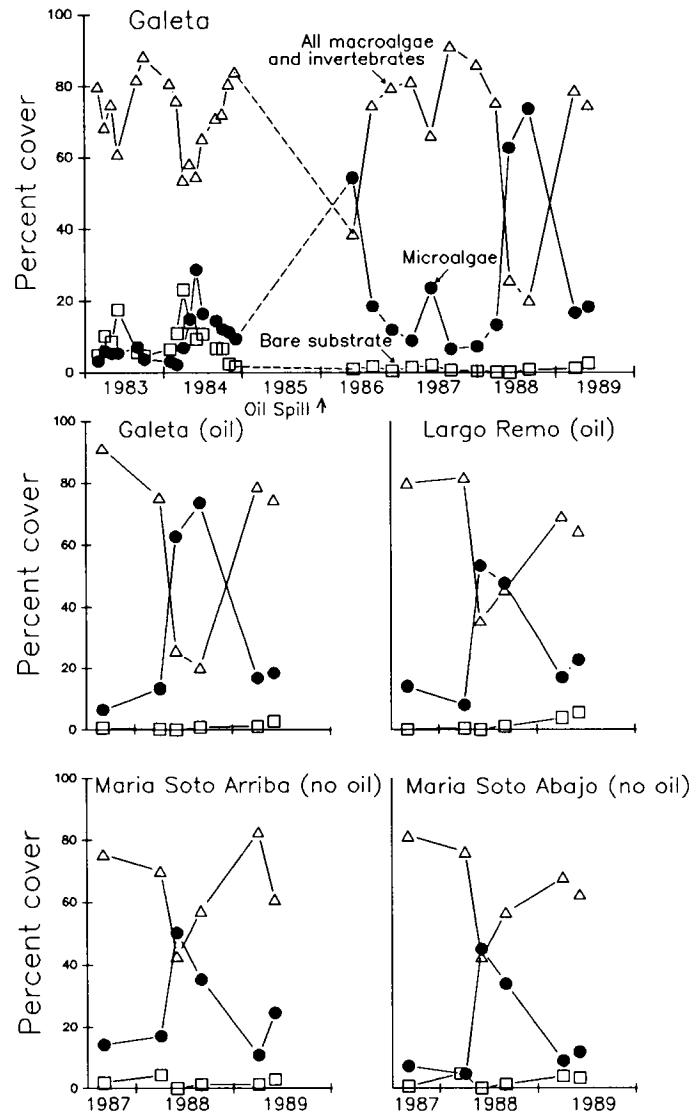


Figure 2.4 Changes in percent cover of macrospecies, microalgae, and bare substratum in the reef-edge transects at all study sites. (Open triangles = macrospecies [i.e., sessile invertebrates and macroalgae]; closed circles = microalgae; and open boxes = bare substratum [i.e., bare rock, loose rubble, and sand].) Together these groups account for all biotic cover or unoccupied substratum at the seaward edge of the reef flat. The monitoring began in 1983 at Punta Galeta and was extended to the other sites in 1987. The dashed line in the top graph spans 1985, when no surveys were made. The arrow denotes the arrival of the first major oil slicks at Punta Galeta in May 1986.

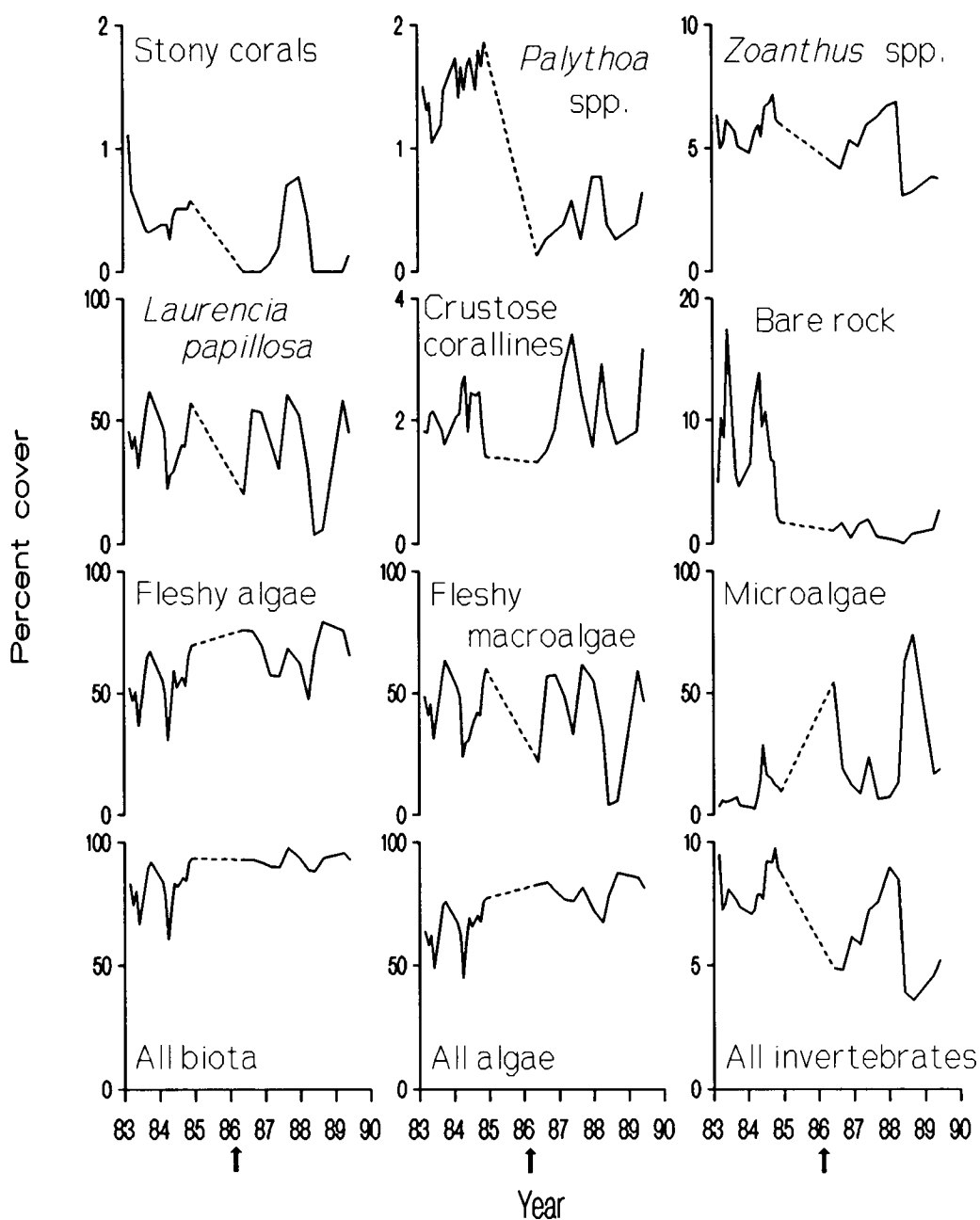


Figure 2.5 Changes in percent cover of major groups of sessile organisms and bare rock in the reef-edge transects at Punta Galeta. The three groups in the top row are invertebrates. Note that the vertical scales differ considerably among graphs to show better the variations in cover for each group. As shown by the dashed line, no surveys were made in 1985. The arrow marks the time when the first oil slicks arrived at Punta Galeta.

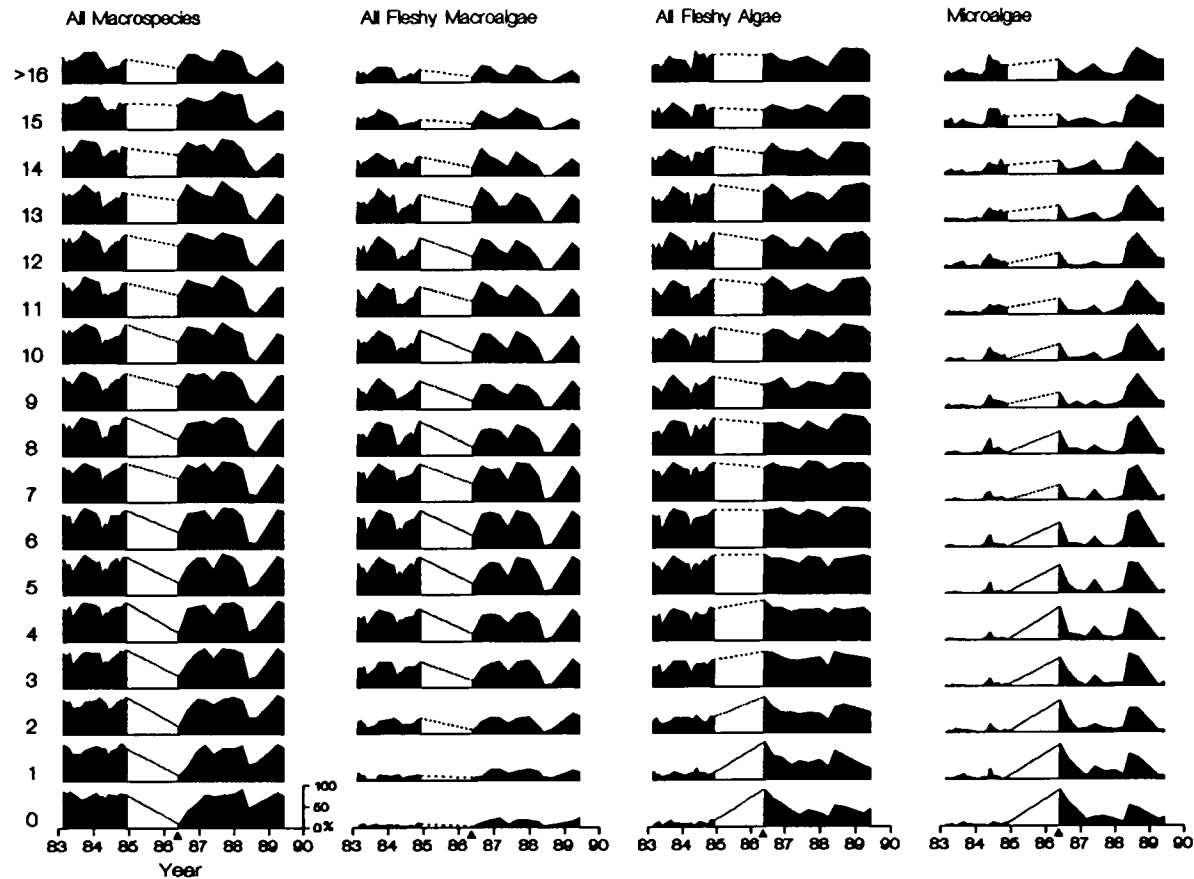


Figure 2.6 Spatial variation in changes of percent cover of major groups of organisms and bare substrata in the reef-edge transects at Punta Galleta. Note that the vertical scales differ among graphs to better show temporal variations in cover for each group. As in the previous graphs, the dashed lines span 1985, when no surveys were made. The numbers 0-16 at the left are meter intervals along the transects; 0 is closest to the sea. The triangle marks the time when the first oil slicks arrived at Punta Galleta.

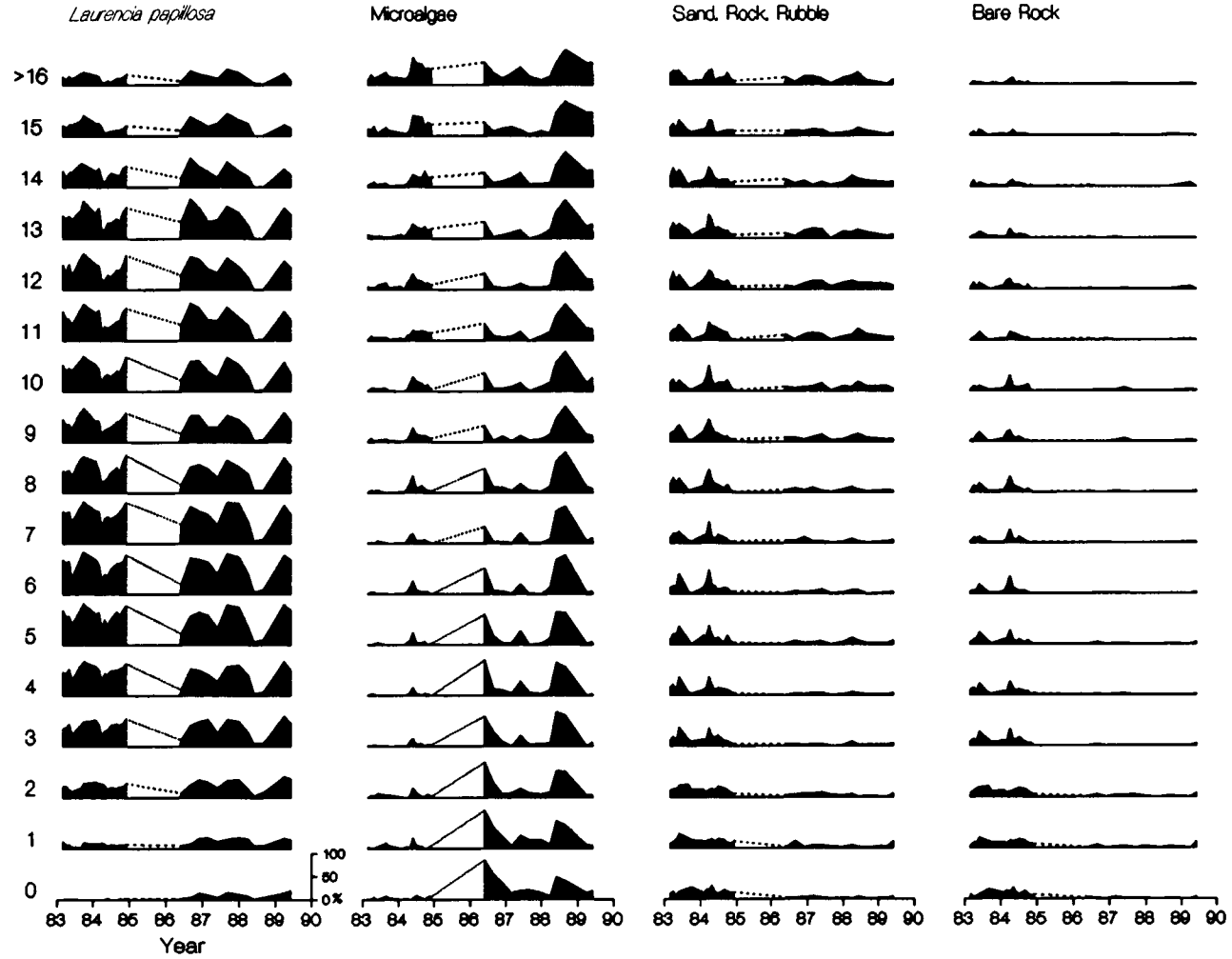


Figure 2.6 Spatial variation in changes of percent cover of major groups of organisms and bare substrata in the reef-edge transects at Punta Galeta (continued).

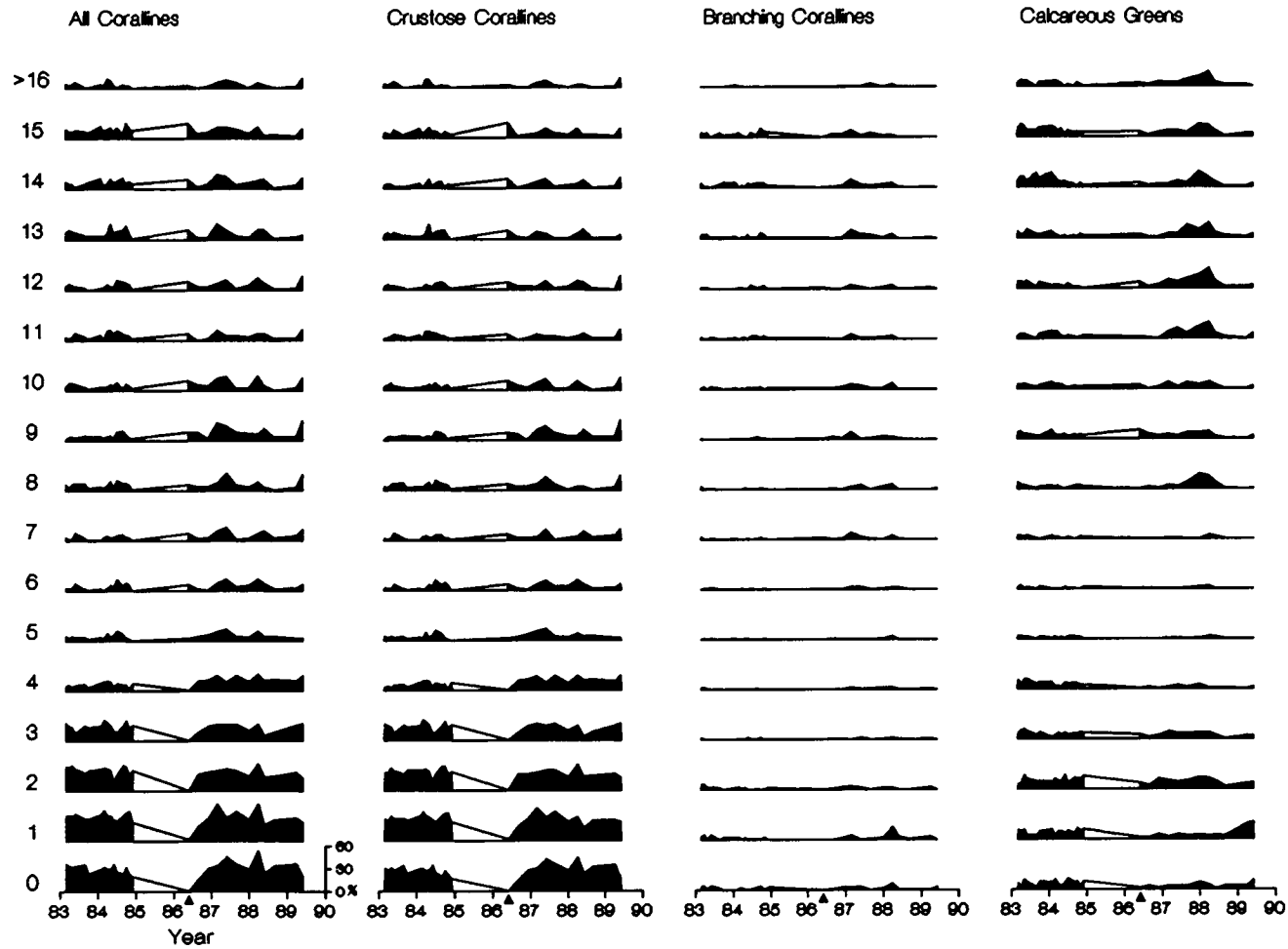


Figure 2.6 Spatial variation in changes of percent cover of major groups of organisms and bare substrata in the reef-edge transects at Punta Galeta (continued).

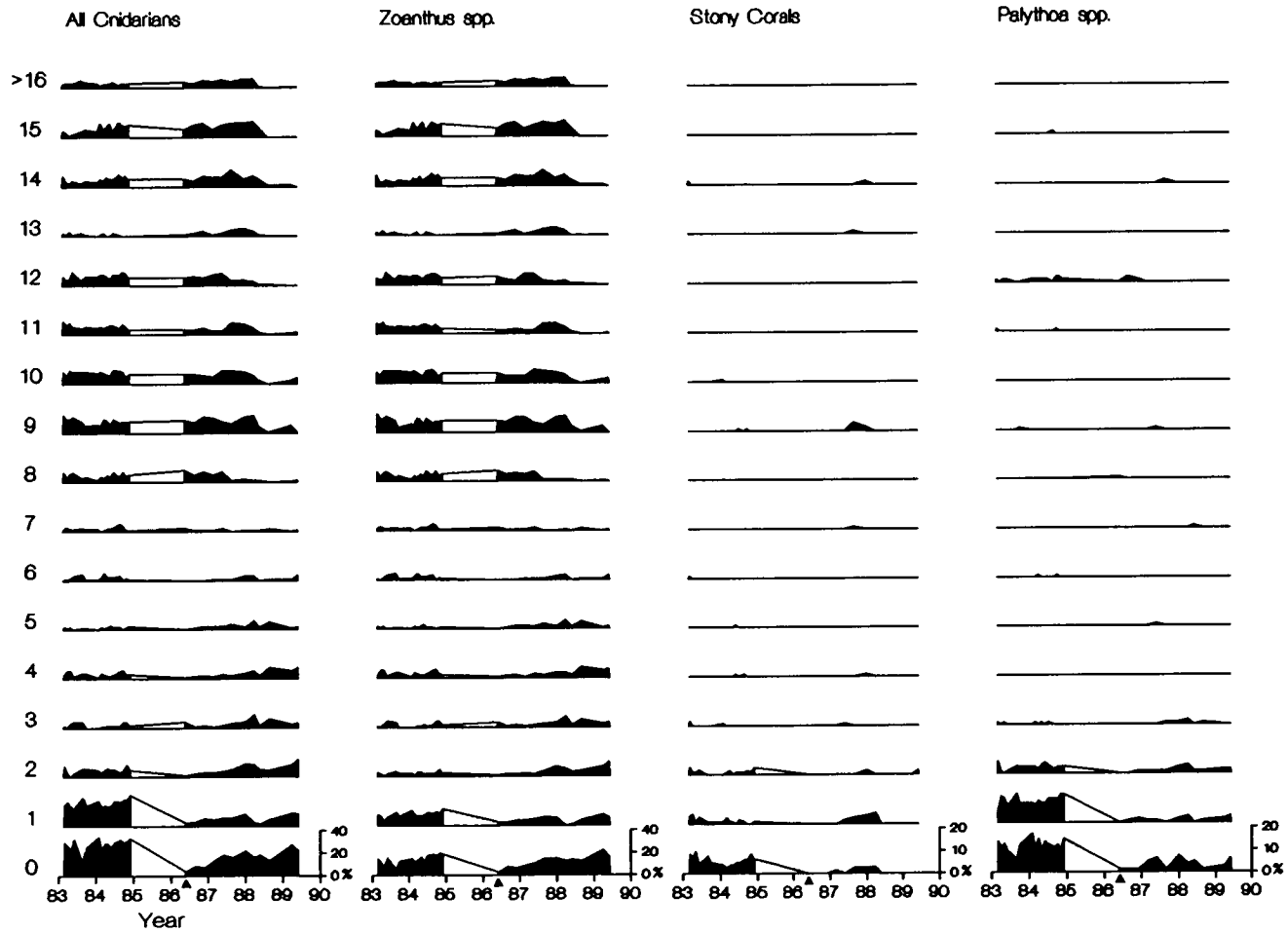


Figure 2.6 Spatial variation in changes of percent cover of major groups of organisms and bare substrata in the reef-edge transects at Punta Galeta (continued).

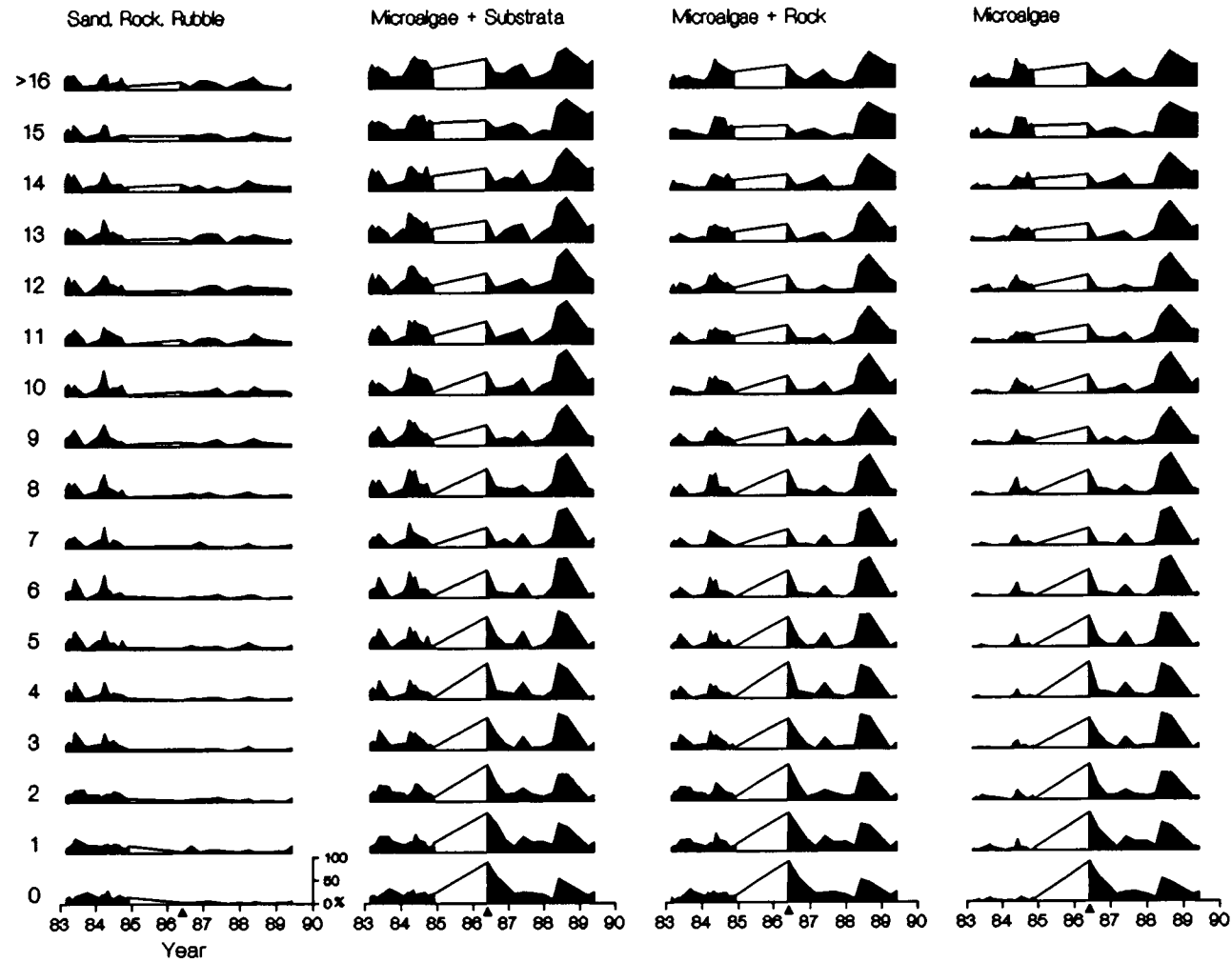


Figure 2.6 Spatial variation in changes of percent cover of major groups of organisms and bare substrata in the reef-edge transects at Punta Galeta (continued).

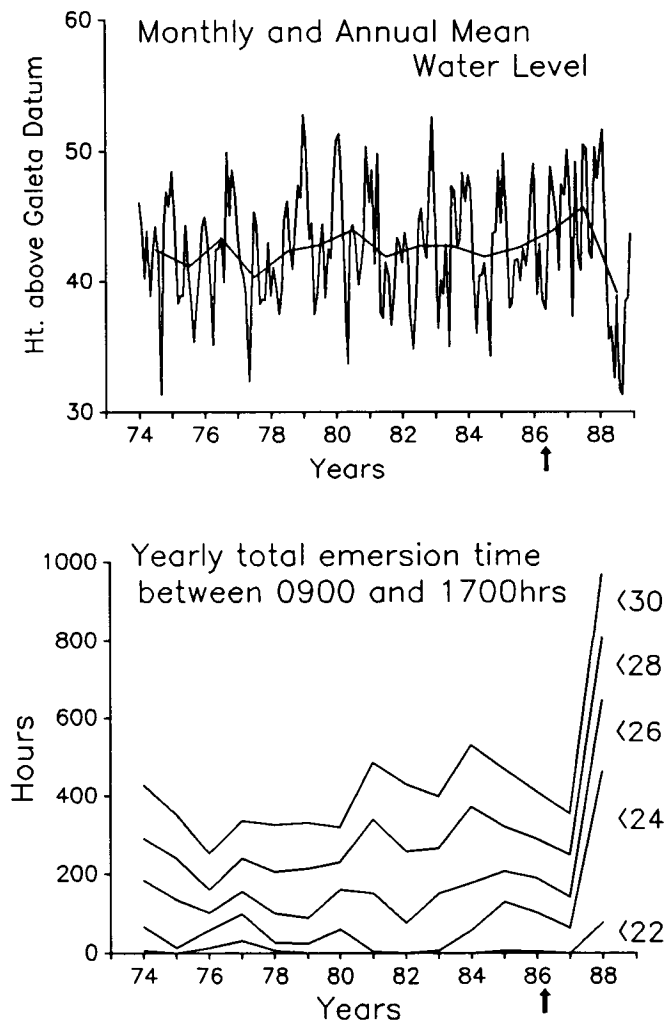


Figure 2.7 Water levels and total hours per year of daytime emersions of the reef flat at Punta Galeta, 1974-1988. All data are derived from hourly records of water level; heights are centimeters above Galeta datum. (The datum level is approximately 30 cm below mean low water; Cubit *et al.* 1988, 1989). In the upper graph, the central line is the annual mean water level, and the more jagged line connects monthly mean water levels. The lower graph shows the total hours per year that water level records were below the cm levels shown at the right during 0900-1700 hours, which is the most stressful portion of the day. For reference, only the highest parts of the reef crest are exposed at 30 cm water levels, whereas nearly all of the reef flat is exposed when water levels are <24 cm. The arrow marks the time when the first oil slicks arrived at Punta Galeta.

1988 reduced the coverage of many of the macrospecies recovering from the oil spill. In contrast to damage from the oil spill, however, damage from the extreme low water levels was concentrated on the higher substrata farther from the seaward edge of the reef flat, as summarized in the plot of "All Macrospecies" in Figure 2.6.

In 1987, waves transported coral fragments (mostly *Millepora* spp.) onto the reef flat from deeper water, producing the pulse of new coral cover shown in Figures 2.5 and 2.6. All post-spill recruitment of corals occurred in this manner. No evidence was seen of any coral developing from any fragment smaller than approximately 2 cm², thus ruling out recruitment from larval stages or individual free polyps. However, unlike the pre-spill coral cover, which was confined primarily to the seaward edge of the reef flat, post-spill recruitment was spread out along the transects. Most of these fragments did not survive through 1988, the year of severe exposures of the reef flat above water level. As of June 1989, the corals and the zoanthid *Palythoa* spp. had not regained their pre-spill abundances (Figure 2.5). In contrast, the algae (as a group) have regenerated quickly and their overall abundance has not been depressed by the 1988 exposures. At the seaward ends of the transects the fleshy algae have been more abundant after the spill than before. These algae (the microalgae and fleshy macroalgae) invaded the seaward zone previously occupied by corals and *Palythoa*. The proportion of microalgae and fleshy macroalgae has varied, but the increased abundance of fleshy algae has continued through all post-spill surveys (as of June 1989).

By September 1986, the coverage of microalgae had decreased to within the range of coverages measured before the oil spill. During 1988 the extensive exposures of the reef flat resulted in another bloom of microalgae having greater total coverage than the post-spill bloom (Figures 2.4 and 2.5); however, the two blooms had almost the reverse patterns of zonation. In the 1988 bloom the microalgae were least abundant at the seaward edge of the reef flat and increased going landward (Figure 2.6).

2.3.3 Populations of Echinoids

Figure 2.8 shows the population variations of sea urchins at Punta Galeta since late 1970. When the oil slicks arrived at Punta Galeta, *Echinometra lucunter* was the most abundant sea urchin on the reef flat; *Echinometra viridis* was the second most abundant. Population variations on the reef flat are highly seasonal, and in the May-June period when the oil came ashore, other genera of sea urchins were normally rare (Cubit *et al.* 1986). In addition, the May-June period is usually a time of recruitment for the *Echinometra* spp., as illustrated for *Echinometra lucunter* in Figure 2.9. In 1986, *Echinometra* populations had been increasing for several months before the oil slicks arrived at Punta Galeta, particularly in the reef-edge transect, where the largest post-spill declines were recorded (Figures 2.8 and 2.9). Lesser decreases were observed in the *Thalassia* transect, 26 m directly landward of

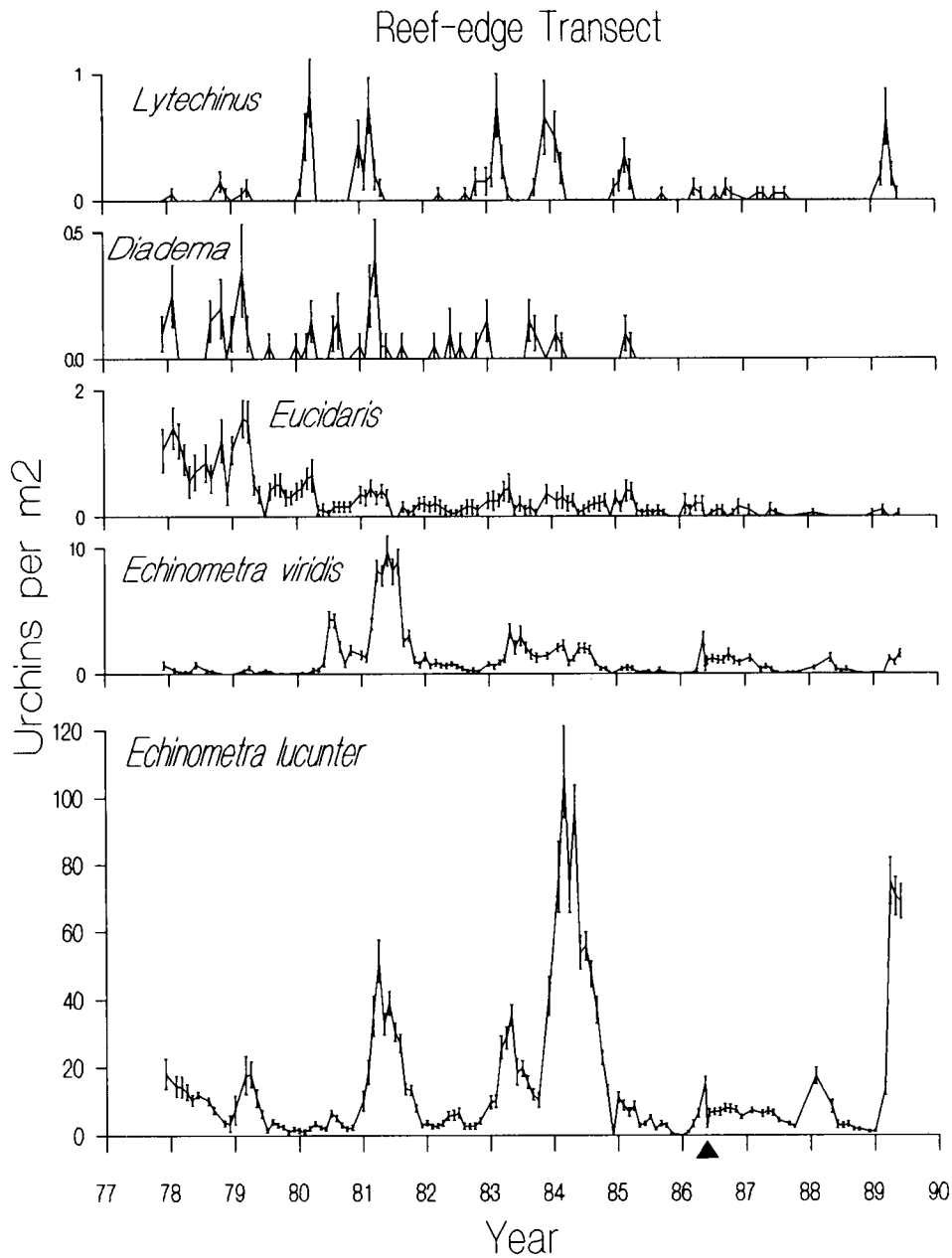


Figure 2.8 Populations of echinoid species on the reef flat at Punta Galeta. These graphs show the abundances of the more abundant species of echinoids in the permanent transects and plots. Note that the vertical scales vary among graphs to show temporal variation more clearly. The triangle marks the time when the first oil slicks arrived at Punta Galeta.

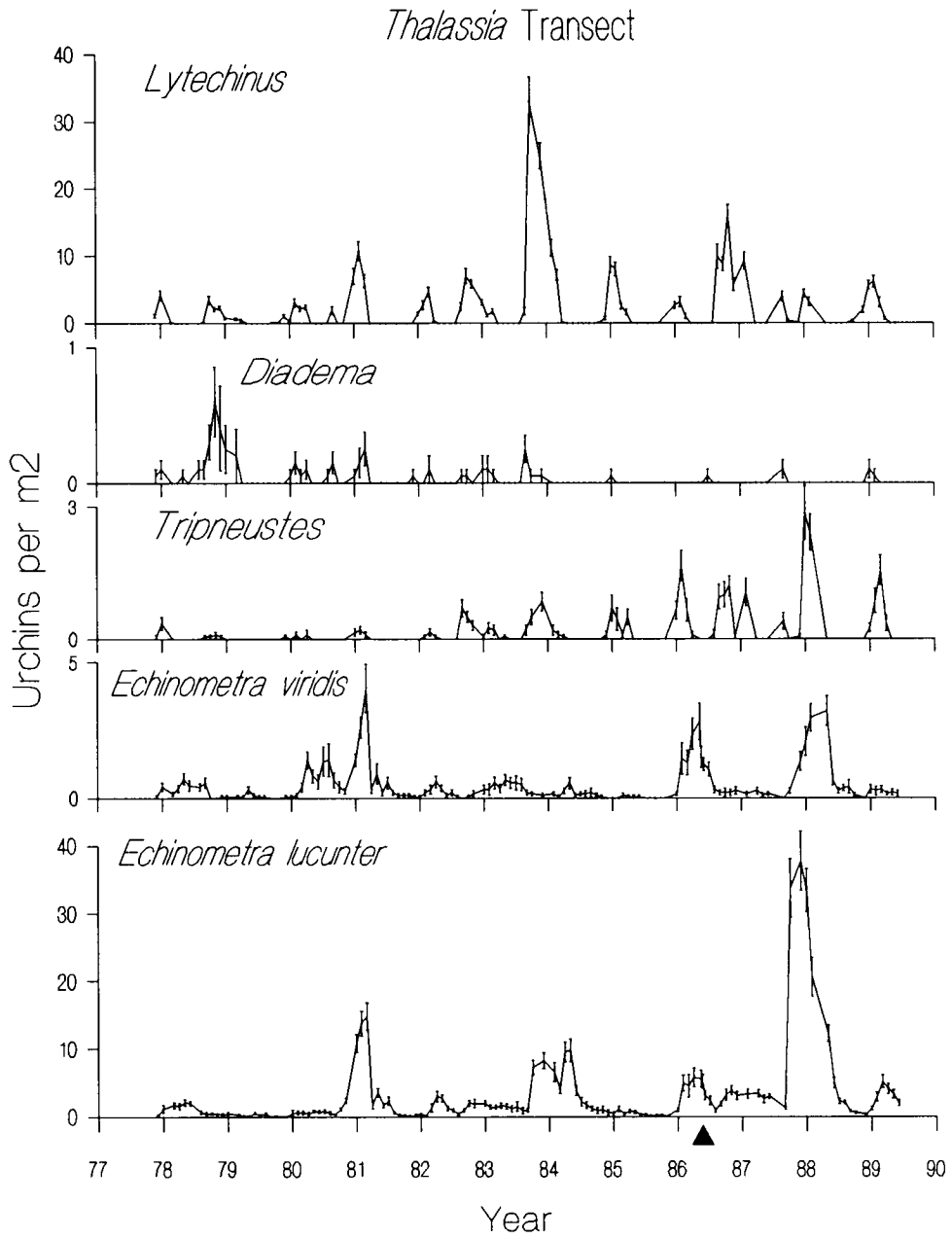


Figure 2.8 Populations of echinoid species on the reef flat at Punta Galeta (continued).

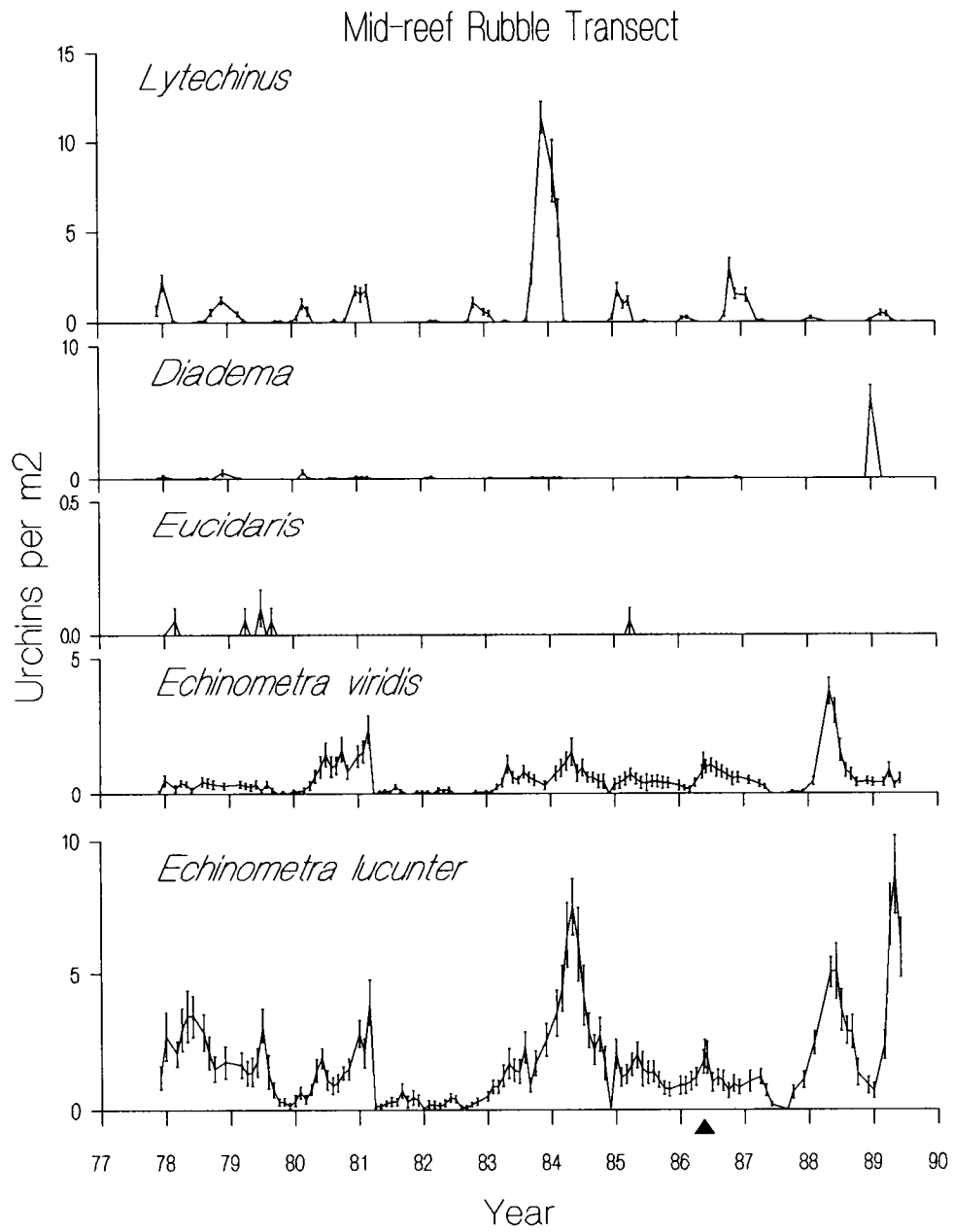


Figure 2.8 Populations of echinoid species on the reef flat at Punta Galeta (continued).

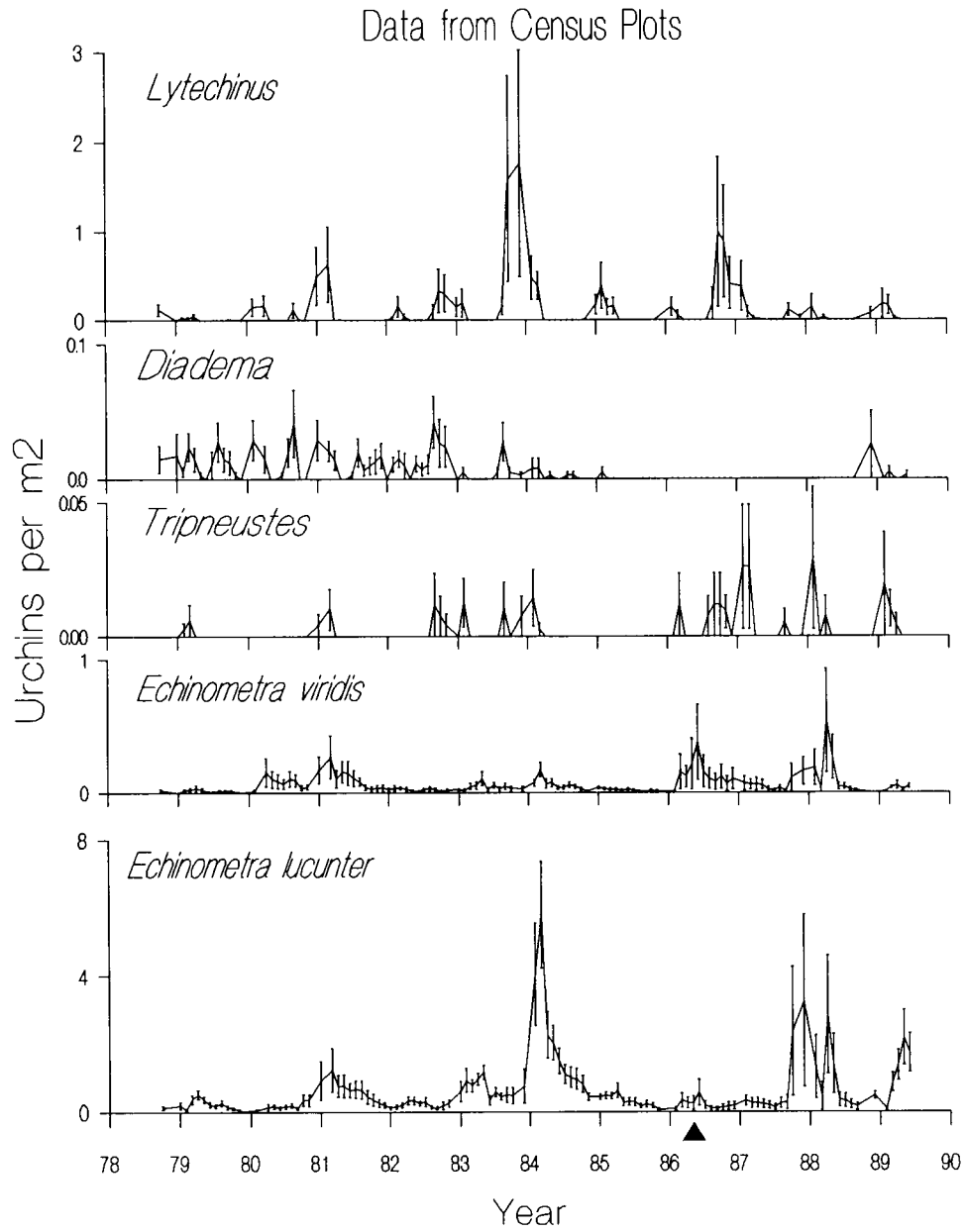


Figure 2.8 Populations of echinoid species on the reef flat at Punta Galeta (continued).

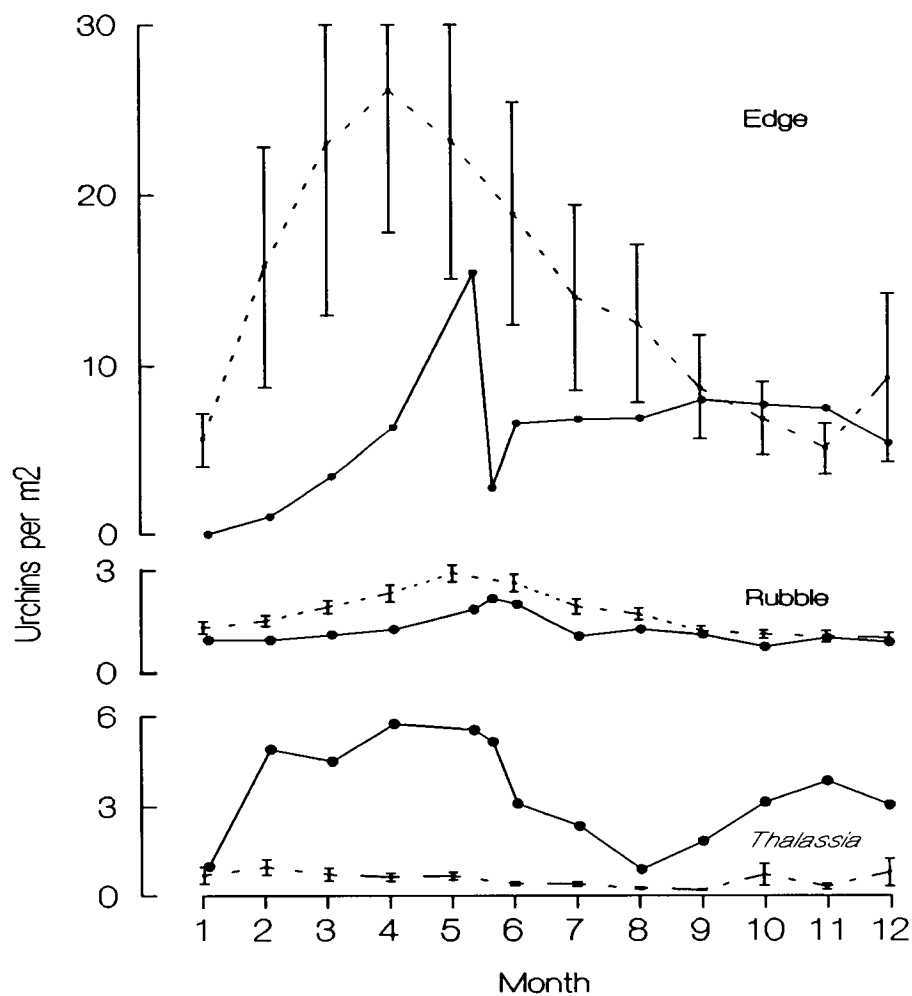


Figure 2.9 *Echinometra lucunter* populations in 1986 in relation to the average seasonal pattern from the 1978-1988 censuses. The 1986 populations are shown in solid lines and the 1978-1988 average pattern is shown as the dashed line with standard error bars. (Edge = reef edge transect; Rubble = east reef transect in area of coral rubble on the reef flat; *Thalassia* = mid-reef transect in *Thalassia* bed.) The first heavy slicks of oil arrived on 9 May 1986. Sea urchin populations in the reef edge transect showed a sharp reduction during the oil spill, but no such reductions were seen in the other transects.

the reef-edge transect. Urchin abundances in the east reef (rubble) transect showed little change. Populations in the remaining census plots were too low before the spill to detect significant decreases, whether they were affected or not.

The decreases occurred at the time of year when exposures of the reef flat above water level were frequent and prolonged (Cubit *et al.* 1986) and mortality of echinoids is naturally high (Hendler 1977a, b). To compare the decline in urchin populations at the time of the oil spill with the natural variability seen in previous years, June populations were regressed on May populations of the same year. This was done for both species of *Echinometra* and for all census areas. The regressions show highly significant May-June relationships for both species (Figure 2.10). Note that the r^2 would be higher if the post-spill outliers were eliminated, but leaving these out could be construed as biasing the data in favor of the hypothesis that oil damaged urchin populations; therefore, as the more conservative course, both pre- and post-spill data were used to calculate the regressions. Both *Echinometra* species in the reef-edge transect showed a larger than usual decrease from May to June 1986 (Figure 2.10). The May-to-June declines of *E. viridis* in the *Thalassia* bed transects were also unusual (Figure 2.10). These decreases are more apparent in graphs of the residuals plotted against population size (Figure 2.11). (Residuals are differences between the regression value and the actual value; i.e., the departure from the regression line as measured on the Y-axis. The residuals thus measure departures from the "expected" or long-term pattern.) In the graphs of the residuals, the departures of post-spill counts in the reef-edge transects cleanly fall out of the pack of counts for other years. For *E. viridis*, the post-spill counts in the *Thalassia* bed at Punta Galeta also were distinctly lower.

Recruitment continued after the oil spill. In the reef-edge transect, populations of both *Echinometra* species increased within six weeks after the heavy mortality in a pattern that was almost identical (Figure 2.8). High variability is a feature of sea urchin populations at all sites (Figure 2.12).

2.3.4 Growth of Sea Urchins

Examination for growth rings failed to demonstrate usable rings for growth, regardless of the plates or methods used. All methods using tetracycline, however, produced marker lines on the jaws of the sea urchins. Although the urchins were harvested less than 72 hours after treatment, the brightest marker lines were imbedded deep in the jaws, at a position usually interpreted to be approximately six months of growth (H. Lessios, pers. comm.). Fainter lines appeared at the margins of the jaws.

2.3.5 Core Samples

Analysis of the core samples is in a preliminary stage. To date, the infauna have been sorted to major taxon, measured, and counted from core samples, as follows. To obtain an initial overview of all sites, we are processing two samples per

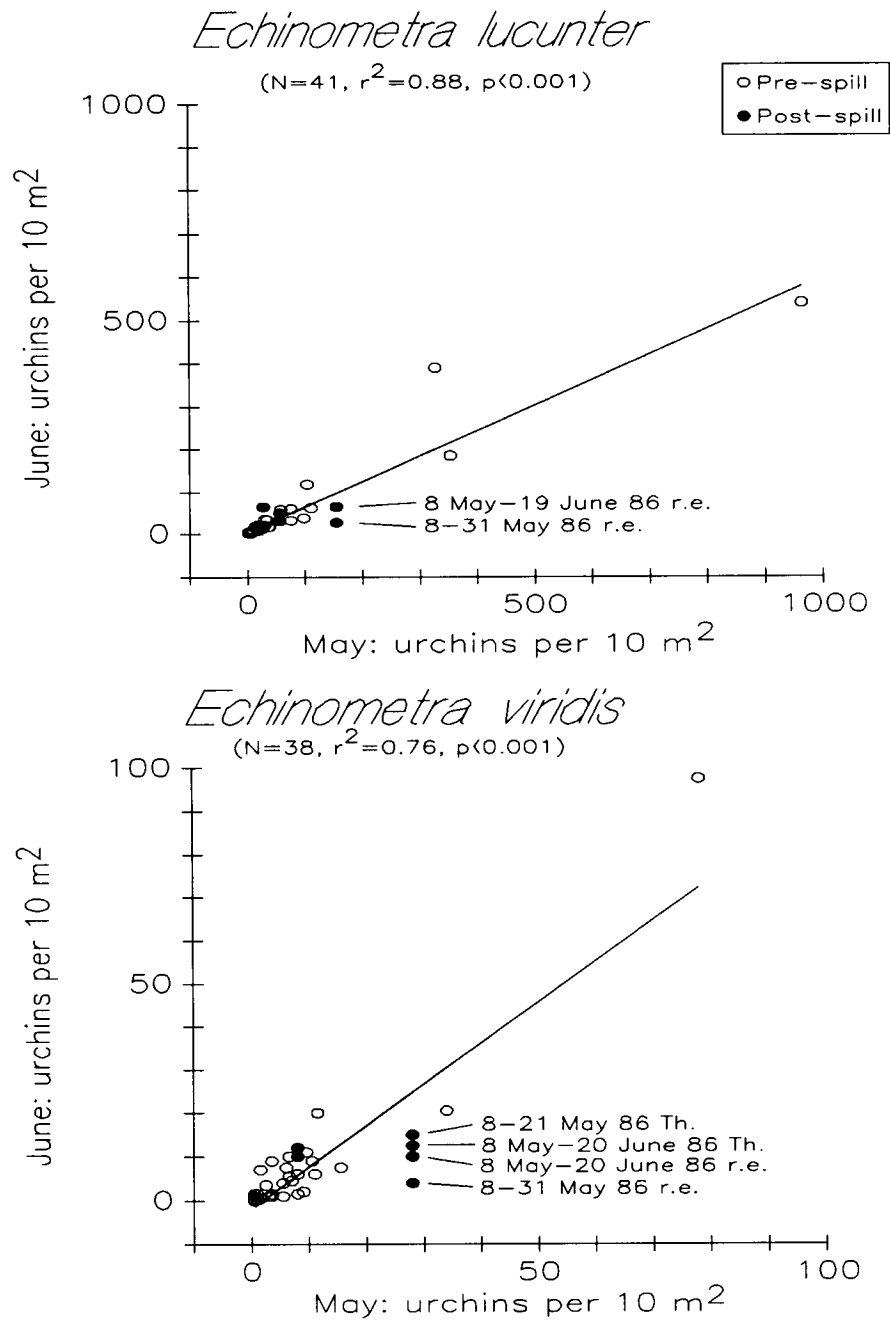


Figure 2.10 Abundances of urchins in June censuses compared with May censuses of the same years. (Th=*Thalassia* transect; r.e.=reef edge transect.)

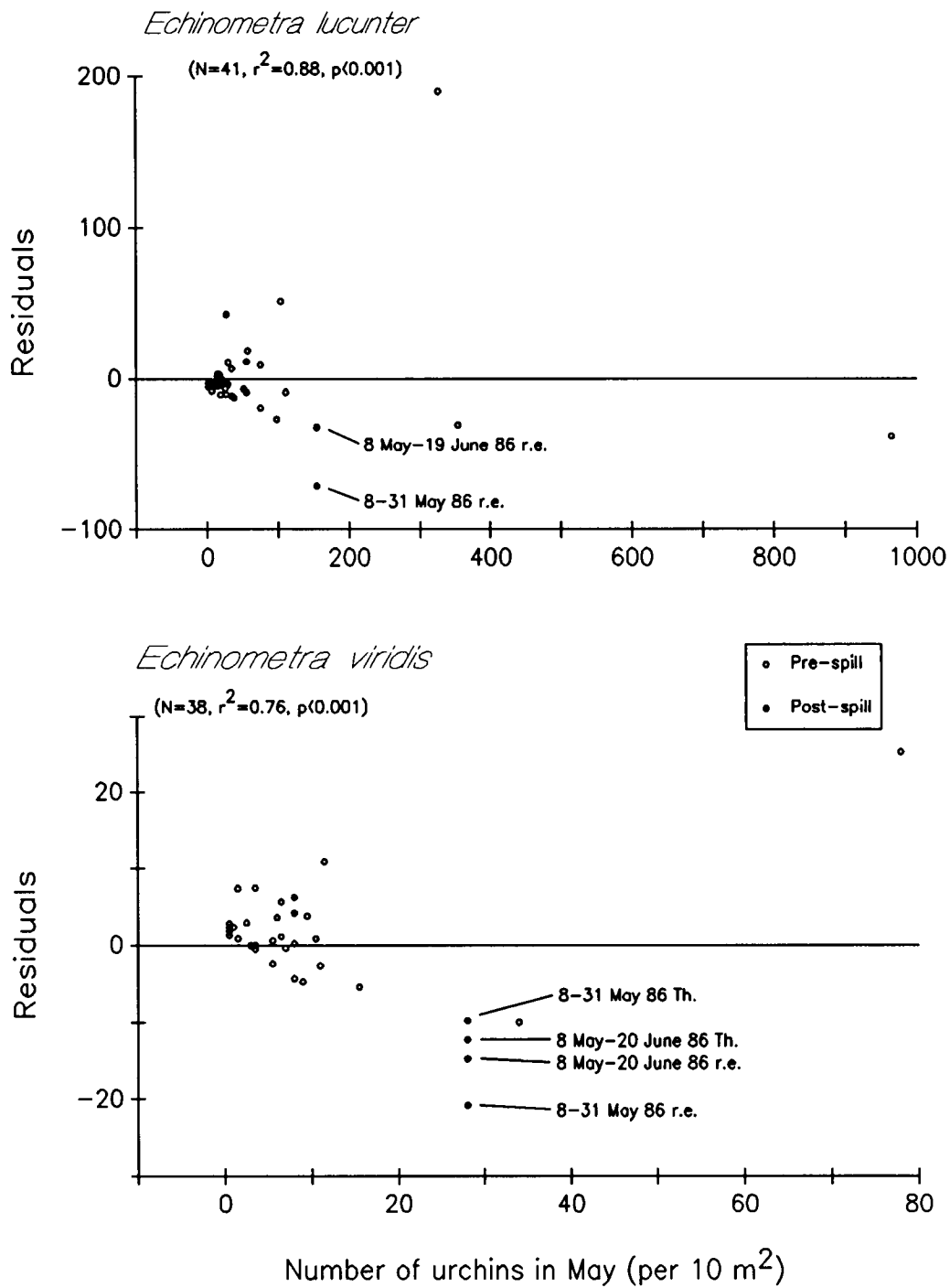


Figure 2.11 Residuals from May-June regressions of *Echinometra lucunter* population densities from previous figure, plotted against May population sizes. (Th=*Thalassia* transect; r.e.=reef edge transect.)

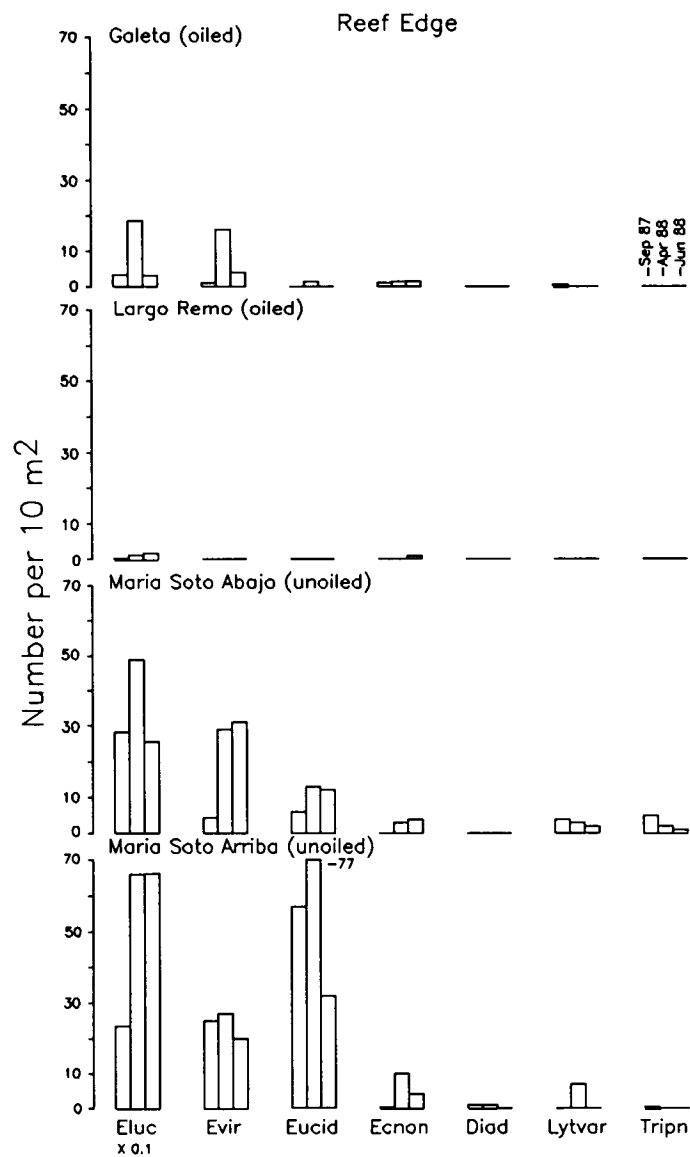


Figure 2.12 Among site comparisons of echinoid populations in post-spill censuses. The graphs on each page are for separate habitats: Reef Edge, Rubble, and *Thalassia* bed. As shown in the upper right of the figures, each cluster of three bars represents the surveys for September 1987, April 1988, and June 1988. Note that the abundances of *Echinometra lucunter* ("Eluc") are shown at one-tenth scale (i.e., a bar height of 30 in the graphs signifies 300 urchins per 10 m²). Species abbreviations are as follows: Eluc = *Echinometra lucunter*; Evir = *Echinometra viridis*; Eucid = *Eucidaris tribuloides*; Ecnon = *Echinoneus cyclostomus*; Diad = *Diadema antillarum*; Lytvar = *Lytechinus variegatus*; Tripn = *Tripneustes ventricosus*.

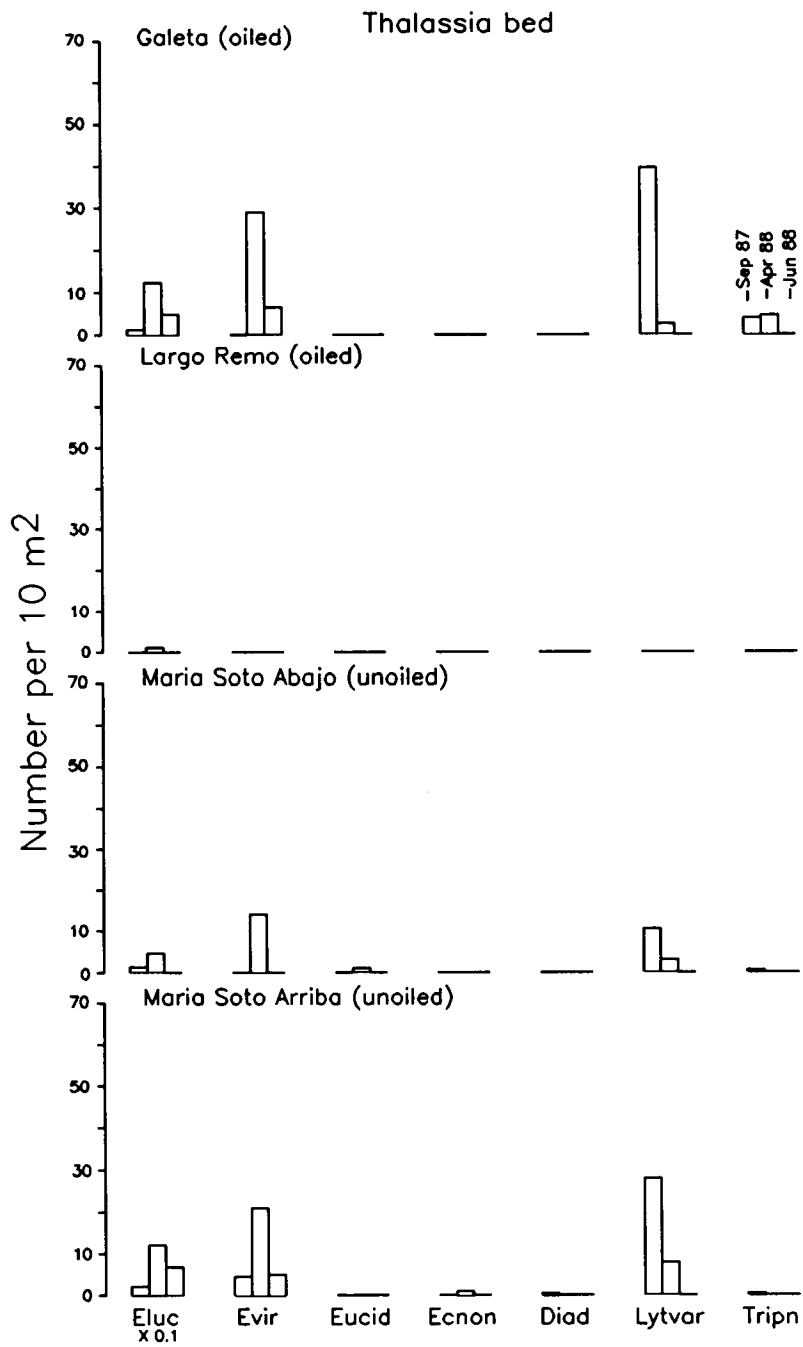


Figure 2.12 Among site comparisons of echinoid populations in post-spill censuses (continued).

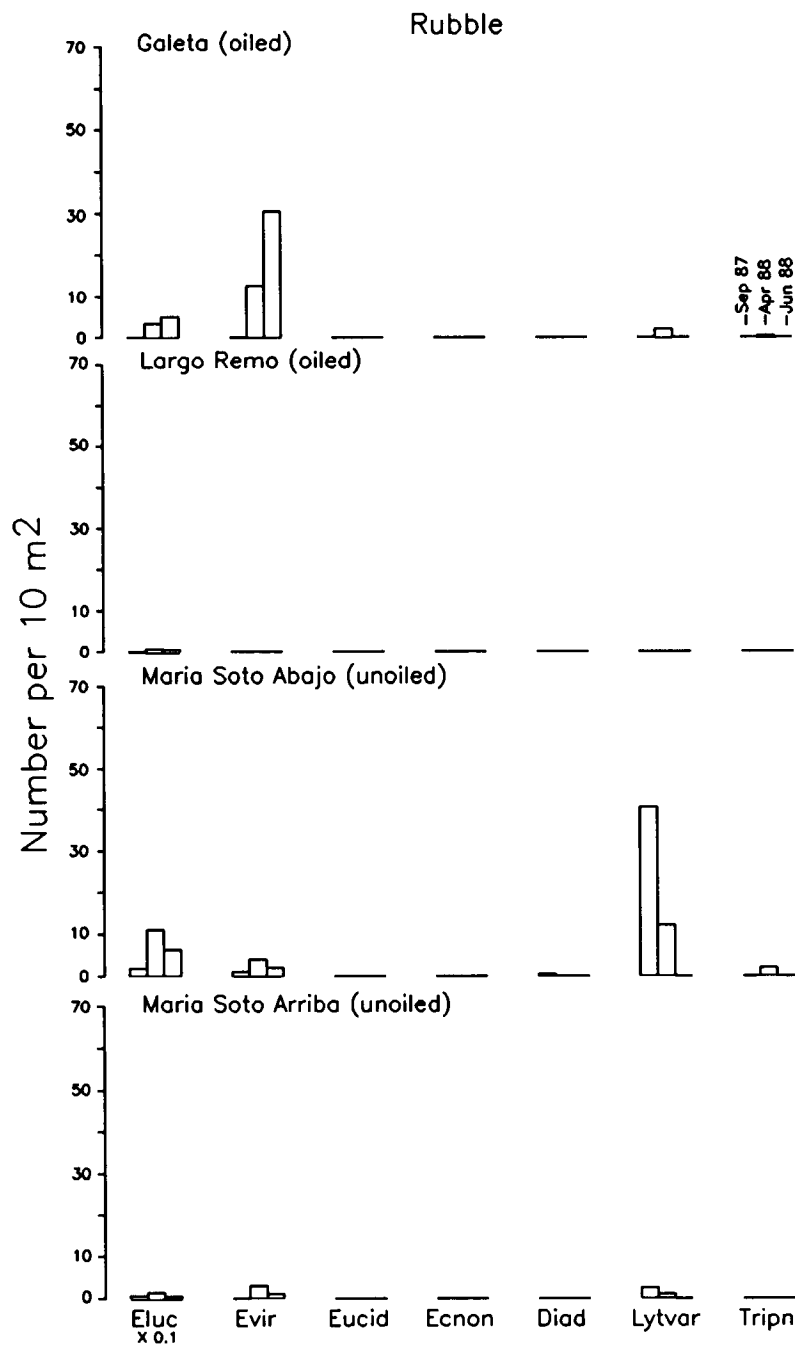


Figure 2.12 Among site comparisons of echinoid populations in post-spill censuses (continued).

cluster of three (see Methods, Chapter 2.2.4). One sample from each cluster from each site has been processed for the September 1987 through April 1989 collections. The first samples (September 1987) showed that the infauna had not been completely eliminated by the oil spill, but preliminary statistical analysis of these data indicates significant differences in the abundances and size distributions of most major groups of invertebrates.

2.4 Discussion and Conclusions

In the first survey after the oil spill in 1986, changes in the biota at Punta Galeta coincided with the spatial pattern of gross oiling of the reef flat. The greatest reductions in the abundances of perennial biota (the macrospecies) of the reef flat were closest to the seaward edge of the flat, where oil accumulated at low tide and the biota were directly immersed in oil rather than seawater. In tidepools and slightly deeper water, where the oil floated above the benthic organisms, we observed much less mortality. An exception to this pattern was the turf-forming alga *Laurencia papillosa*. Because the thalli of this alga dried at low tide, oil adhered to it over the full lengths of the transects. Afterwards the fronds died back to the basal tissue, reducing the percent cover of this alga. The alga quickly regenerated, however, and within five months after the oil spill had regained total abundances within the range measured before the spill. Continued spatial expansion of this and other species of algae has increased their space occupancy in the zone previously occupied by *Palythoa* and corals. As reported from other studies, the algae may have excluded the invertebrates (e.g., Loya and Rinkevitch 1980).

The frequent and extensive exposures of the reef flat above water level in 1988 provided a strong, natural test of the hypothesis that the post-spill reductions of the reef flat biota in 1986 were effects of stresses during seasonally low water levels that occurred naturally at the time of the oil spill. These seasonal stresses and their effects are well documented for the reef at Punta Galeta, producing natural declines in the abundance of the sessile macrobiota in the period between April and November (e.g., Birkeland *et al.* 1976; Cubit 1985; Cubit *et al.* 1986, 1988b). The spatial patterns of reduced abundances were much different between the post-spill censuses in June of 1986 and the post-exposure censuses of 1988. Moreover, in each situation the reductions in abundances corresponded to the expected mortality from oil (nearer the seaward edge) and from emersion stress (higher on the reef crest). This occurred even though the emersions of the reef flat were the worst in 15 years of records and the reductions in overall coverage were more severe than in any other survey. If natural physical stresses produced the post-spill reductions of biota near the reef edge in 1986, the reductions would have been even higher in 1988. They were not, indicating it is highly probable that the reductions in coverage of sessile biota and numbers of urchins after the oil spill were caused by oil and not by natural stresses.

The mortality of sessile species during the oil spill and the exposures of 1988 resulted in little bare substratum. In both cases the open space was immediately

colonized by a bloom of microalgae. The post-spill bloom developed as the oil was still coming ashore, suggesting these microalgae have exceptional tolerance to crude oil, or the crude oil was not particularly toxic to them. The latter explanation is more consistent with the pattern of mortality of the macrospecies: mortality was concentrated where the oil was in prolonged direct contact with the benthic biota, essentially replacing seawater with crude oil.

Both the post-spill and post-emersion blooms of microalgae occurred in the same spatial patterns as the mortality of the biota on the reef flat. The post-spill bloom was concentrated at the seaward edge of the reef flat and diminished going landward. The post-emersion bloom was concentrated near the crest of the reef flat, and diminished going seaward.

Blooms of benthic algae have been reported after oil spills elsewhere in the tropics (Russell and Carlson 1978) and in the temperate zones (Bellamy *et al.* 1967; Southward and Southward 1978): they appear to be a universal phenomenon. The cause of these blooms is not certain. The consensus explanation is that they are caused by mortality of herbivores (e.g., O'Brien and Dixon 1976; Southward and Southward 1978). This is consistent with our observations of the initial reductions in numbers of sea urchins, grazing crabs, and infauna, followed by a rapid regeneration of these herbivores and concomitant disappearance of the algal bloom. However, release from competition and stimulation by increased nutrients cannot be ruled out as contributing to the algal bloom (Russell and Carlson 1978). To our knowledge, the information from this study is the only before-and-after quantitative description of such a bloom and its demise following an oil spill.

Much still remains to be established by the continuing analysis of the data. For example, surveys during the extreme emersions of the reef flat environments will also provide additional critical examinations of the effects of oil. By analyzing changes over time in the oiled and unoled sites, we can examine the hardiness and resilience of populations at the two categories of sites in responding to extreme stresses. At the time of the 1988 emersions, these populations had much different histories: populations at the oiled sites were recently regenerated, or were still in the process, but the populations at the unoled sites were relatively old and established. Analysis of the most recent data, which is now underway, should measure the relative ability of the younger, reestablishing populations to resist stress, recuperate after stress, or both.

Although the analysis of the core samples is still in the preliminary stages, it is clear from the samples processed so far that infauna were not permanently removed in the two heavily oiled sites. Most of the infauna are arthropods (e.g., decapods, tanaids, amphipods, and isopods), which have hydrophobic exoskeletons and respiratory systems that should have made them susceptible to being killed by oil. Moreover, the *Laurencia* beds were heavily oiled and deteriorated over wide areas. None of these animals appeared in the samples taken in June 1986. Therefore, we expected to find conspicuous signs of severe population reductions at a scale large enough to cause slow population recovery. The abundant, diverse infauna at the oiled sites indicates recolonization was rapid. Preliminary statistical

analyses, however, indicate a persistent difference in the size-abundance distributions of the infauna, which may indicate the demography of these populations was affected by mortality and recruitment patterns during and after the oil spill.

As more of the past databases are incorporated into these analyses, more data will be brought to bear on the pre- and post-spill comparisons. These data extending from the early 1970's will provide a strong basis for distinguishing the effects of oil from the events that occur naturally.

Remaining to be investigated are the changes in the reef-flat communities associated with the expected release of oiled sediment from the dead and decomposing mangroves backing the reef flats at the oiled sites. Mass release of fine sediments alone from these oil-killed mangroves has destructive potential for marine biota. Such mass releases, combined with the oil entrapped in the sediment, may cause even more severe effects. Certainly the oil-sediment combination will be heavier than the original oil itself, and therefore more likely to come in prolonged contact with the submerged biota. To our knowledge, such second-stage impacts of oil spills have never been investigated, despite their obvious implications for the management of oil spills and clean-up operations.

Chapter 3

Effects of an Oil Spill on the Gastropods of a Tropical Intertidal Reef Flat

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3.1 Introduction

On April 27, 1986, an oil storage tank at the refinery on Isla Payardi in the Republic of Panama ruptured, spilling at least 50,000 barrels of medium weight crude oil into the Caribbean Sea (Cubit *et al.* 1987, 1988a; Jackson *et al.* 1989). The Galeta Laboratory of the Smithsonian Tropical Research Institute, located on a nearby reef flat, has been the focus of detailed investigations since 1968 (e.g., Birkeland *et al.* 1976; Cubit and Williams 1983). Oil began washing ashore at Punta Galeta in early May, as the area between the Caribbean entrance to the Panama Canal and the town of María Chiquita was extensively oiled (see Figure 1.1).

Gastropod molluscs are abundant and diverse consumers on many tropical shores (Vermeij 1978), but relatively little is known about molluscs on coral reef flats (Kohn and Leviten 1976; Leviten and Kohn 1980). The relative abundance and size structure of molluscs in different habitats on the Galeta Navy Reef, located approximately 0.5 km west of Punta Galeta Reef (Figure 1.1), were sampled quantitatively in 1982-3 (Garrity and Guttierrez, unpublished). These are the most comprehensive data on gastropod ecology at Galeta Navy Reef before the spill (but see also Radwin 1969; Birkeland *et al.* 1976; Brattstrom 1985).

Little information exists on even short-term effects of oiling on the molluscs of reef flats (Birkeland *et al.* 1976; Eisler 1973, 1975a, b; Chan 1976, 1977; Maynard *et al.* 1977; Maynard 1984). In August 1986, roughly three months after oil began coming ashore on the Punta Galeta reef flat, we initiated quarterly monitoring of snail abundance and size structure both at Galeta Navy Reef and at unoiled sites, using methods identical to those in 1982-3.

These data provide an opportunity to examine effects over time of a major oil spill on the gastropods of a tropical reef flat. We present (1) data taken at Galeta Navy Reef before the oil spill, (2) data from twelve quarterly, post-spill monitorings of the Galeta Navy Reef site (oiled) and (3) comparative data from twelve quarterly monitorings of control (unoiled) reef flats. A control site was oiled by a small diesel

spill in May 1988; we discuss its effects and recovery of this site relative to overall results, through May 1989.

3.2 Methods

Six discrete habitats (= zones) within the reef flat were sampled (Figure 3.1). Running roughly from the shore toward the reef's seaward edge, they included: high rubble; low rubble; sand and reef rock; and beds of seagrasses, primarily *Thalassia testudinum*, and algae, primarily *Laurencia papillosa*. The high rubble habitat was on the reef's shoreward margin and consisted of an elevated, consolidated matrix of dead coral, shell fragments and sand, with some loose rubble and shell (Figure 3.2). It was never submerged, although waves might splash or surge onto the high rubble habitat during extremely rough weather (pers. obs.). The low rubble habitat occurred just below and seaward; it was less consolidated, and consisted mainly of large fragments of coral rubble embedded in sand, and other relatively loose debris (Figure 3.3). This habitat lay partly within the intertidal zone; it was regularly to intermittently submerged and emersed, depending upon tidal cycle and wave action. Movement of snails between the high and low rubble zones was possible because the zones were contiguous sections of shore. The sand habitat (Figure 3.4) also occurred at the shoreward margin of reef flats, partly in and partly below the intertidal zone (Figure 3.1). We monitored snails in the intertidal portion of this habitat. The reef-rock habitat occurred seaward of the above habitats, and was a relatively horizontal area of jagged and pocked rock (Figure 3.5). It was well within the intertidal zone, so underwent fairly regular cycles of submergence and emersion. It was physically isolated from the shoreward zones by deeper water. *Thalassia* beds (Figure 3.6) have been described by Heck (1977). Located within the lagoon and in deeper areas of the reef flat, they were partly emersed during low tides. *Laurencia* beds (Figure 3.7, see also Connor 1984) occurred near the seaward fringe of the reef flat and were exposed above water level only when calm seas occurred along with minus tides.

Cubit *et al.* (1986, 1989) discuss thoroughly the details of intertidal exposures at Punta Galeta. Tidal fluctuations are small and often overridden by winds and currents, leading to an irregular pattern of submersion and emersion of the reef flat. In general, water levels and wave action are high in the late wet (December) and especially dry seasons (January - March). In the early rainy season (April - May), onshore winds cease and water levels drop, often leading to long periods of exposures of the reef flat above water level. Exposures of the reef flat occur most often in this part of the year, but may happen at any time when winds and tides are both low.

Monitoring was done during daylight, and generally when water levels and wave action were low to moderate. Within each habitat, gastropod abundance was estimated as follows: 0.25 m² quadrats were laid at 20 to 30 randomly selected points along a 50-100 m transect line, and all gastropods visible within each quadrat were identified and counted. We did not sample cryptic or burrowing gastropods because sampling would have involved considerable disturbance to most habitats.

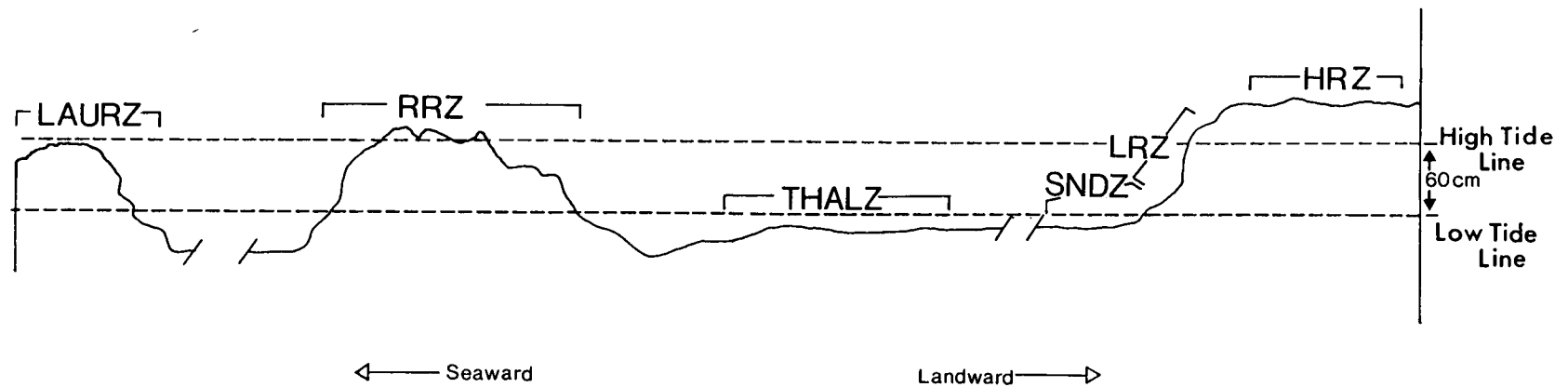


Figure 3.1 Composite diagram of the six reef flat zones. Tide limits are approximate, and actual water levels may exceed upper and lower limits because of weather conditions. See text. *Thalassia* zone shown as subtidal, but seagrass blades may be exposed to air during low tide. LAURZ=*Laurencia* zone, RRZ=reef rock zone, THALZ=*Thalassia* zone, SNDZ=sand zone, LRZ=low rubble zone, HRZ=high rubble zone.

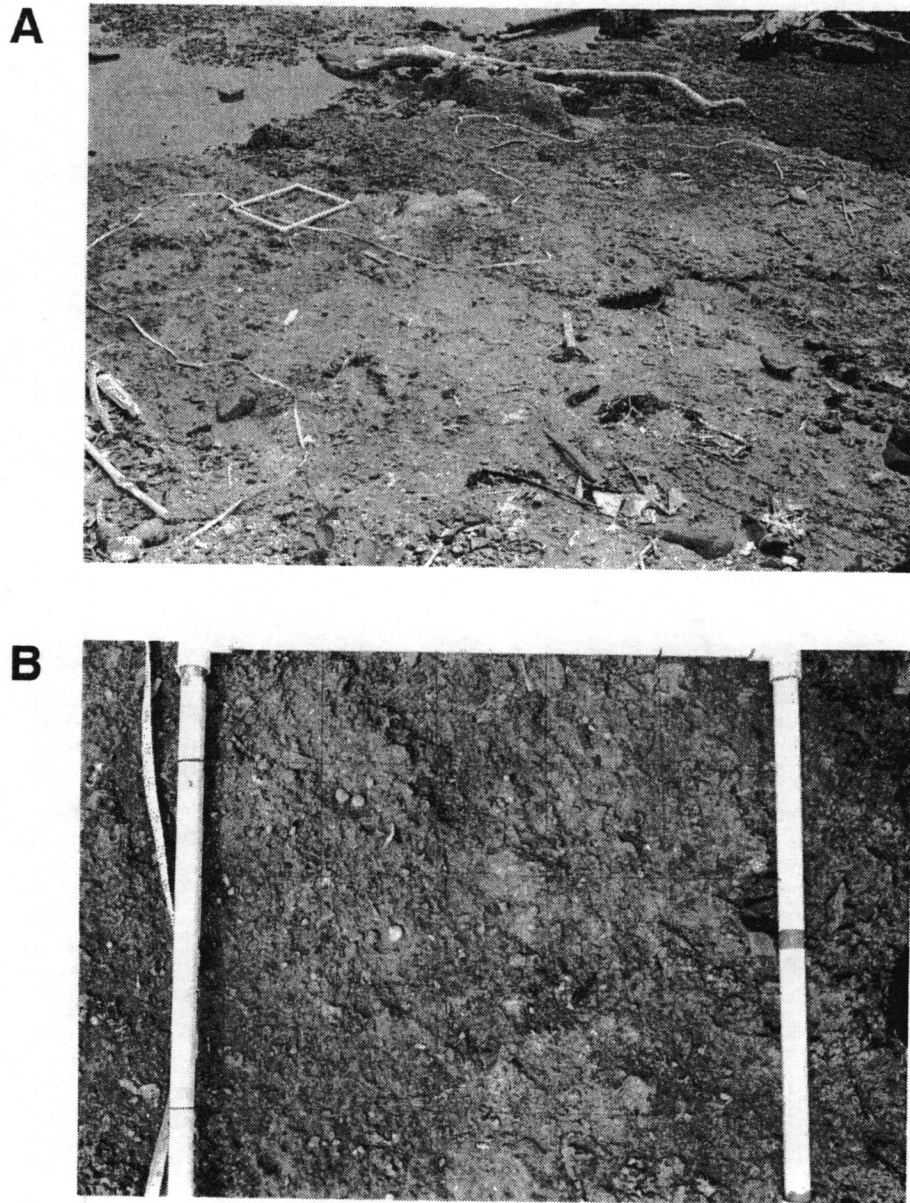


Figure 3.2

High Rubble Zone, May 1989.

A. General view, facing seaward, Toro Point. Quadrat is 0.5x0.5 m. Note transect tape and storm debris.

B. Closeup of 0.25 m² quadrat, Toro Point. Note consolidated coral rubble substratum. Visible snails include *Tectarius*, *Nodilittorina* and several species of *Littorina*.

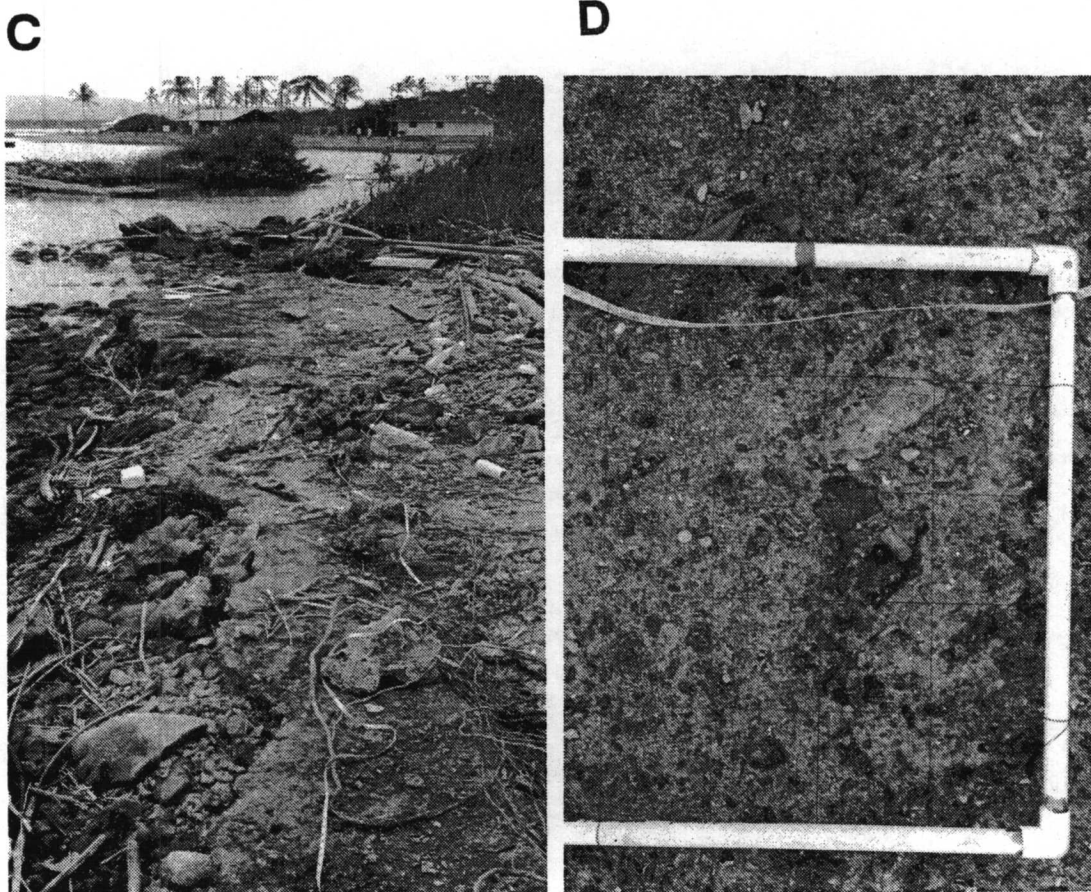


Figure 3.2 High Rubble Zone, May 1989 (continued).

C. General view, Galeta Navy Reef. Note storm debris and erosion of zone.

D. Closeup of 0.25 m² quadrat, Galeta Navy Reef. Visible snails include *Tectarius* and *Littorina lineolata*.

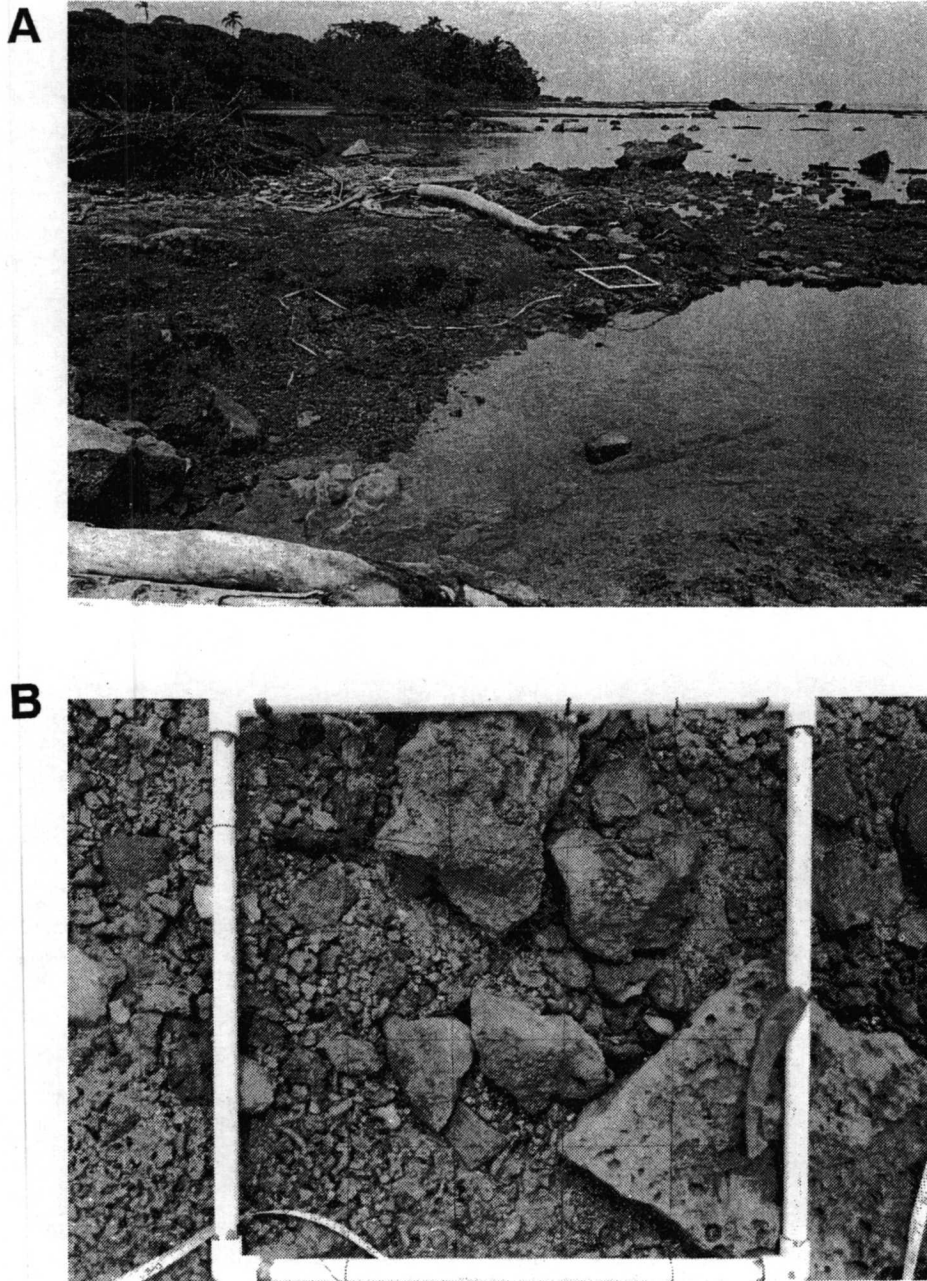


Figure 3.3 Low Rubble Zone, May 1989.

A. General view, Toro Point. Quadrat and line are in the low rubble zone; high rubble zone visible above and to the left. Reef rock zone visible in the background at the extreme upper right.

B. Closeup of 0.25 m² quadrat, Toro Point. Note unconsolidated nature of the substratum relative to the high rubble zone. Visible snails are *Planaxis nucleus* in the lower left of quadrat.

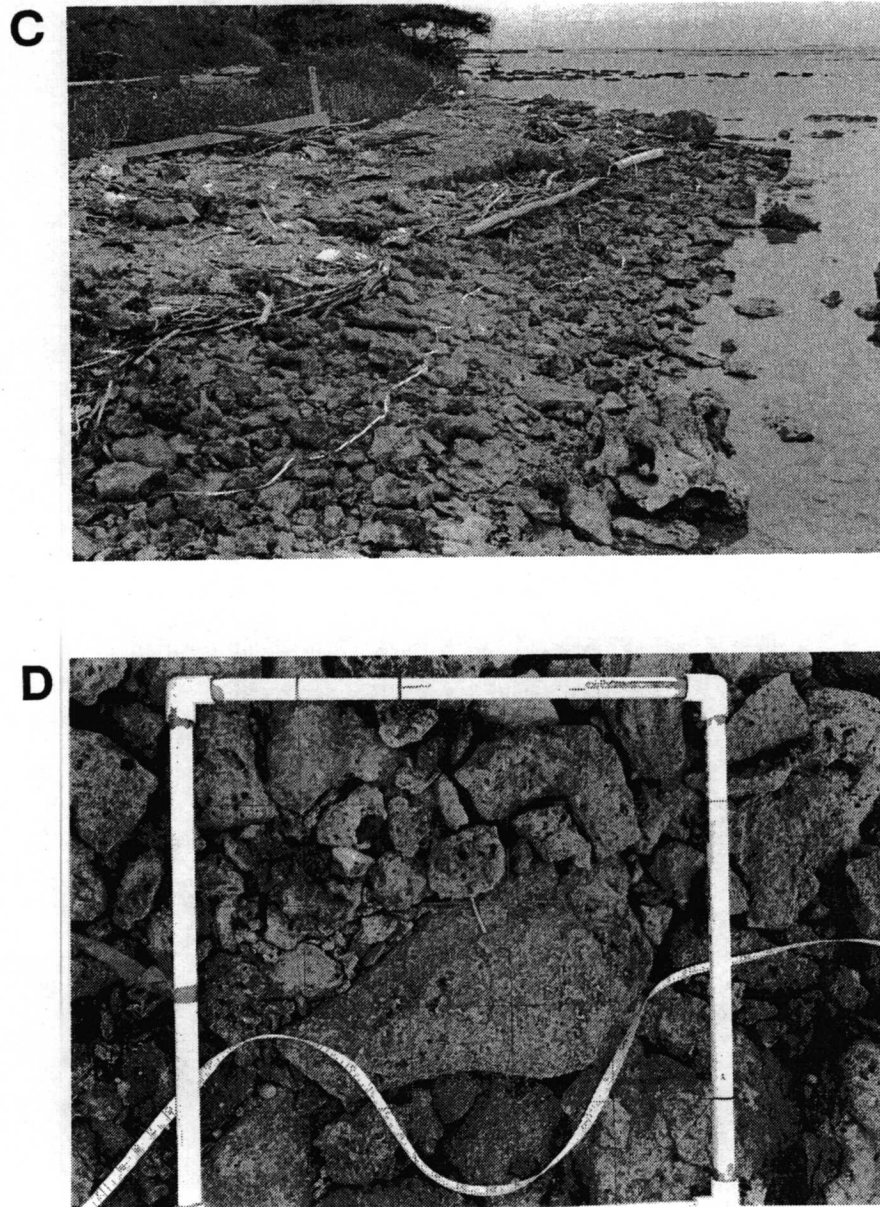


Figure 3.3 Low Rubble Zone, May 1989 (continued).

C. General view, Galeta Navy Reef. Note litter at high tide mark.

D. Closeup of 0.25 m² quadrat, Galeta Navy Reef. No visible snails; note tarballs in lower right quadrat and outside of the quadrat to the upper right.

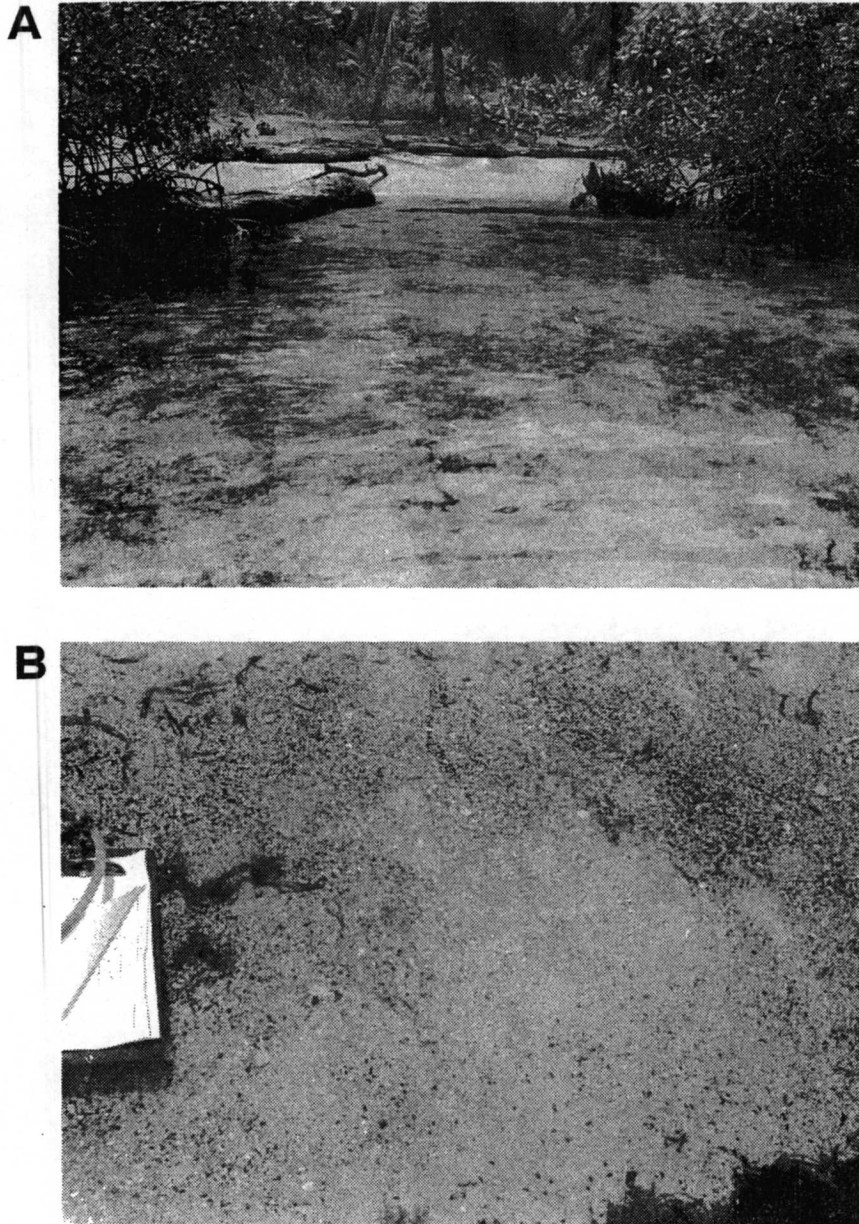
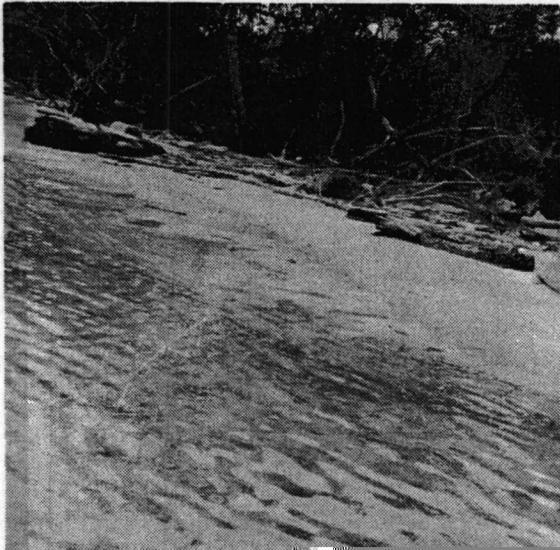


Figure 3.4 Sand Zone, May 1989.

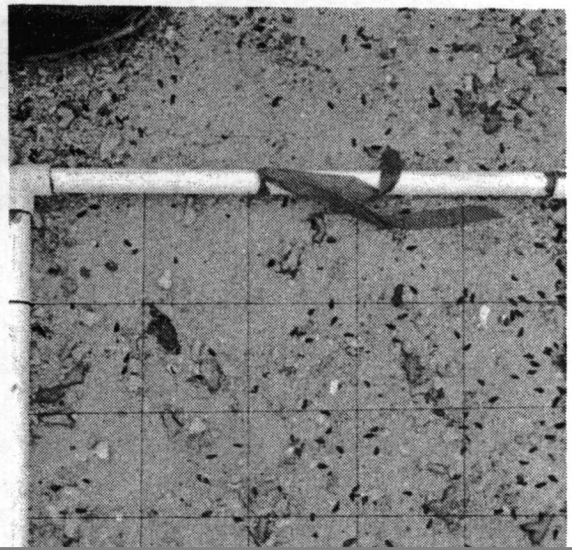
A. General view, María Soto. Note lack of fringing hard substratum. Trees are red mangrove, *Rhizophora mangle*.

B. Closeup of 0.25 m² quadrat, María Soto. Clipboard visible to left for scale. Small black snails in the foreground are *Batillaria minima*.

C



D



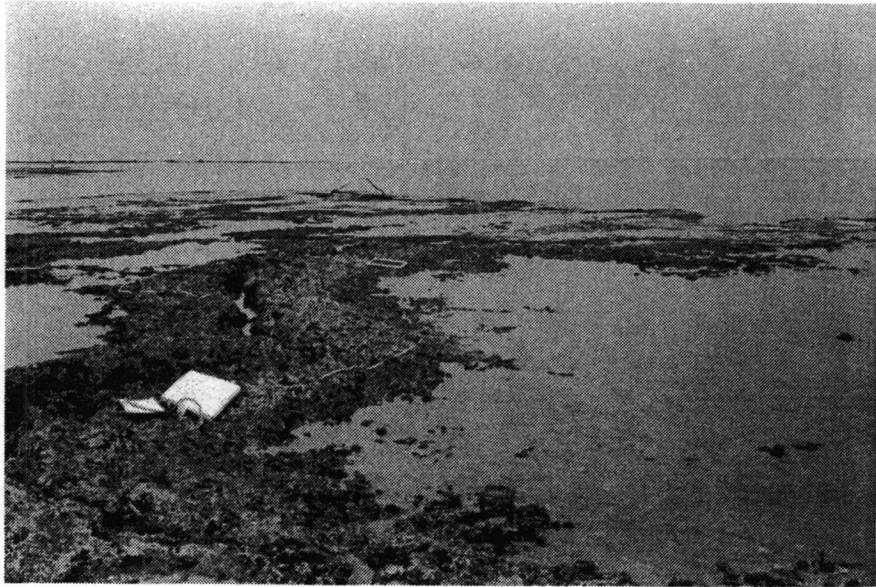
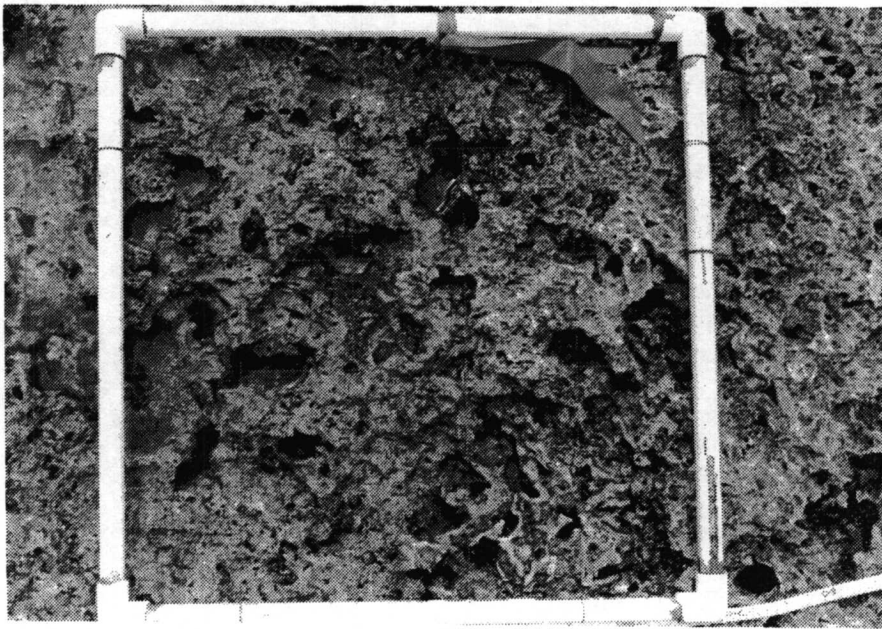
A**B**

Figure 3.5 Reef Rock Zone, May 1989.

A. General view, Toro Point.

B. Closeup of 0.25 m² quadrat, Toro Point. Note pocked and creviced nature of rock. The chiton *Acanthopleura granulata* is in the upper left of photograph.

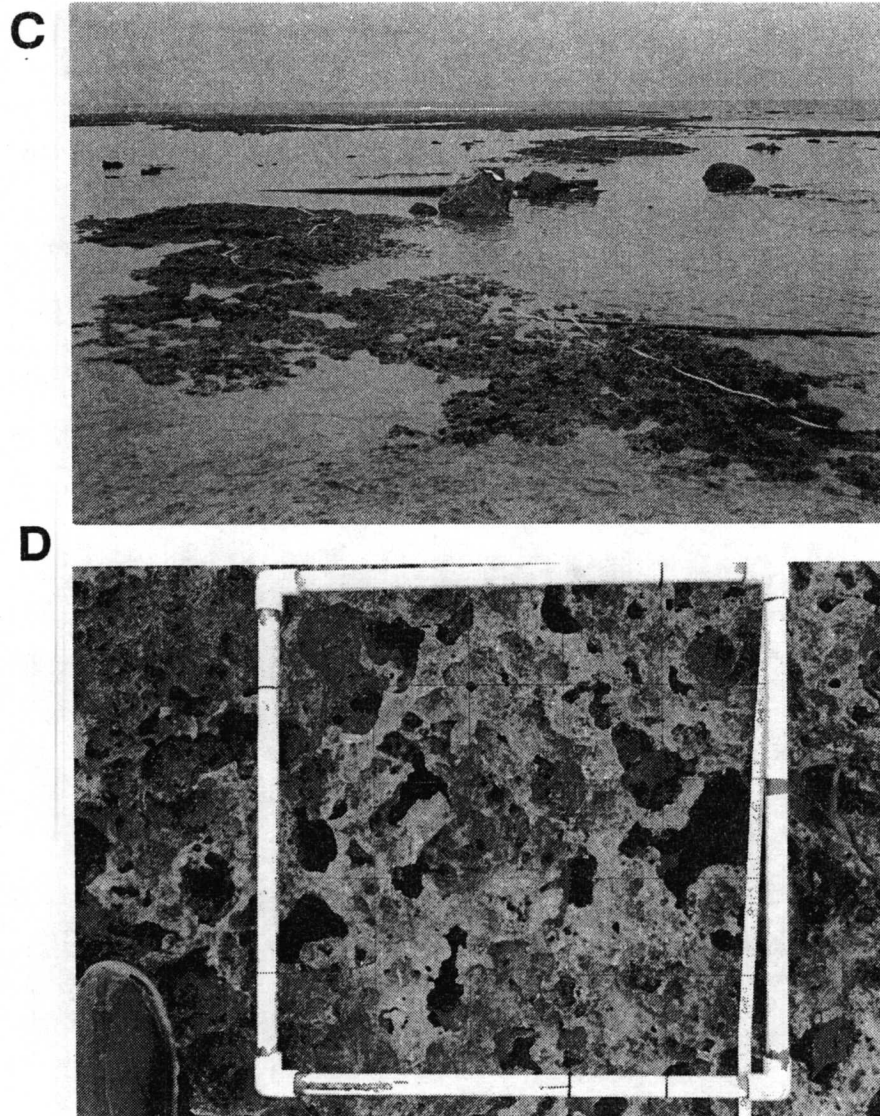


Figure 3.5 Reef Rock Zone, May 1989 (continued).

C. General view, Galeta Navy Reef. Note how deeper water isolates zone.

D. Closeup of 0.25 m² quadrat, Galeta Navy Reef. Substratum is pocked and creviced as at Toro Point.

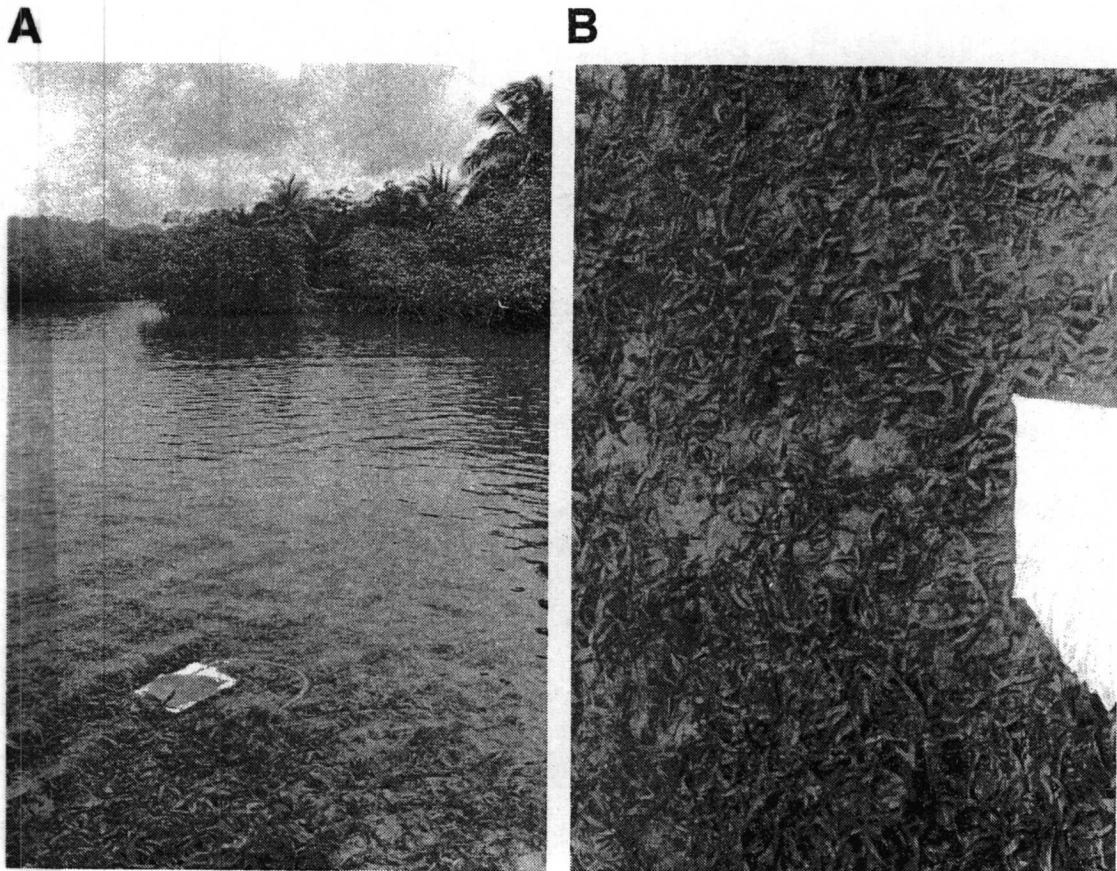


Figure 3.6 *Thalassia* Zone, May 1989.

A. General view, María Soto, high tide. Object in foreground is a clipboard for scale.

B. Closeup of 0.25 m² quadrat, María Soto. Clipboard visible to left for scale.

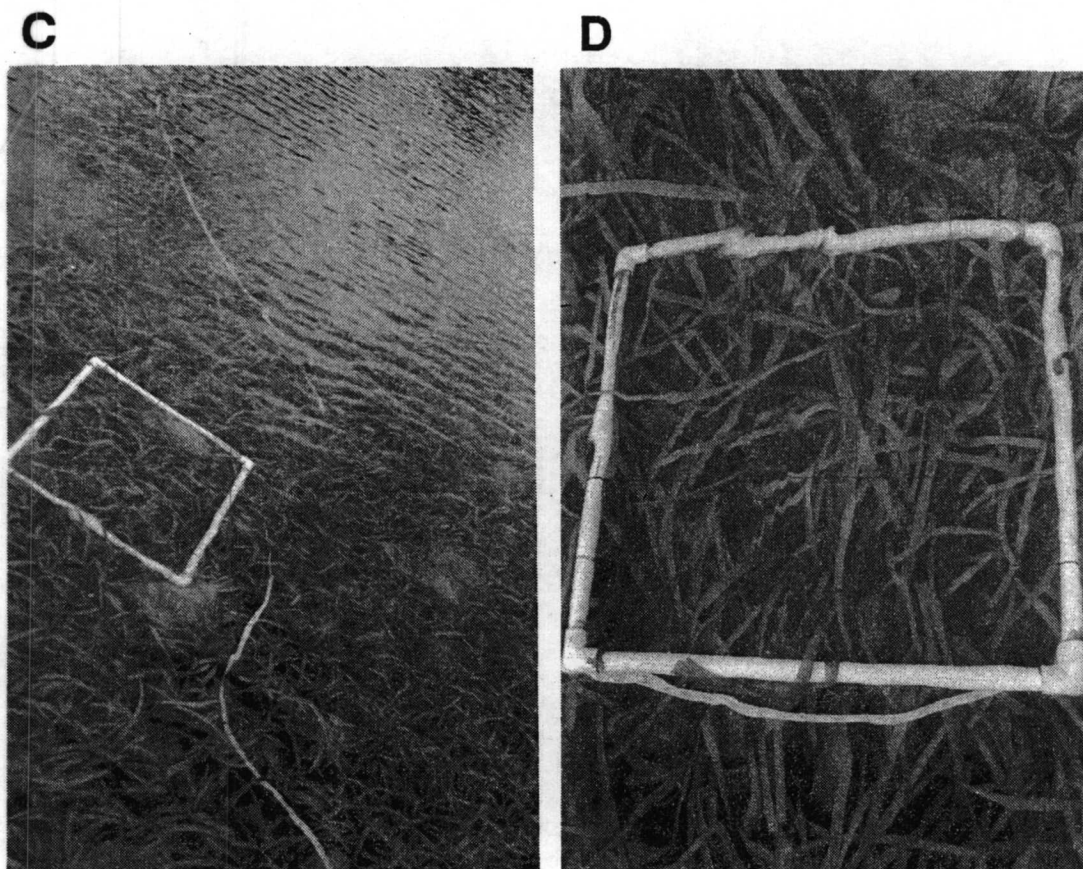


Figure 3.6 *Thalassia* Zone, May 1989 (continued).

C. General view, Galeta Navy Reef, high tide.

D. Closeup of 0.25 m² quadrat, Galeta Navy Reef. No snails visible.

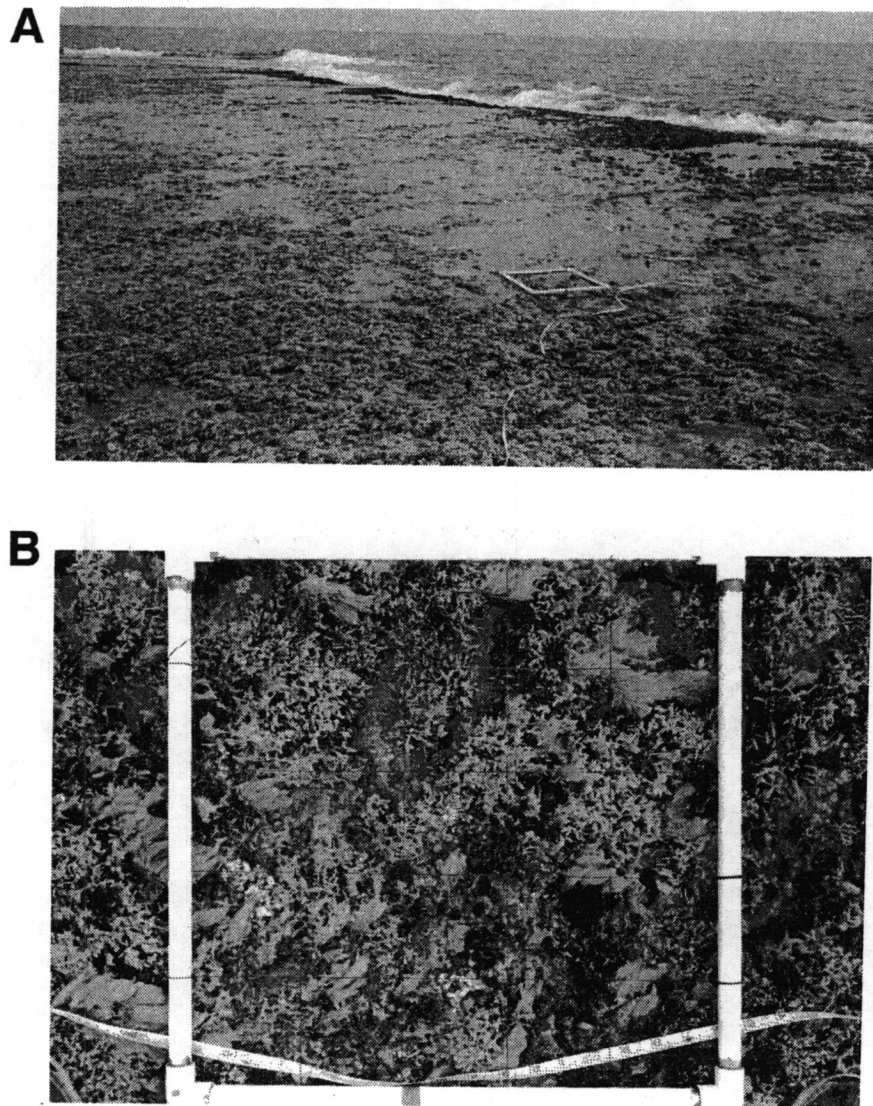


Figure 3.7 *Laurencia* Zone, May 1989.

A. General view, Toro Point. Ships anchored in background are awaiting transit through the Panama Canal.

B. Closeup of 0.25 m² quadrat, Toro Point. Note bleaching of some algae.

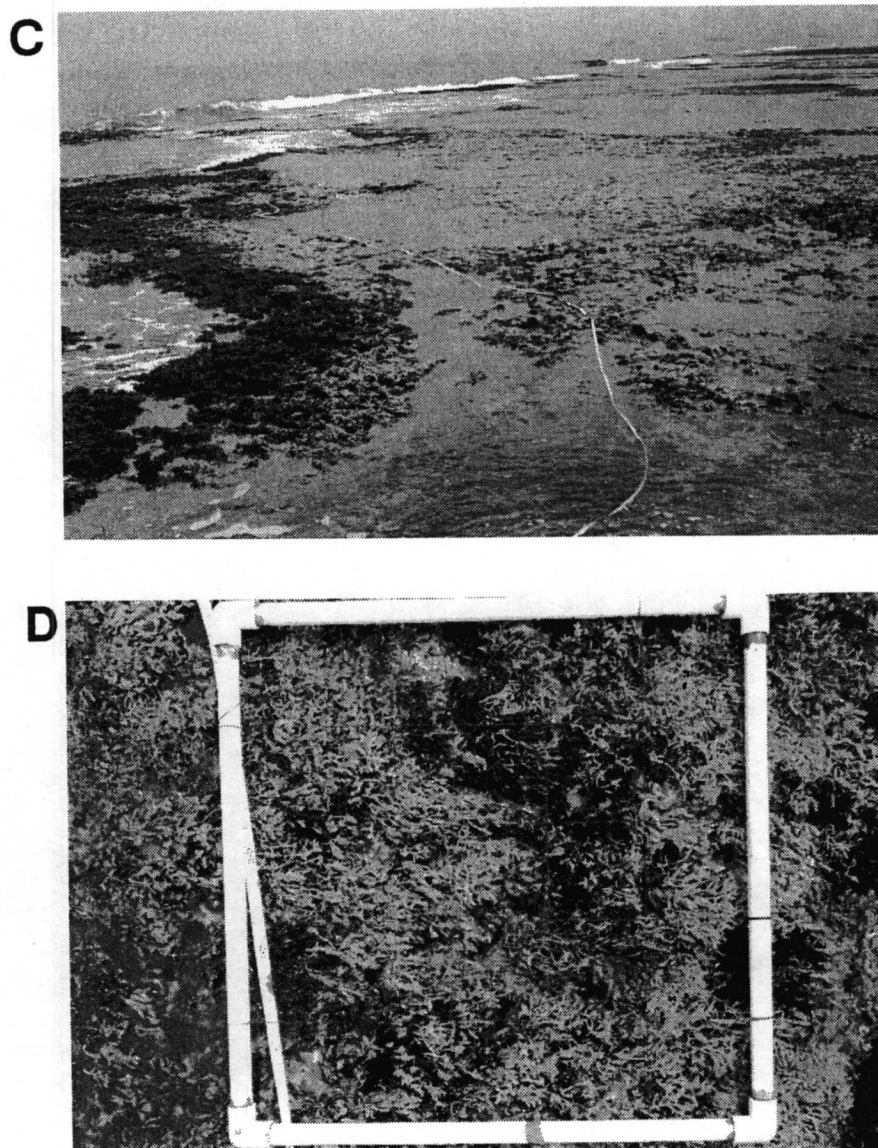


Figure 3.7 *Laurencia* Zone, May 1989 (continued).

C. General view, Galeta Navy Reef. Most algae are bleached except those on the outer fringes of the reef.

D. Closeup of 0.25 m² quadrat, Galeta Navy Reef. No snails visible. Most algae are bleached.

To examine the size structure of common species of snails, individuals (usually ≥ 200) of each species were collected and shell lengths measured to the nearest millimeter. All snails were collected by hand from a series of contiguous, 0.25 m² quadrats running from each zone's upper to lower limits. Forceps were used to collect tiny individuals and those in crevices. Snails were measured with vernier calipers and returned immediately to the appropriate habitat. Snail species were identified and vouchers retained for verification. We use generic names in the text for genera with only one species present at our study sites (e.g., *Tectarius* for *Tectarius muricatus*). Size data were used to test for size-specific mortality following the spill and to examine patterns of recruitment over time. A species was considered to have recruited if a pulse of small individuals that was previously absent appeared. Because monitoring was quarterly, pulses of rapidly growing species could have been missed; estimates of recruitment should be considered conservative.

Pre-spill monitoring at Galeta Navy Reef was done between November 1982 and January 1983. It was first remonitored during July and August 1986, three months after oiling. Less than one week after this monitoring, a clean-up crew manually removed oil and oiled debris from the high and low rubble habitats and used water hoses to blow oil from the sand habitat. These three habitats were remonitored within two weeks to assess changes brought about by cleaning. Reef flat habitats at Galeta Navy Reef have been monitored at quarterly intervals since.

The following additional data from Galeta Navy Reef were collected soon after the spill. In August 1986, the amount of oil adhering to the substratum in each habitat was assessed. In the high rubble, low rubble and reef rock habitats, percent cover of oil was visually estimated within 20 randomly placed 0.25 m² quadrats. Extensive field notes were taken of oiling in the intertidal sand habitat and in *Laurencia* and *Thalassia* beds, where the nature of the substratum precluded percent cover measurements. During August 1986 sampling, snails within quadrats were recorded as having oiled or unoiled shells. In both August and November 1986, dead snails that were either (1) cemented by oil to the rock in the low rubble habitat or (2) found in the high rubble habitat (where movement of shells by wave action did not occur) were separately counted, along with live individuals in abundance transects. Dead snails were collected in these two habitats for size measurements. Size-selective mortality was examined by comparing the size frequency distributions of live and dead snails collected at the same time and in the same habitat.

The section of reef flat sampled was in front of the U.S. Navy antenna site and was thus protected from human interference (Galeta Navy Reef, GNRF, Figure 3.8). Beginning in August 1986, unoiled reef flats were sampled similarly for gastropod distribution, abundance and size structure. Two unoiled sites were used because no one area had all the habitats found at Galeta Navy Reef (Figure 3.8). The unoiled flats were located (1) west of Toro Point, where high rubble, low rubble, reef rock and *Laurencia* were monitored (TPR); and (2) seaward of María Soto,

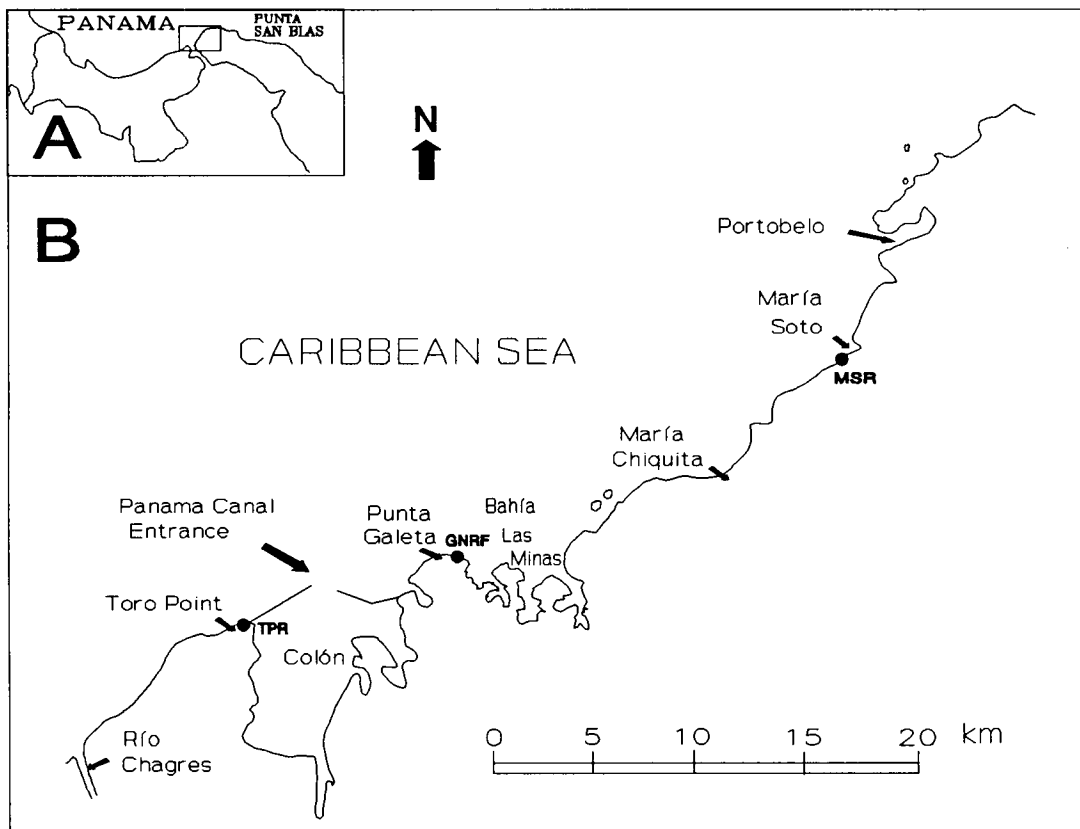


Figure 3.8 Map of study sites. (A) Location of study area in Panama. (B) GNRF = Galeta Navy Reef (oiled), TPR = Toro Point (unoiled), MSR = María Soto (unoiled). See Figure 1.1 for further details.

where we monitored gastropods found on sand and in the *Thalassia* bed (MSR).

At both Toro Point and Galeta Navy Reef, some erosion of the surface was observed in the high rubble habitat during the dry season (December - March) of 1987-88. This erosion followed periods of heavy wave action. Using the same quadrats used to measure density in May 1988, we visually estimated the extent of erosion as percent cover of newly eroded substratum. Further, on May 14, 1988, a week prior to quarterly monitoring, at least part of Toro Point was contaminated by a small diesel spill from a grounded sailboat. The monitoring immediately after this event is marked by an arrow on all figures, and observations are presented in the appropriate sections below. We again estimated percent cover of oil in the high rubble, low rubble and reef rock zones at both Toro Point and Galeta Navy Reef in November 1988.

Time considerations limited the monitoring to one replicate of each of the six oiled and unoiled habitats (henceforth referred to as "zones"). This lack of replication of zones within treatments prevents direct statistical comparison between oiled and unoiled treatments. Our data are thus best used to indicate the magnitude of reductions in mollusc populations at Galeta Navy Reef from 1983 levels and to follow population events there over time. Population differences for common species between unoiled (Toro Point and María Soto) and oiled sites (Galeta) over time are also useful; we suggest some of these are related to oiling.

3.3 Results

3.3.1 Gastropod Abundances Prior to Oiling

Table 3.1 shows mean densities for gastropod species found in six reef flat habitats at Galeta Navy Reef in 1982-1983. The high rubble zone was characterized by the large-bodied, herbivorous snail *Tectarius muricatus* (mean density = 6.1/0.25 m²) and the smaller *Littorina angustior* (10.8/0.25 m²). *Nodilittorina tuberculata* (2.8/0.25 m²) and *L. lineolata* (3.6/0.25 m²) were also relatively common while *L. ziczac*, *Nerita versicolor* and *Melampus coffeus* were rare (< 1 snail/0.25 m²).

Nine species occurred in the low rubble habitat at Galeta Navy Reef prior to the spill. *Nerita tessellata* and *N. versicolor* were most abundant (~12/0.25 m²), and *N. fulgurans* and *N. peloronta* were rare (< 1 snail/0.25 m²). *Littorina lineolata*, *L. angustior* and *L. ziczac* occurred in roughly the same abundance as in high rubble while *Nodilittorina* (we use generic names for genera with only one species present) was rarer (< 1 snail/0.25 m²) compared to higher on the shore. *Planaxis nucleus*, which forages under rocks, was present in low abundance (< 1 snail/0.25 m²).

Nerita tessellata was the most abundant snail (10.8/0.25 m²) on reef rock at Galeta Navy Reef while *Nerita fulgurans*, *Littorina ziczac*, *L. angustior*, *L. meleagris* and *Nodilittorina* occurred in low density (≤ 1 snail/0.25 m²).

Table 3.1 Snail abundances by zone at Galeta Navy Reef 1982-1983. Mean \pm one standard deviation/0.25 m², N=20 quadrats, except in *Thalassia* and *Laurencia* where n=30.

Species	High Rubble	Low Rubble	Reef Rock	<i>Thalassia</i>	Sand	<i>Laurencia</i>
<i>Tectarius muricatus</i>	6.1 \pm 8.6	-(*)	-	-	-	-
<i>Nodilittorina tuberculata</i>	2.8 \pm 3.7	0.9 \pm 1.7	0.1 \pm 0.2	-	-	-
<i>Littorina angustior</i>	10.8 \pm 14.2	8.8 \pm 9.0	1.0 \pm 4.0	-	-	-
<i>L. lineolata</i>	3.6 \pm 5.2	3.3 \pm 4.4	-	-	-	-
<i>L. ziczac</i>	0.3 \pm 0.7	1.3 \pm 2.7	0.2 \pm 0.9	-	-	-
<i>Melampus coffeus</i>	0.3 \pm 1.1	-	-	-	-	-
<i>Nerita versicolor</i>	0.7 \pm 1.6	11.4 \pm 16.6	-	-	-	-
<i>N. fulgurans</i>	-	0.8 \pm 1.5	0.3 \pm 0.6	-	-	-
<i>N. peloronta</i>	-	0.3 \pm 0.7	-	-	-	-
<i>N. tessellata</i>	-	12.1 \pm 22.7	10.8 \pm 9.5	-	-	-
<i>Planaxis nucleus</i>	-	0.6 \pm 2.2	-	-	-	-
<i>L. meleagris</i>	-	-	1.0 \pm 2.8	-	-	-
Gastropod sp. 1	-	-	0.1 \pm 0.2	-	-	-
<i>Cerithium eburneum</i>	-	-	-	3.0 \pm 5.8	-	-
<i>C. literatum</i>	-	-	-	12.1 \pm 14.7	-	-
<i>Thais rustica</i>	-	-	-	0.03 \pm 0.2	-	-
<i>Batillaria minima</i>	-	-	-	-	96.4 \pm 97.3	-
<i>Neritina virginea</i>	-	-	-	-	0.3 \pm 0.9	-
Gastropod sp. 2	-	-	-	-	0.1 \pm 0.3	-
<i>Astrea ?phoebia</i>	-	-	-	-	-	0.2 \pm 0.5
<i>Cyprea zebra</i>	-	-	-	-	-	1.4 \pm 2.6
<i>Diodora dysoni</i>	-	-	-	-	-	0.1 \pm 0.2
<i>Leucozonia nassa</i>	-	-	-	-	-	0.03 \pm 0.2
<i>Smaragdia</i> sp.	-	-	-	-	-	0.2 \pm 0.4
<i>Thais deltoidea</i>	-	-	-	-	-	0.3 \pm 0.9
Nudibranch sp.	-	-	-	-	-	0.1 \pm 0.4
Gastropod sp. 6	-	-	-	-	-	0.03 \pm 0.2

(*) - = not present.

Two species, *Batillaria minima* and *Neritina virginea*, occurred on intertidal sand of the Galeta Navy Reef in 1982; *Batillaria* was patchily abundant ($\sim 100/0.25$ m²), while *Neritina* was rare ($<1/0.25$ m²). *Thalassia* beds were characterized by two species of cerithids, *Cerithium eburneum* and *C. literatum* at a total density of ~ 15 individuals/0.25 m². These snails were found on and around *Thalassia* blades.

Laurencia beds had relatively high gastropod diversity, but low abundances in 1982 (total density <3 individuals/0.25 m²). Both predaceous and herbivorous gastropods occurred within this habitat; the most common were *Cypraea zebra*, *Smaragdia* sp. and *Thais deltoidea*.

3.3.2 Extent of Oiling

Oil from the 1986 spill was not deposited uniformly across Galeta Navy Reef. The high rubble zone, which is wetted only by spray and splash, had a percent cover of $2.3 \pm 2.9\%$ oil in August 1986 (mean \pm one standard deviation, n=20 quadrats). In contrast, the low rubble and reef rock zones were heavily oiled. In the low rubble zone oil covered $69.0 \pm 17.6\%$ and diatoms growing on hardened oil averaged $15.5 \pm 14.0\%$ cover. On reef rock, hardened oil or oil overgrown by diatoms accounted for $91.7 \pm 9.6\%$ cover. Oil was present in the sand, *Thalassia* and *Laurencia* zones, but the amount could not be quantified as was done on the rocky substratum of the three zones higher in the intertidal. However, slicks and oil globules were seen at high tide in August 1986 (S. D. Garrity, pers. obs.), and oil seeped visibly from sand disturbed at low tide. In *Thalassia* beds, plants were heavily covered with epiphytes, especially the red alga *Acanthophora spicifera*, bluegreen algae and diatoms. The *Laurencia* bed was heavily silted and had high cover of turf-like microalgae and bare space ($17.5 \pm 17.9\%$).

Control areas showed no oil coverage attributable to the spill. No sign of oiling was found at María Soto through the three years of the study. However, at Toro Point, located adjacent to the Atlantic entrance to the Panama Canal, a few tar balls were noted initially and have been found with increasing frequency in the low rubble and reef rock zones at each visit (Figure 3.9). This low-level, chronic oiling appears related to ship traffic. Additionally, in May 1988, a 75-foot sailboat ran aground on the reef at Toro Point, releasing an undetermined amount of diesel fuel. An oily sheen was seen across the flat, and dead crabs and snails were found in several habitats (C. González, pers. obs.). Diesel fuel is more volatile than crude oil; this and the small size of the spill suggest that while it may have caused short-term effects, it was unlikely to have persistent effects relative to the Bahía las Minas spill of 1986.

Percent cover of oil was monitored again at both Galeta Navy Reef and Toro Point in November 1988. In the high rubble zone at Galeta Navy Reef, $2.6 \pm 4.8\%$ (mean \pm one standard deviation, n=20 quadrats) of the substratum was covered with oil, most of which was highly weathered mats of tar. This is approximately equal to the cover of oil in the high rubble zone immediately after the spill. More oil was recorded at Toro Point ($5.3 \pm 17.1\%$) because a large tar ball covered 76% of one quadrat. In the low rubble zone, oil cover had dropped to $3.6 \pm 3.7\%$ in November 1988, as compared with $\sim 75\%$ cover in August 1986. Only traces of oil were recorded at Toro Point. Oil only covered $14.4 \pm 21.6\%$ of the reef rock zone at Galeta Navy Reef (vs. $\sim 90\%$ in August 1986); this remaining oil was heavily



Figure 3.9 Tarball in the reef rock zone, Toro Point, May 1988. The forceps to the upper left of the tarball are 25.4 cm long.

overgrown by diatoms. Only traces of oil were recorded at Toro Point in an equivalent area. Because of the continuing presence of tar balls and the May 1988 diesel spill, Toro Point should be considered lightly oiled compared to Galeta Navy Reef (heavily oiled) or María Soto (unoiled).

3.3.3 Initial Effects on Gastropod Populations: Oiling and Mortality of Snails

Although the amount of oil deposited on the high rubble zone at Galeta Navy Reef was small (see above), a greater proportion of snails than of substratum was oiled in August 1986. Forty-eight percent of *Tectarius* counted in quadrats, 25% of *Nodilittorina*, 28% of *L. angustior*, 17% of *L. ziczac* and 6% of *L. lineolata* had oiled shells in August 1986. Dead individuals of ten species were collected ($n = 283$ snails) within the high rubble zone outside of density quadrats (Table 3.2 A). Included were *Nodilittorina*, *L. angustior*, *L. lineolata* and *L. ziczac*, normally resident in the high rubble zone; *N. versicolor*, *N. tessellata*, *N. peloronta*, and *Planaxis nucleus*, usually found lower on the shore; and *Melampus* and *L. angulifera*, primarily inhabitants of mangroves (*N. versicolor* and *Melampus* were both rare in the high rubble zone before the spill). Live *L. lineolata* did not differ in size from empty shells (Table 3.3, $p > 0.1$, G test with Williams' correction, Sokal and Rohlf 1981). For all other dead snails collected, too few live individuals were present for comparison.

Dead snails of ten species were also collected from the high rubble in November 1986, about seven months after the spill ($n = 262$ snails); nine species appeared in density quadrats (Table 3.2 A). Because all empty shells had been removed when found in August, these data represent mortality of snails between the August and November monitorings. They also suggest the higher proportion of oiled snails relative to oiled substratum in the high rubble zone (observed in August, above) was due to upward movement of oiled individuals following the spill. The absence of live individuals of species found dead from this zone supports this hypothesis (Table 3.2 vs. Table 3.4), but prevents a test for size-selective mortality.

Of the snails found in low rubble in August 1986, 100% of *P. nucleus*, *L. angustior*, and *L. lineolata* had oiled shells, as did 68% of *N. versicolor* and 14% of *N. tessellata*. The single *L. ziczac* found was unoiled. Much of the low rubble zone was coated with a thick, viscous layer of oil (see above). We counted the number of dead snails stuck to or covered with oil (Table 3.2 B) along with counts of live individuals, then removed the dead snails. At least five species were found (there were individuals of both *Littorina* and *Nerita* spp. too oiled to identify). Live and dead individuals of *N. versicolor* and *N. tessellata* were abundant enough to test for size-selective mortality; in both cases, small snails died more than large ones ($p < 0.005$, G test with Williams' correction, Table 3.3).

Table 3.2 Abundances of dead snails in high and low rubble zones at Galeta Navy Reef. Data are the mean (one standard deviation)/0.25 m². N=20 quadrats, + =present, but not found in a quadrat, - =absent. *Nerita* spp. and *Littorina* spp. are individuals that were too oiled to identify to species.

Species	August 1986	November 1986
A. High Rubble Zone		
<i>Tectarius muricatus</i>	-	+
<i>Nodilittorina tuberculata</i>	+	0.1 (0.31)
<i>Littorina angustior</i>	+	0.3 (0.55)
<i>L. ziczac</i>	+	0.2 (0.67)
<i>L. lineolata</i>	+	0.1 (0.22)
<i>L. angulifera</i>	+	0.1 (0.22)
<i>Nerita tessellata</i>	+	1.0 (1.50)
<i>N. versicolor</i>	+	0.3 (0.73)
<i>N. peloronta</i>	+	-
<i>Melampus coffeus</i>	+	0.9 (1.50)
<i>Planaxis nucleus</i>	+	0.3 (0.57)
B. Low Rubble Zone		
<i>Nerita fulgurans</i>	0.6 (1.15)	-
<i>N. versicolor</i>	0.9 (1.40)	+
<i>N. tessellata</i>	0.3 (0.92)	+
<i>Nerita</i> spp.	1.6 (1.88)	-
<i>Littorina ziczac</i>	0.1 (0.22)	-
<i>L. angustior</i>	-	+
<i>Littorina</i> spp.	0.8 (1.27)	-
<i>Planaxis nucleus</i>	0.4 (0.67)	+

Individuals of four species were found dead in November 1986 (Table 3.2 B). For *N. versicolor* and *P. nucleus*, small snails died more frequently than large ones (Table 3.3, $p < 0.005$, G test with Williams' correction). The opposite was true of *N. tessellata* ($p < 0.005$, G test with Williams' correction), but only 12 live *N. tessellata* were found. Seven of these were less than 8 mm long while the smallest live snail found in August was 9 mm long.

Table 3.3 Size selective mortality: high and low rubble zones at Galeta Navy Reef. Data are the number of snails in millimeter size classes found living or dead at Galeta after the oil spill.

		Size in millimeters:																	Probability *
		4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	
High Rubble Zone																			
<i>Littorina lineolata</i>																			
8-86	Live					2	4	2	4	2	1								> 0.1
	Dead					16	17	17	23	48	7								
Low Rubble Zone																			
<i>Nerita versicolor</i>																			
8-86	Live					1	2	2	1	10	10	8	22	25	26	12	12	4	<< 0.005
	Dead	2	1			9	5	14	19	9	22	28	25	9	7	1	1		
11-86	Live					1	4	3	23	24	33	37	33	17	10	2	1	1	<< 0.005
	Dead					3	3	3	2	5	4	7	5	6	1	2	1		
<i>N. tessellata</i>																			
8-86	Live							2	7	14	27	34	22	22	15	5	1	<< 0.005	
	Dead	1	4	9	11	19	21	24	42	54	36	37	16	6	1				
11-86	Live	2	2	2	1						1	2	1	1				<< 0.005	
	Dead			2	4	2	6	3	7	10	17	19	9	5	1				
<i>Planaxis nucleus</i>																			
11-86	Live							1	2	5	10	21	7	4				<< 0.005	
	Dead					2	1	2	4	9	6								

* Calculated using the G test with Williams' correction, Sokal and Rohlf 1981.

3.3.4 Initial Effects on Gastropod Populations: Effects of Reef Cleanup Crew, August-September 1986

Gastropod abundances at Galeta Navy Reef were monitored twice in August-September 1986: before and immediately after the activities of a cleaning crew (Table 3.4). Cleaning occurred in the high and low rubble zones, where oiled rocks, rubble and other debris were collected by hand and removed by wheelbarrow, and in the sand zone, where cleaning was attempted using salt-water pumps and hoses. Here, oil, snails and small detritus were washed seaward several meters into deeper water (pers. obs.).

Table 3.4 Density of gastropods before and after cleaning in the high rubble, low rubble and sand zones at Galeta Navy Reef, 1986. Data are the mean number (one standard deviation)/0.25 m². N = 20 quadrats at each monitoring in 1986.

	August Post Spill	August-September Post Cleaning	November
A. High Rubble Zone			
<i>Littorina angulifera</i>	0.1 (0.2)	0	0
<i>L. angustior</i>	14.8 (12.8)	13.0 (18.9)	10.4 (13.5)
<i>L. lineolata</i>	0.9 (1.4)	0	0.4 (0.8)
<i>L. ziczac</i>	0.6 (1.6)	0	0.8 (0.3)
<i>Nodilittorina tuberculata</i>	2.5 (3.5)	1.4 (2.7)	1.1 (1.2)
<i>Tectarius muricatus</i>	14.8 (10.2)	7.2 (13.3)	12.1 (11.3)
<i>Nerita versicolor</i>	0.1 (0.2)	0.7 (2.7)	0
<i>L. nebulosa</i>	0	0.1 (0.2)	0
<i>N. peloronta</i>	0	0.1 (0.3)	0
<i>N. tessellata</i>	0	0	0.6 (2.5)
<i>Melampus coffeus</i>	0	0	0.1 (0.5)
B. Low Rubble Zone			
<i>Littorina angustior</i>	0.2 (0.7)	1.4 (5.8)	1.0 (1.3)
<i>L. ziczac</i>	0.1 (0.2)	0	0.5 (1.1)
<i>Littorina</i> spp. (*)	0.1 (0.2)	0	0
<i>Nerita tessellata</i>	1.1 (2.6)	0.1 (0.2)	0.1 (0.3)
<i>N. versicolor</i>	1.3 (2.2)	0.3 (0.9)	4.3 (4.0)
<i>Planaxis nucleus</i>	0.1 (0.2)	0	1.2 (2.2)
<i>P. lineatus</i>	0	0.1 (0.5)	0
<i>Tectarius muricatus</i>	0	0.1 (0.5)	0
<i>Melampus coffeus</i>	0	0	0.1 (0.5)
C. Sand Zone			
<i>Batillaria minima</i>	11.1 (41.2)	0.1 (0.2)	0
<i>Cerithium eberneum</i>	24.3 (69.3)	0	0
<i>Neritina virginea</i>	0	0	415.0 (497.4)

(*) too oiled to identify to species

In the high rubble zone, six of seven species found before cleanup were less abundant immediately afterwards, with the abundance of *Tectarius* dropping 50%. One rare species increased slightly in abundance, and two new species appeared. In the low rubble zone, four of five species decreased in abundance or disappeared, one increased, and two new species were found after cleanup. In sand, the most abundant snail disappeared, and another common species decreased in density by a factor of ten following cleanup.

Changes in the high and low rubble zones appeared ephemeral. Table 3.4 also shows snail abundances for the following quarterly monitoring (November 1986). Four species which had appeared in the two rubble zones after cleanup did not persist. However, reductions in density of *Batillaria* and *C. eberneum* in the sand zone continued. These two species disappeared, and a third species, *Neritina virginea*, not previously found, appeared in high density.

3.3.5 Long-term Effects on Gastropod Abundances

High Rubble Zone

Overall Abundance - After the spill, total snail abundance (all species combined) remained similar (~ 30 snails/ 0.25 m^2) between Toro Point and Galeta Navy Reef in August and November 1986 (Figure 3.10). From February 1987 through May 1989, however, density was greater at Toro Point than Galeta Navy Reef in all ten monitorings. When we compared the number of species found in each monitoring's transect, more species occurred at Toro Point in eight of 12 instances, with one tie. More species were found at Galeta Navy Reef in the two first monitorings after the spill (when two "nonresident" species, *L. angulifera* and *Melampus* were found) and in May 1988 (after a diesel spill at Toro Point).

Individual Species Abundances - Figure 3.11 shows the relative contribution of common species to overall snail abundance at each site. Between November 1986 and February 1987, a single species, *Littorina lineolata*, increased markedly in abundance at Toro Point only. This caused most of the difference in abundance between sites. Another contributor, *Littorina angustior*, was initially abundant only at Galeta Navy Reef, then disappeared during the same period. Two species, *Nodilittorina tuberculata* and *Tectarius muricatus*, showed roughly similar changes in abundance at both sites.

Figures 3.12 and 3.13 show long-term abundance data for individual species in this zone. Despite some gastropod mortality immediately after the spill (Chapter 3.3.3), abundances of most species at Galeta Navy Reef in August 1986 were roughly similar to those in 1982. *Tectarius* was the exception, having more than doubled in density (to $\sim 15/0.25 \text{ m}^2$). The unoiled site at Toro Point, first monitored in August 1986, had three main differences from Galeta Navy Reef: *L. angustior* was

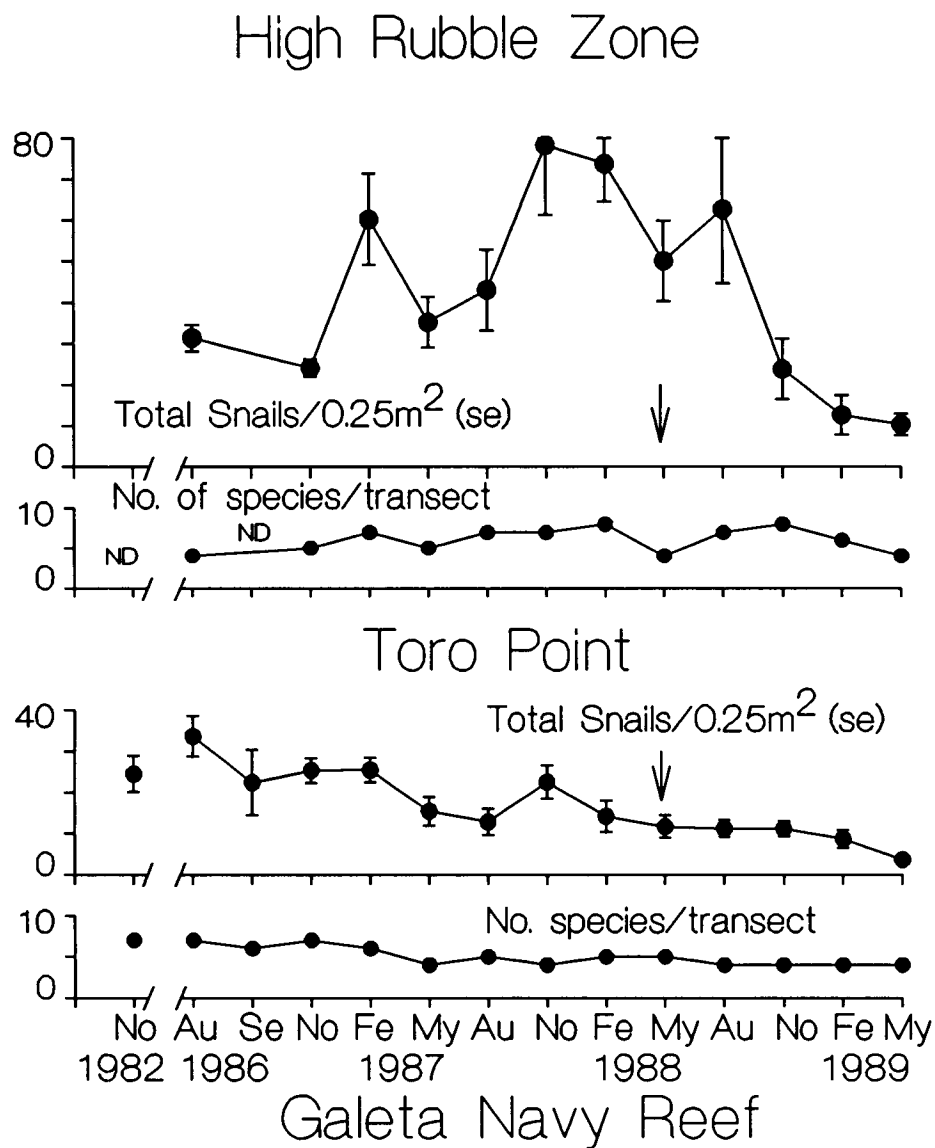


Figure 3.10 Total gastropod abundance and number of species in transect sampling, high rubble zone. Abundance data are mean densities (standard error) of all gastropod species combined, for each site at each sampling date. Vertical arrow indicates monitoring done just after a diesel spill at Toro Point in May 1988. Number of species is the total number of species recorded in 20 0.25 m² quadrats at each sampling date. See text for sampling methods. ND = No data.

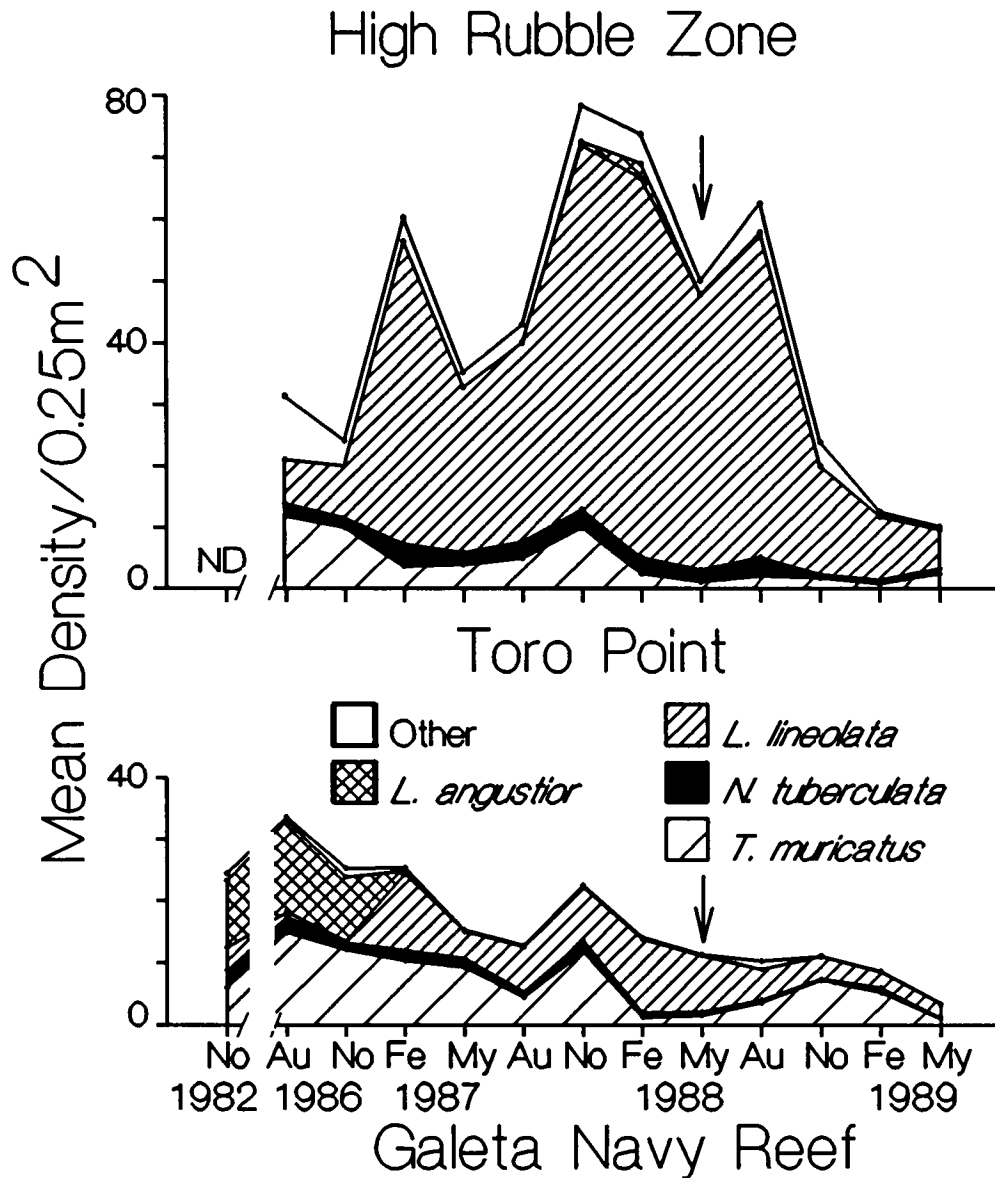


Figure 3.11 Total gastropod abundance in the high rubble zone by component species or taxonomic groups. Differentially shaded areas in graph illustrate the relative contributions of individual species or groups to overall mean abundance of gastropods at each monitoring. Vertical arrow indicates monitoring done just after diesel spill at Toro Point in May 1988. ND = No data.

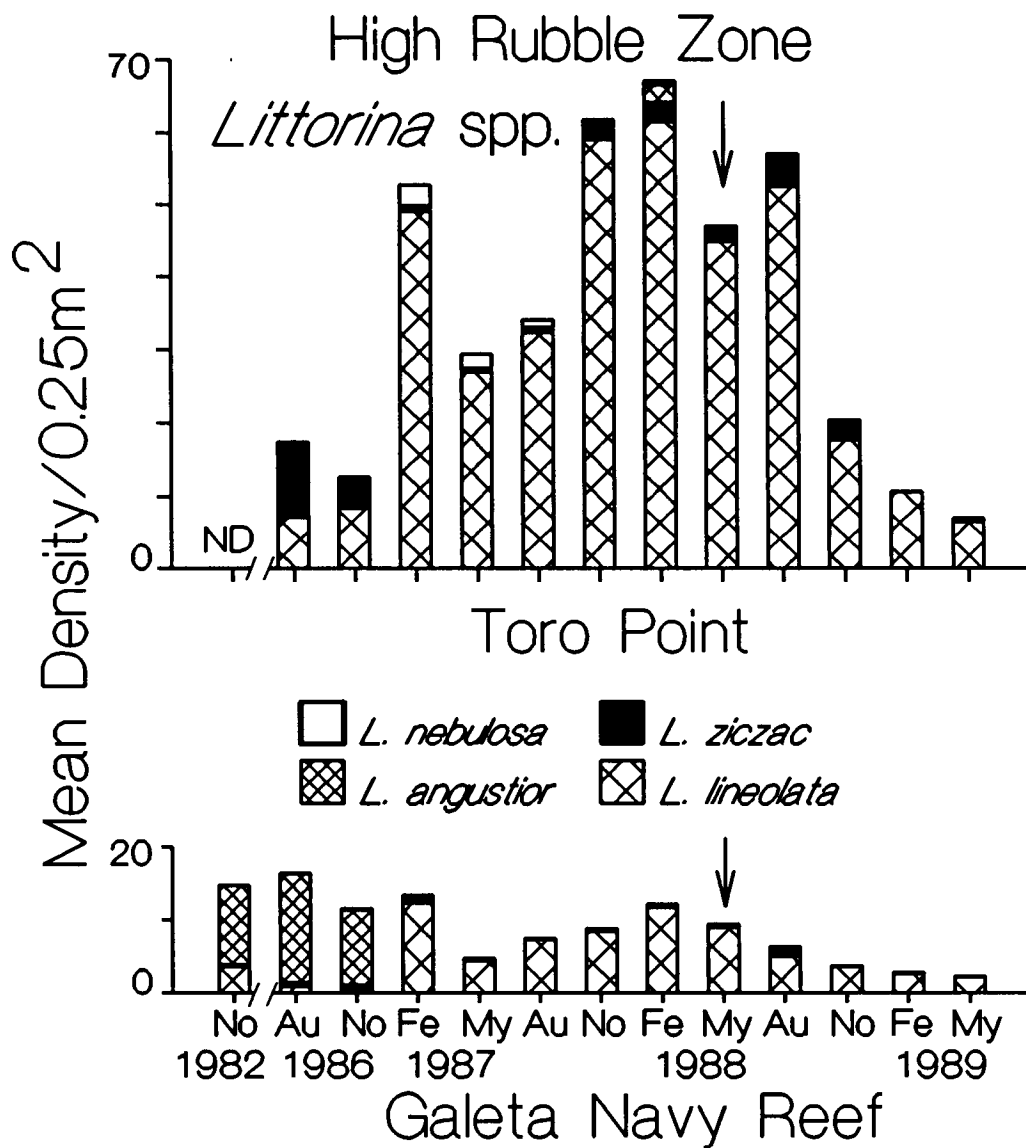


Figure 3.12 Abundance of *Littorina* species in the high rubble zone. Differentially shaded areas of each bar are mean density of each species of littorinid snail found at each monitoring period. Vertical arrow indicates monitoring done just after diesel spill at Toro Point in May 1988. See text for details. ND = No data.

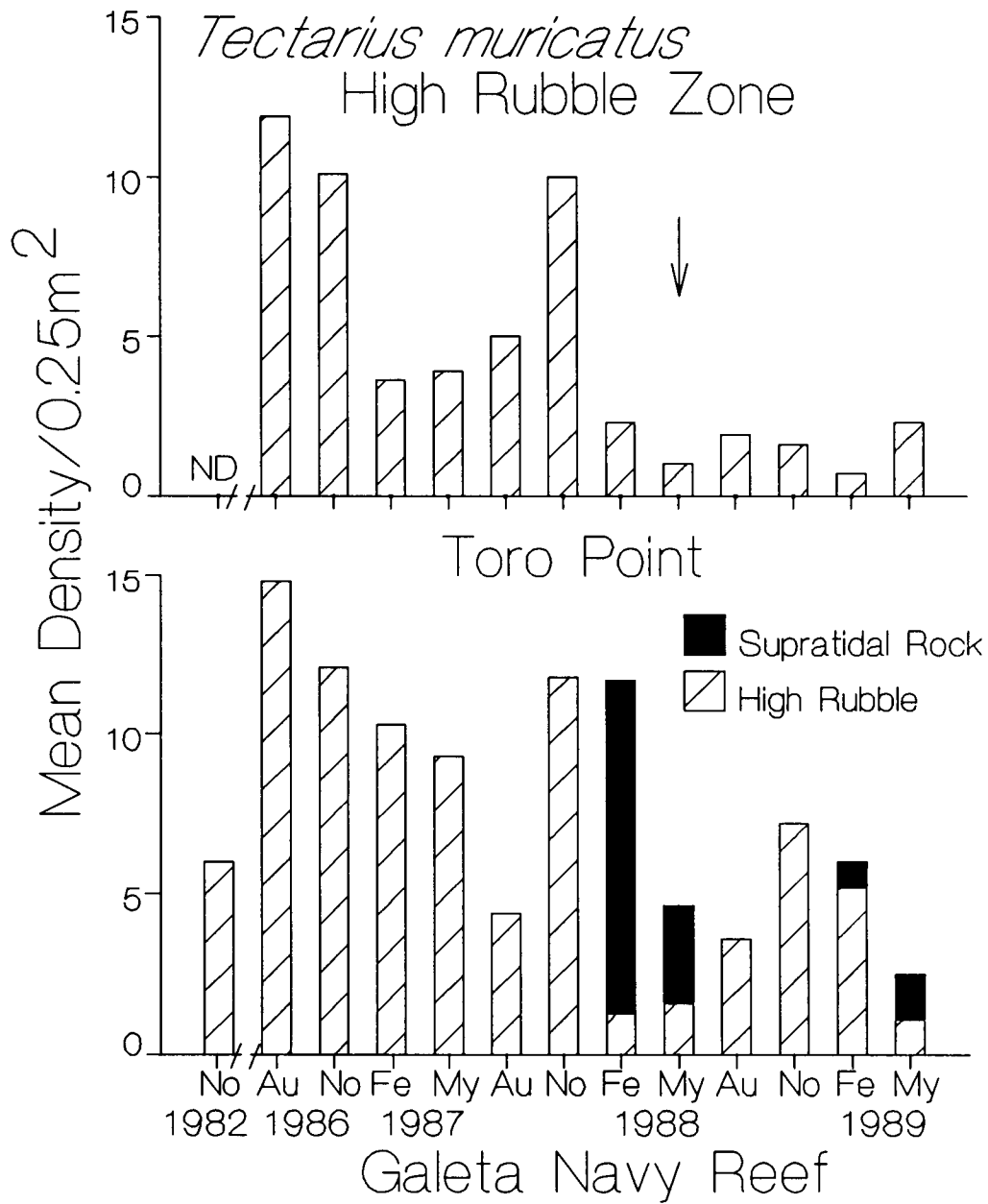


Figure 3.13 Abundance of *Tectarius muricatus* in the high rubble zone. Data are means of 20 quadrats at each sampling date. Fully shaded portions of bars for Galeta are means of 20 quadrats run along a transect line above the high rubble zone when snails were observed in the supratidal. Vertical arrow indicates monitoring done just after diesel spill at Toro Point in May 1988. ND = No data.

absent, *L. ziczac* was far more common than at Galeta Navy Reef, and only four species were recorded.

Density at both oiled and unoiled sites remained about the same in November 1986, except for a decrease in *L. ziczac* at Toro Point and an increase in *Tectarius* at Galeta Navy Reef. Both oiled and unoiled sites had markedly increased numbers of *L. lineolata* in February 1987. *L. angustior*, abundant in all previous monitorings at the oiled site, essentially disappeared in February 1987. *L. ziczac* became rare at the unoiled site at the same time, and *Tectarius* also dropped sharply in abundance. *L. ziczac* remained rare at both sites (<1 snail/ 0.25 m^2) through the next several monitorings (rainy season), then increased at Toro Point during late rainy/early dry season 1987-88 ($2-4/0.25\text{ m}^2$). *L. lineolata*, after a decrease in May 1987, increased at both sites through dry season 1988, then decreased again in May 1988.

The 1988 dry season was windy, with extremely high wave action (R. C. Thompson, pers. comm., pers. obs.). By May, up to 75% of the substratum in the high rubble zone at Galeta Navy Reef had been recently eroded (mean = 24.8%, range 0-75%, $n=20$). There was somewhat less erosion at Toro Point (mean = 10.5%, range 0-50, $n=20$). In February 1988, *Tectarius* decreased greatly in abundance at both Galeta Navy Reef and Toro Point. At Galeta Navy Reef, this reduction in numbers was partly caused by an upward migration of snails into supratidal debris (mean density \pm one standard deviation of *Tectarius* above its normal range = $10.5 \pm 8.1/0.25\text{ m}^2$, $n=20$ quadrats, Figure 3.13). In May 1988, *Tectarius* was rare at Galeta Navy Reef in both the high rubble zone and above its normal range ($2.9 \pm 3.3/0.25\text{ m}^2$). This suggests delayed mortality. Similar reductions in density were observed at Toro Point, where less supratidal rock was available. The absence of a supratidal refuge equivalent to that at Galeta Navy Reef apparently led to greater immediate *Tectarius* mortality during storm conditions, and probably caused the reduction in numbers of *Tectarius* in February 1987 as well.

In May 1988, density for all species dropped at both Galeta Navy Reef and Toro Point (Figure 3.10, 3.11); this also occurred in February to May 1987. It may reflect a regular migration of snails to lower levels of the shore at the end of the dry season, when water levels predictably drop and the reef flat is exposed for long periods (Cubit *et al.* 1986). However, at Toro Point, there was a small spill of diesel fuel just prior to the May monitoring. Species number dropped from eight to four (the four rarest species disappeared). *N. versicolor* was one; it had been present in four of seven previous monitorings. This species died in large numbers in the lower intertidal after the diesel spill and may have been affected indirectly in the high rubble zone. Members of the genus *Nerita* typically exhibit rhythmic feeding migrations (e.g., Levings and Garrity 1983; Peckol *et al.* 1989), making them likely to be affected by conditions in a broad range of the intertidal. Overall, data suggest that the diesel spill at Toro Point in May 1988 had effects in the high rubble zone

only on *N. versicolor* and perhaps on three rare species of *Littorina*. Density returned to previous levels in August 1988.

Over the next four monitorings (August 1988 - May 1989), *L. lineolata* slowly continued to decline at Galeta Navy Reef; at Toro Point, after a small increase in August 1988, it also declined through the remainder of the study. Rare species, including *L. ziczac* and *L. nebulosa*, also became rarer or disappeared at both sites. This general trend was correlated with further physical degradation of the habitat at both sites through continued erosion by wave action (pers. obs.).

Low Rubble Zone

Overall Abundance - Total gastropod abundance in the low rubble zone was generally greater, but more variable at Toro Point than at Galeta Navy Reef (Figure 3.14). For three years following the spill, snail density was higher at Toro Point in 10 of 12 monitorings. In two of the three years, snail density was highest in mid-rainy season (August) at the unoiled site, while there was little seasonality evident at Galeta Navy Reef (Figure 3.14). At Galeta Navy Reef, overall snail density remained 50-95% lower than it had been in 1982-3 until August 1988 (over nine quarterly monitorings).

The number of species found per transect was higher at Galeta Navy Reef than Toro Point in seven of 12 monitorings; the unoiled site had more or the same number of snail species only for the first four monitorings following the spill and in the final (May 1989) monitoring (Figure 3.14).

Individual Species Abundances - The relative contributions of various species or groups to overall snail abundance are shown in Figure 3.15. Littorinids and *Planaxis* showed strongest differences between oiled and unoiled sites, while neritids did not differ as strongly. At Galeta Navy Reef only, neritids appeared to have a seasonal cycle of abundance, with lowest densities in May (early rainy season) in all three years examined. At Toro Point, littorinid abundance decreased greatly through rainy season 1987 and never recovered. *Planaxis*, the major contributor to overall snail density, also showed highly seasonal patterns of abundance, with peaks in mid to late rainy seasons 1987 and 1988 at Toro Point (and to a lesser degree at Galeta Navy Reef).

Figures 3.16 and 3.17 show individual species' densities for littorinid and neritid snails. In the first monitoring after the spill, only five species were found at Galeta Navy Reef (Figure 3.14, August 1986). *Nerita fulgurans*, *N. peloronta*, *L. lineolata* and *Nodilittorina* disappeared there between 1982 and August 1986; abundances of remaining species dropped roughly by a factor of ten (Figures 3.16, 3.17). Immediately following activities of the clean-up crew, *L. lineolata*, *L. ziczac* and *P. nucleus* disappeared while a few *P. lineatus* appeared (Table 3.4).

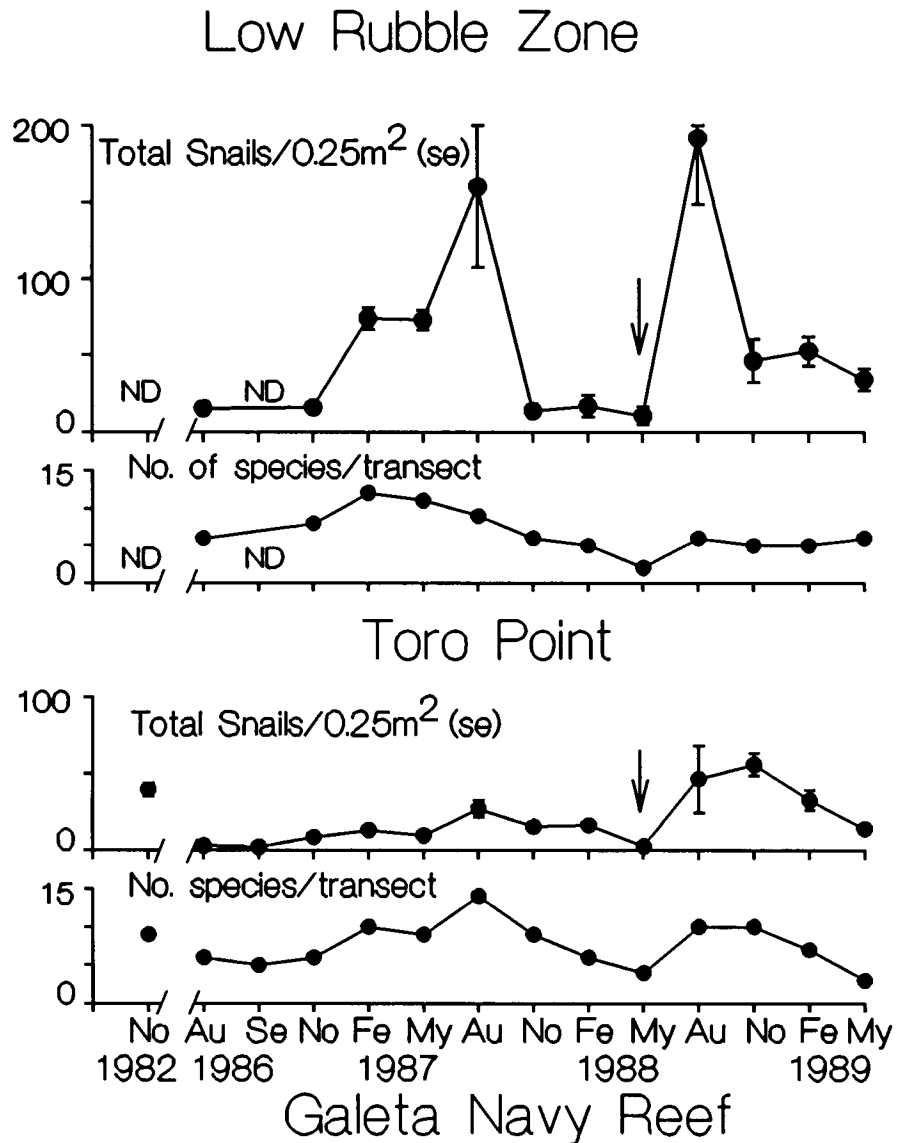


Figure 3.14 Total gastropod abundance and number of species in transect sampling, low rubble zone. Abundance data are mean densities (standard error) of all gastropod species combined, for each site at each sampling date. Vertical arrow indicates monitoring done just after a diesel spill at Toro Point in May 1988. Number of species is the total number of species recorded in 20 0.25 m² quadrats at each sampling date. See text for sampling methods. ND = No data.

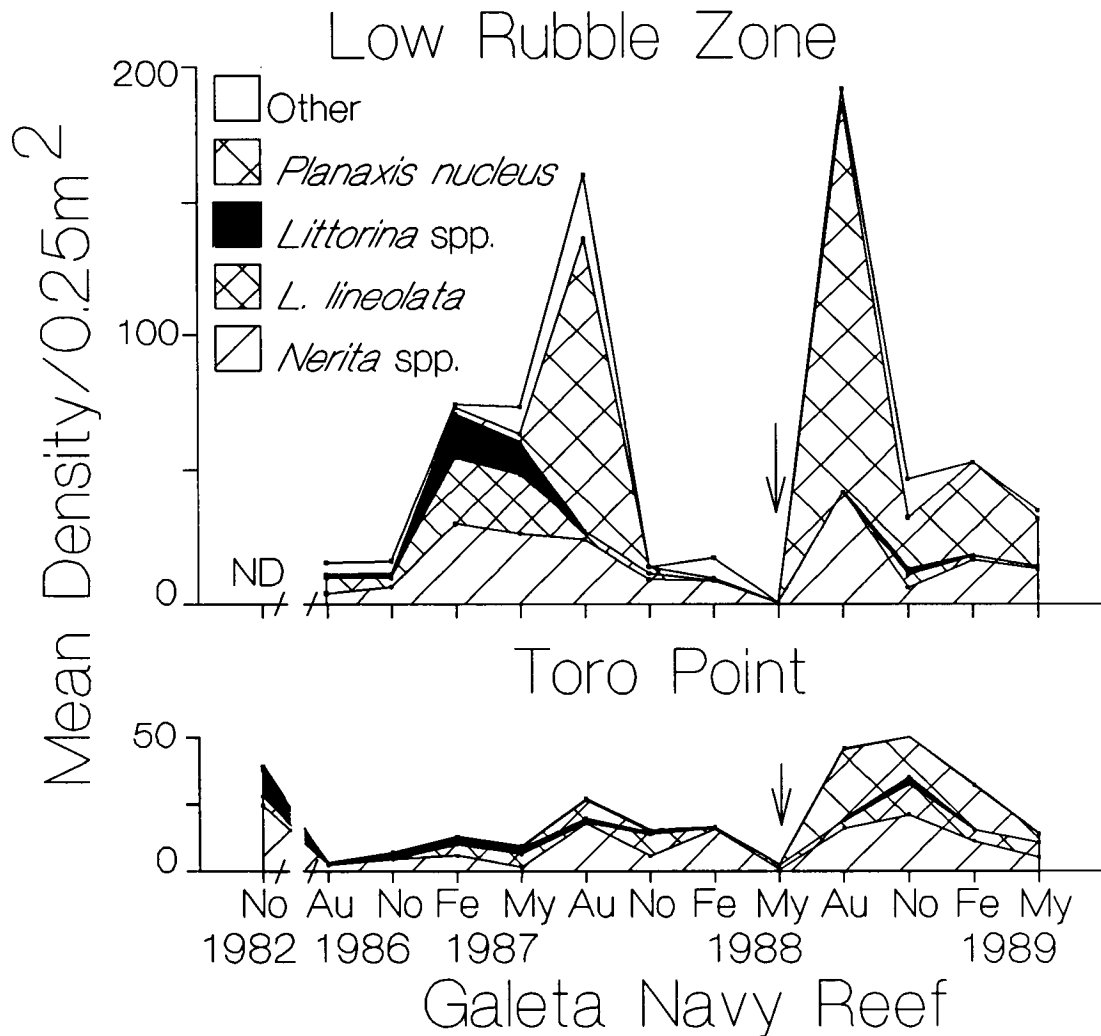


Figure 3.15 Total gastropod abundance in the low rubble zone by component species or taxonomic groups. Differentially shaded areas in graph illustrate the relative contributions of individual species or groups to overall mean abundance of gastropods at each monitoring. Vertical arrow indicates monitoring done just after diesel spill at Toro Point in May 1988. ND = No data.

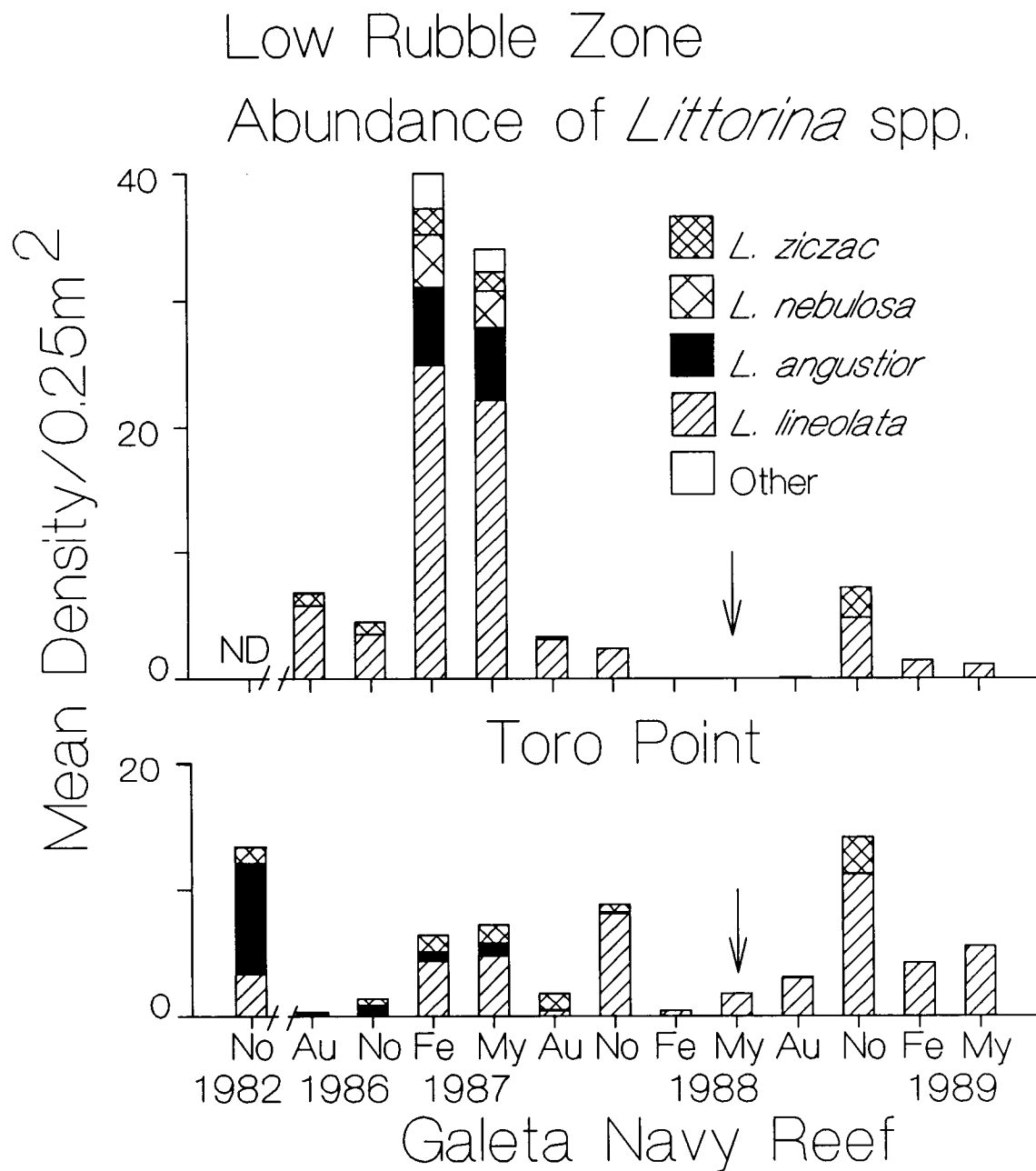


Figure 3.16 Abundance of species of *Littorina* in the low rubble zone. Differentially shaded areas of each bar are mean density of each species of littorinid snail found at each monitoring period. Vertical arrow indicates monitoring done just after diesel spill at Toro Point in May 1988. See text for details. ND = No data.

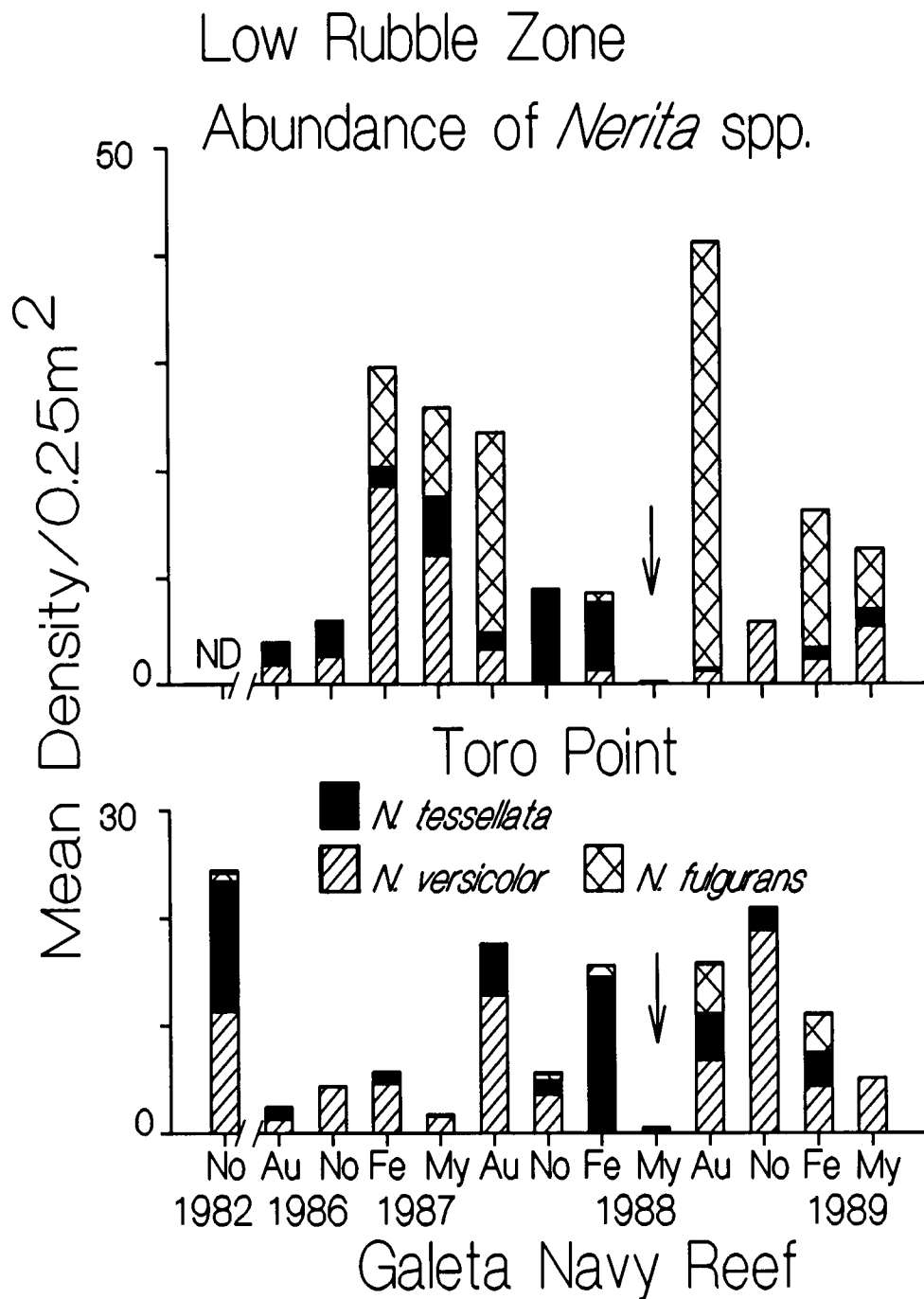


Figure 3.17 Abundance of species of *Nerita* in the low rubble zone. Differentially shaded areas of each bar are mean density of each species of neritid snail found at each monitoring period. Vertical arrow indicates monitoring done just after diesel spill at Toro Point in May 1988. See text for details. ND = No data.

In November 1986, four species present in 1982 were absent, and one new species appeared (Tables 3.1 and 3.4). The latter, *Melampus coffeus*, usually is found in mangrove detritus. Of the two previously most abundant species, *N. tessellata* dropped further in density between August and November 1986, while *N. versicolor* increased. There were no major changes in density for any species of gastropod at the unoiled site (Toro Point) during the same period (Figures 3.16, 3.17).

In February 1987 there were increases in density for ten species (most notably *L. lineolata* and *N. versicolor*) at Toro Point (Figures 3.16, 3.17). Three of these species also increased at Galeta Navy Reef (*N. tessellata*, *L. nebulosa*, *Nodilittorina*), but abundances remained comparatively low (Figures 3.16, 3.17). In contrast to events in the high rubble zone, most species in the low rubble zone did not decrease markedly in density in May 1987 (early rainy season) - only *N. tessellata* and several rare species (combined as "other" on Figure 3.16) became less abundant at Galeta Navy Reef. At Toro Point, most species declined slightly, and *N. tessellata* increased (Figures 3.16 3.17).

In August 1987, *P. nucleus* (Figure 3.15) and *N. fulgurans* (Figure 3.17) markedly increased in abundance at the unoiled site, and *N. versicolor*, *N. tessellata* and *P. nucleus* increased at Galeta Navy Reef (Figures 3.15, 3.17). Most other species declined in abundance at both the oiled and unoiled site, notably *L. lineolata* and *L. angustior* (Figure 3.16), or at Toro Point only (*N. versicolor* and *N. tessellata*, Figure 3.17).

In November 1987, snail density at Toro Point dropped markedly. The two species whose densities had increased greatly in August were relatively rare in November. Only *N. tessellata* increased in density, and three previously rare species were not found. At Galeta Navy Reef, the three species that had increased in abundance in August, as well as *L. ziczac*, markedly decreased. Three species that had been rare in August were not found in November samples, but *L. lineolata* increased by a factor of 20 (Figures 3.15, 3.16, 3.17).

Densities of snails in February 1988 were generally lower than in November 1987 at both sites. A notable exception was *Nerita tessellata* at Galeta Navy Reef; it increased by a factor of ten (Figure 3.17). The number of species decreased at both sites as well (from nine to six at Galeta Navy Reef, and from six to five at Toro Point). This contrasted sharply to events seen the previous February. Field notes document storm damage and erosion of the habitat between November 1987 and February 1988.

For most species, density at both Galeta Navy Reef and Toro Point dropped considerably between February and May 1988 (Figures 3.15, 3.16, 3.17); only *L. lineolata* at Toro Point showed a small increase. The overall decreased densities, perhaps due to continuing effects of rough dry season weather, complicate interpretation of the effects of the diesel spill that occurred only at Toro Point. However, there is some evidence for mortality due to the spill. Only two species were found at Toro Point in May, both at greatly reduced density; five species were recorded in February. All *Batillaria minima* in density quadrats were dead (mean

density of dead snails \pm one standard deviation = 10.3 ± 25.1 , $n=20$). At Galeta Navy Reef, total snail density also dropped, largely as a result of changes in numbers of *N. tessellata*. Three rare species disappeared (of six present at the previous monitoring), while one was added (*Nodilittorina*).

At the next monitoring (August 1988), three neritid species, particularly *N. fulgurans*, and *P. nucleus* had increased in abundance both at Toro Point and at Galeta Navy Reef (Figure 3.17). Littorinid abundances remained low, although *L. lineolata* continued to increase slowly at Galeta Navy Reef only (Figure 3.16). In November 1988, *N. fulgurans* disappeared from both sites, and *N. versicolor* increased (Figure 3.17). *L. lineolata* continued to increase at Galeta Navy Reef and reappeared, along with *L. ziczac* (both sites) at Toro Point (Figure 3.16).

After November 1988, littorinid abundance declined overall, with disappearances of *L. ziczac* at both sites, and a decrease in *L. lineolata* as well (with a slight increase in the latter at Galeta Navy Reef in May 1989 - Figure 3.16). In February 1989, *N. versicolor* declined in abundance at both sites, while *N. tessellata* and *N. fulgurans* reappeared at both sites. Neither species occurred at Galeta Navy Reef in May 1989; *N. versicolor* was the only neritid found. At Toro Point, *N. versicolor* and *N. tessellata* were more abundant than in February, but *N. fulgurans* was rarer.

Reef Rock Zone

Overall Gastropod Abundance - The effects of oiling were severe in the reef rock zone. Total gastropod abundance was higher at Toro Point than at Galeta Navy Reef in 11 of 12 monitorings; only in May 1988, following the localized diesel spill, was snail abundance as low at Toro Point as at Galeta Navy Reef (Figure 3.18). Similarly, the number of gastropod species found per transect was higher at Toro Point at all monitorings except May 1989. Like overall snail abundance, species number at Toro Point was lowest immediately after the May 1988 diesel spill, although there were still more species of molluscs found (three) than at Galeta Navy Reef (one).

The relative contribution of species or groups to overall patterns of abundance in the reef rock zone is shown in Figure 3.19. Both littorinids and neritids, present before the spill, were almost entirely absent from Galeta Navy Reef's rock zone for two years following the spill. Even after three years, density at Galeta Navy Reef was <7.5 snails/ 0.25 m^2 (all species combined). At Toro Point, there were 4-10 neritids/ 0.25 m^2 , except after the diesel spill (May 1988), when they were rare (0.2 snails/ 0.25 m^2). At Toro Point, littorinids were both more abundant and more variable than neritids, with no apparent pattern of seasonality and lowest density immediately after the diesel spill. Several rare species, including *Purpura patula* and

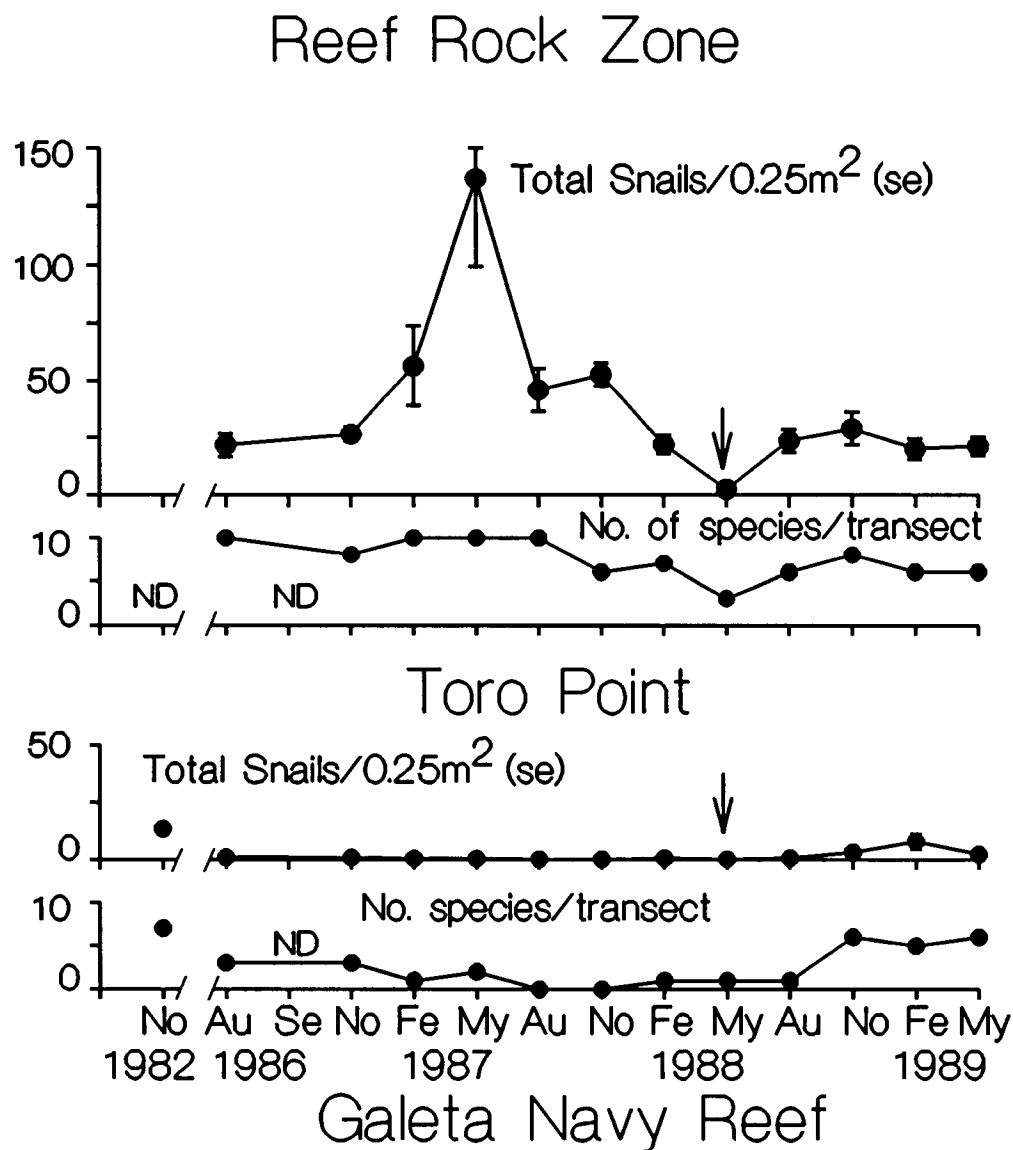


Figure 3.18 Total gastropod abundance and number of species in transect sampling, reef rock zone. Abundance data are mean densities (standard error) of all gastropod species combined, for each site at each sampling date. Vertical arrow indicates monitoring done just after a diesel spill at Toro Point in May 1988. Number of species is the total number of species recorded in 20 0.25 m² quadrats at each sampling date. See text for sampling methods. ND = No data.

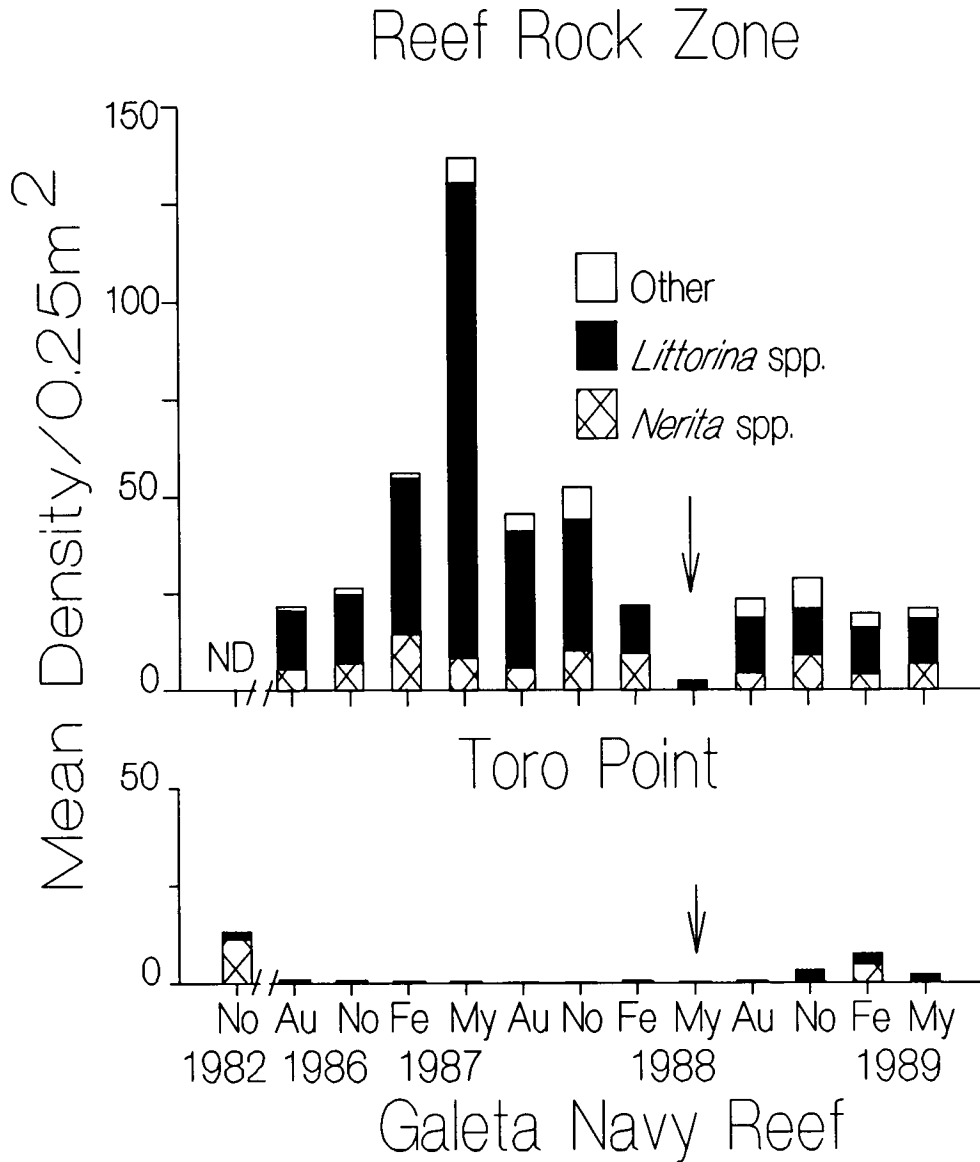


Figure 3.19 Total gastropod abundance in the reef rock zone by component taxonomic groups. Differentially shaded areas of bars illustrate the relative contributions of individual species or groups to overall mean abundance of gastropods at each monitoring. Vertical arrow indicates monitoring done just after diesel spill at Toro Point in May 1988. ND = No data.

Acanthopleura granulata, were found at Toro Point in nine of 12 monitorings, but only one individual was found at Galeta Navy Reef.

Individual Species Abundances - At Galeta Navy Reef in August 1986, of the species found prior to the spill only two species of neritids remained, and both were less abundant than in 1982 (Figure 3.20; the mean density of *N. fulgurans* in August 1986 was 0.1/0.25 m² and cannot be seen in the figure). *N. fulgurans* was not found between August 1986 and August 1988. *N. versicolor*, not found in 1982, was present in August 1986 (mean density = 0.1/0.25 m², not visible in Figure 3.20), but was not recorded from May 1987 through February 1989. *N. tessellata* remained rare, never reaching a density of one snail/0.25 m². No littorinids reappeared through August 1988. A single chiton, *Acanthopleura granulata*, was found in November 1986. Recovery appeared to begin in earnest only in 1989 and was not complete in May when the study terminated.

At the unoiled site, the reef rock zone had more vertical relief and was more emergent than at Galeta Navy Reef. Three or four species of neritids were counted during all monitorings except May 1988, when only one species was found.

Four species of littorinids occurred in the reef rock zone (Figure 3.21). Numbers of *L. lineolata* fluctuated, but remained high through February 1988, dropping in May 1988. *L. ziczac* remained rare until August 1988. *L. angustior* varied from 5 to 28 snails/0.25 m² between August 1986 and August 1987; none was recorded after August 1987. *L. meleagris* was ephemeral; it was found only in 1987.

Seven species were recorded at Toro Point in the reef rock zone in February 1988, at a total snail density of 21.8/0.25 m². In May, only three species were found, with a total snail density of 2.4/0.25 m² (an order of magnitude fewer snails than had been recorded in seven previous monitorings). On May 23, more than 120 dead *N. versicolor* and numerous dead crabs were counted in a 5 m² area. Dead individuals of five species (*N. tessellata*, *N. versicolor*, *N. peloronta*, *N. fulgurans* and *Tectarius*, n=81 total, most *N. tessellata*) were found on June 1. These data strongly suggest an immediate impact of diesel at Toro Point in this zone. Density recovered in August 1988 and remained similar to that in February 1988 for the duration of the study.

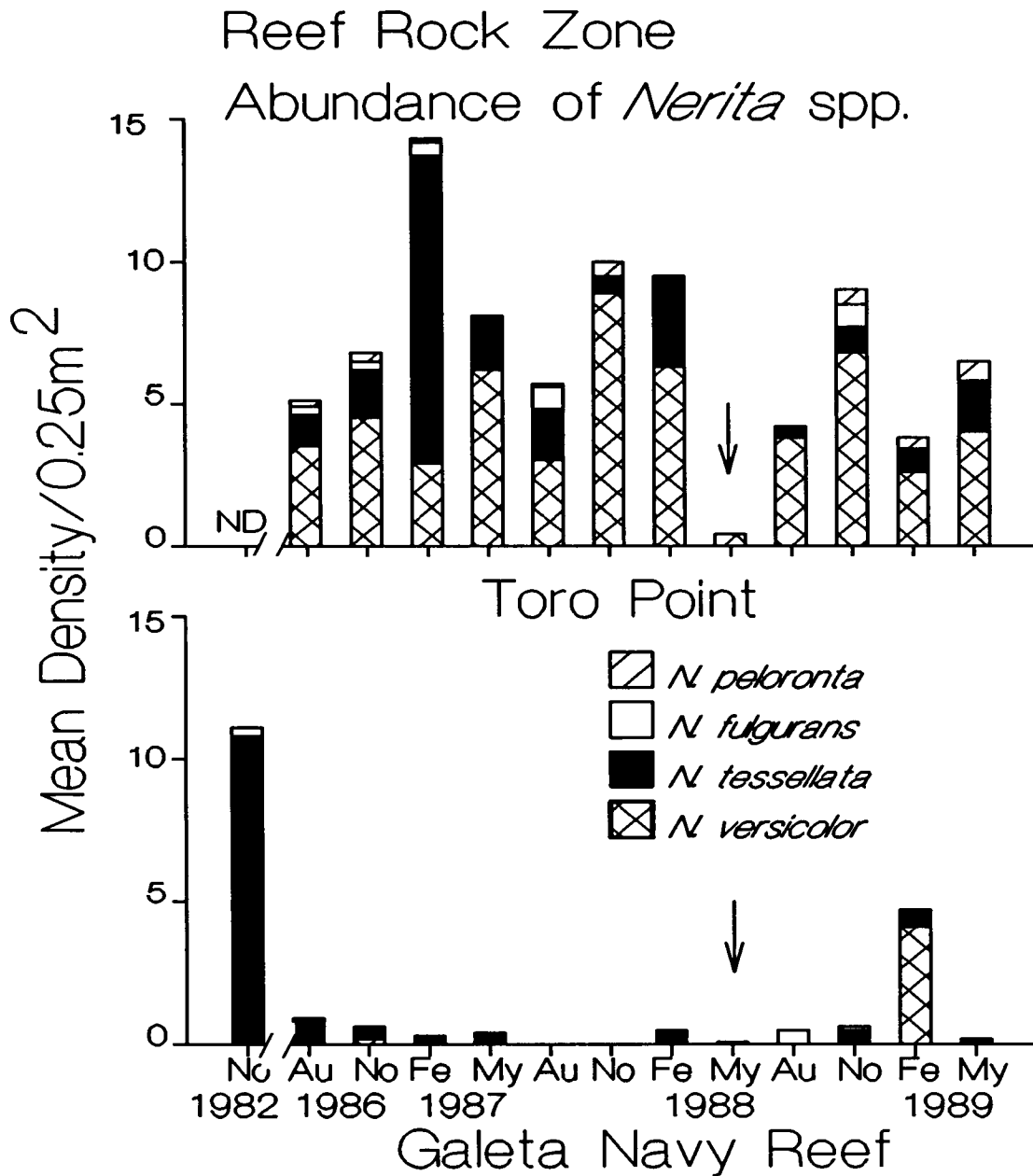


Figure 3.20 Abundance of species of *Nerita* in the reef rock zone. Differentially shaded areas of each bar are mean density of each species of neritid snail found at each monitoring period. Vertical arrow indicates monitoring done just after diesel spill at Toro Point in May 1988. See text for details. ND = No data.

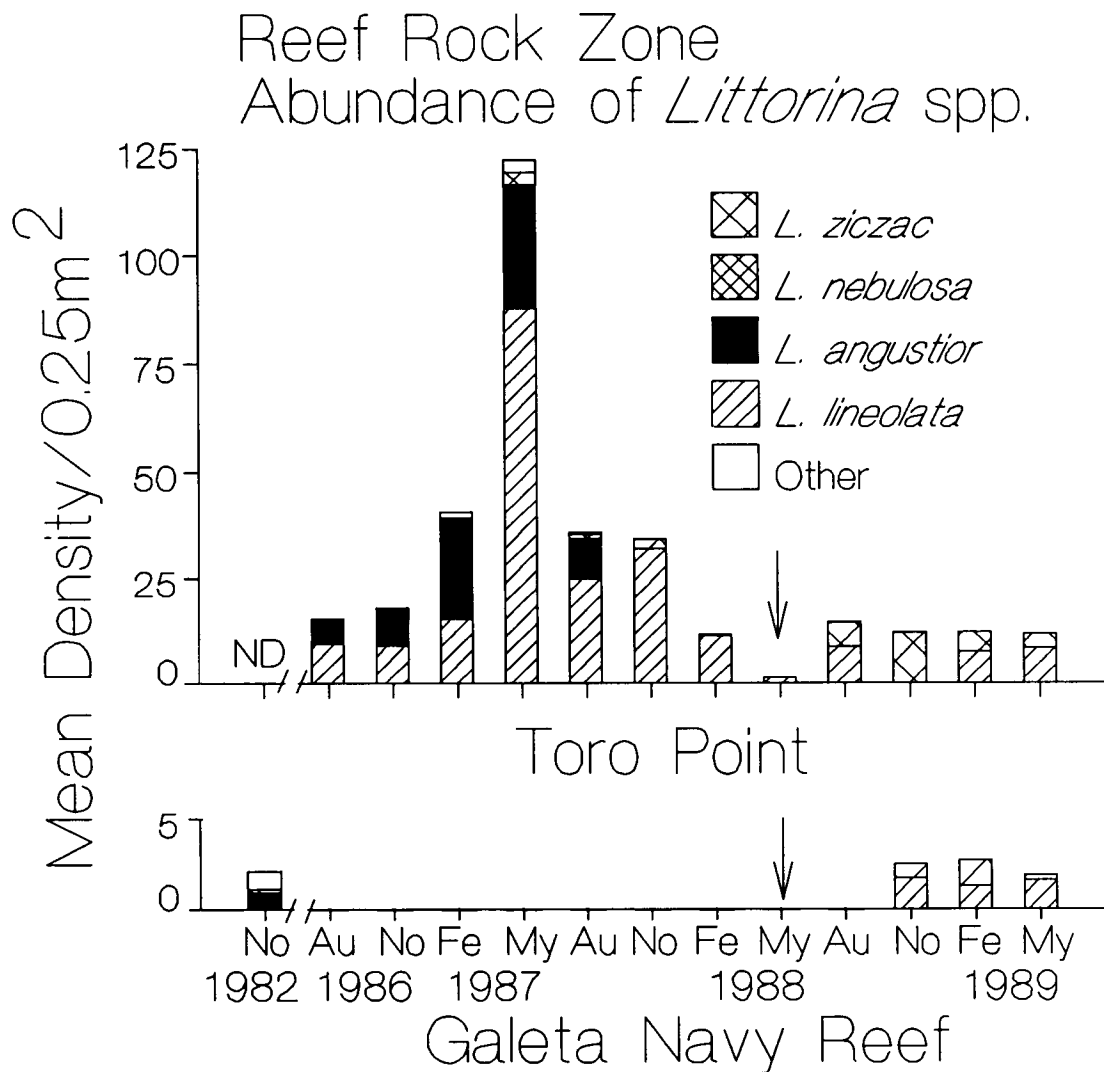


Figure 3.21 Abundance of species of *Littorina* in the reef rock zone. Differentially shaded areas of each bar are mean density of each species of littorinid snail found at each monitoring period. Vertical arrow indicates monitoring done just after diesel spill at Toro Point in May 1988. Note difference in scales between Galeta and Toro Point. See text for details. ND = No data.

Sand Zone

At Galeta Navy Reef, *Neritina* was not found and *Batillaria* abundance was far lower in August 1986 than it had been in 1982 (Tables 3.1, 3.5). No live *Batillaria* were found in November 1986. Only a few *Batillaria* were found at each monitoring until August 1987, and they were extremely rare again in November 1987. Since then, numbers have fluctuated from <1 to >50 snails/0.25 m². *Cerithium eburneum* was not found in 1982, but was common in August 1986. It was also washed seaward by cleaning, and only two individuals, found in August 1987, have since been counted. The small-bodied *Neritina*, absent in August and September 1986, appeared in high density in November 1986. Its numbers gradually declined through 1987, and none has been recorded since August 1987.

At the unoiled site (María Soto), *C. eburneum* was absent in August and rare in November 1986, after which none was found. *Batillaria* has been patchily abundant in all monitorings. *Neritina* was abundant in November 1986. None was found in February 1987 and only a few in May 1987. None has been recorded since the May 1987 monitoring.

Thalassia testudinum Zone

At Galeta Navy Reef in August 1986, *C. eburneum* was not found, and *C. literatum* was rare; in November 1986, neither species was found (Table 3.6). On the unoiled reef at María Soto, both species occurred in low abundance during the August and November 1986 monitorings. Snails have been rare at both oiled and unoiled sites since then. No snails have been recorded at either site since November 1987, except for a few *Batillaria* at María Soto in August 1988 and February 1989.

Laurencia papillosa Zone

Gastropods were rare in this zone (Table 3.7). No gastropods were found in August and November 1986 at Galeta Navy Reef, but *T. deltoidea* reappeared in low abundance in February through August 1987. Two even rarer species were found then, *Olivella* in May 1987 and *Engina* in August 1987. No snails were found in sampling from November 1987 through May 1989.

At Toro Point, one to four species were found in low abundance at each monitoring through August 1987. Since then, like at Galeta Navy Reef, no snails have been recorded. No snails were found at Toro Point in May 1988 after the diesel spill and early wet season exposures. In July 1988, an unrelated search at this site for *Morum oniscus*, a cryptic species, yielded only dead individuals (C. González, pers. obs.).

Table 3.5 Density of gastropods in the sand zone. Data are the mean number (one standard deviation)/0.25 m². N = 20 quadrats.

	Galeta Navy Reef			María Soto		
	<i>Cerithium eberneum</i>	<i>Batillaria minima</i>	<i>Neritina virginea</i>	<i>Cerithium eberneum</i>	<i>Batillaria minima</i>	<i>Neritina virginea</i>
Date:						
1986 August	24.3 (69.3)	11.1 (41.2)	0	0	901.3 (1137)	1.5 (6.5)
November	0	0	415.0 (497.4)	0.3 (1.1)	172.8 (176)	91.5 (146.3)
1987 February	0	5.1 (13.9)	116.5 (12.6)	0	418.8 (527)	0
May	0	0	71.9 (94.4)	0	26.6 (35)	10.7 (18.6)
August	0.2 (0.4)	77.4 (163.5)	20.7 (30.3)	0	164.4 (552)	0
November	0	0.3 (0.8)	0	0	96.7 (189)	0
1988 February	0	0.4 (0.8)	0	0	147.1 (233.4)	0
May	0	42.3 (121.6)	0	0	57.2 (68.6)	0
August	0	52.4 (137.6)	0	0	128.0 (182.7)	0
November	0	7.8 (12.7)	0	0	8.2 (8.4)	0
1989 February	0	14.9 (27.1)	0	0	69.3 (69.1)	0
May	0	19.1 (47.3)	0	0	24.8 (27.2)	0

Table 3.6 Density of gastropods in the *Thalassia testudinum* zone. Data are the mean number (one standard deviation)/0.25 m². N = 20 quadrats.

	Galeta Navy Reef		María Soto		
	<i>Cerithium eberneum</i>	<i>Cerithium literatum</i>	<i>Cerithium eberneum</i>	<i>Cerithium literatum</i>	<i>Batillaria minima</i>
Date					
1986 August	0	0.05 (0.2)	0.2 (0.5)	0.3 (0.6)	0
November	0	0	0.9 (1.1)	0.6 (1.5)	0
1987 February	0.1 (0.3)	0	0	0	0
May	0	0	0	0.1 (0.3)	0
August	0.1 (0.3)	0	0.7 (1.5)	0	0
November	0	0	0	0	0
1988 February	0	0	0	0	0
May	0	0	0	0	0
August	0	0	0	0	1.7 (2.6)
November	0	0	0	0	0
1989 February	0	0	0	0	0.5 (1.3)
May	0	0	0	0	0

Table 3.7 Density of gastropods in the *Laurencia papillosa* zone. Data are the mean number/0.25 m². N = 20 quadrats. - = no data. No molluscs have been recorded at either site since November 1987.

Species	1982-1983	1986		1987			
		August	November	February	May	August	November
A. Galeta Navy Reef							
<i>Thais deltoidea</i>	0.3	0	0	0.1	0.1	0.1	0
<i>Engina turbinella</i>	0	0	0	0	0	0.1	0
<i>Leucozonia nassa</i>	0.03	0	0	0	0	0	0
<i>Astraea phoebia</i>	0.2	0	0	0	0	0	0
Opisthobranch sp. 1	0.1	0	0	0	0	0	0
<i>Diodora dysoni</i>	0.1	0	0	0	0	0	0
<i>Smaragdia</i> sp.	0.3	0	0	0	0	0	0
Gastropod sp. 6	0.03	0	0	0	0	0	0
<i>Cypraea zebra</i>	1.4	0	0	0	0	0	0
<i>Olivella pusilla</i>	0	0	0	0	0.1	0	0
Total species	8	0	0	1	2	2	0
B. Toro Point							
<i>Thais deltoidea</i>	-	0.2	0.3	0.2	0.5	0.3	0
<i>Olivella pusilla</i>	-	0.4	0.2	0.1	0	0	0
<i>Mitra</i> sp.	-	0	0.1	0	0	0	0
<i>Aplysia</i> sp. 1	-	0	0	0.2	0	0	0
<i>Conus</i> sp. 1	-	0.1	0.1	0	0	0	0
<i>Heliacus ?cylindricus</i>	-	0	0	0	0	0.1	0
<i>Engina turbinella</i>	-	0	0	0	0	0.1	0
Total species	-	3	4	3	1	3	0

3.3.6 Patterns of Recruitment: 1986-1989

Changes in size classes (of shell lengths) over time in a given species were used to monitor recruitment events. Recruitment differed among species, as well as between oiled and unoiled sites. Size-frequency data on shell lengths are shown for three species with different patterns of recruitment in Figure 3.22. *Batillaria minima* (Figure 3.22), here shown from the sand zone at María Soto, showed a relatively static population size structure from August 1986 through May 1987. A sharp pulse of recruitment in August 1987 was followed by increases in shell size between November 1987 and July 1988. A lesser event of recruitment occurred in December 1988, followed by rapid growth over the dry season. The relative increase in some smaller size classes in May 1989 may not have been recruitment, since no size classes were found smaller than those found in the previous monitoring. Rather, this probably represented movement or mortality of some larger snails and was not counted as a recruitment event.

In contrast, *Tectarius muricatus*, shown from the high rubble zone at Galeta Navy Reef, remained relatively static throughout the study, with little or no change in shell size (Figure 3.22). There were no large pulses of recruitment and only occasional appearances of small numbers of small individuals. A third species, *Littorina lineolata*, from the high rubble zone at Toro Point (Figure 3.22) recruited in moderate numbers in November 1986, February 1987, August 1987, February 1988 and February 1989.

Episodes of recruitment are listed for all species abundant enough to follow in Table 3.8. Most species have planktonic larvae (Vermeij 1978), with the exception of members of the genus *Cerithium*. Dispersal and settlement should thus be independent of snail population size at any given site.

In the high rubble zone, for the first year following the spill (August 1986-May 1987), only *Littorina lineolata* recruited at Galeta Navy Reef; this species, another littorinid and two neritids recruited at Toro Point. Over the second year (August 1987 - May 1988), *L. lineolata* again was the only species to recruit in the high rubble at Galeta Navy Reef, compared to three littorinids and two neritids at Toro Point. In the final year, *L. ziczac* and *Nodilittorina tuberculata* recruited at Galeta Navy Reef, and these two plus *L. lineolata* and *Tectarius* recruited at the unoiled site. Overall, between August 1986 and May 1989, four pulses of recruitment were recorded in the high rubble zone at Galeta Navy Reef, while 13 were observed at Toro Point, and there were three species that recruited only at Toro Point (Table 3.8).

In the low rubble zone, ten species recruited at Toro Point between August 1986 and May 1987, compared to five at Galeta Navy Reef (Table 3.8). Only one of these five, *L. lineolata*, did not also recruit at the unoiled site in this zone; it did in the high rubble zone. In both the second and third years following the spill, about the same number of species recruited in the low rubble zone, although individual

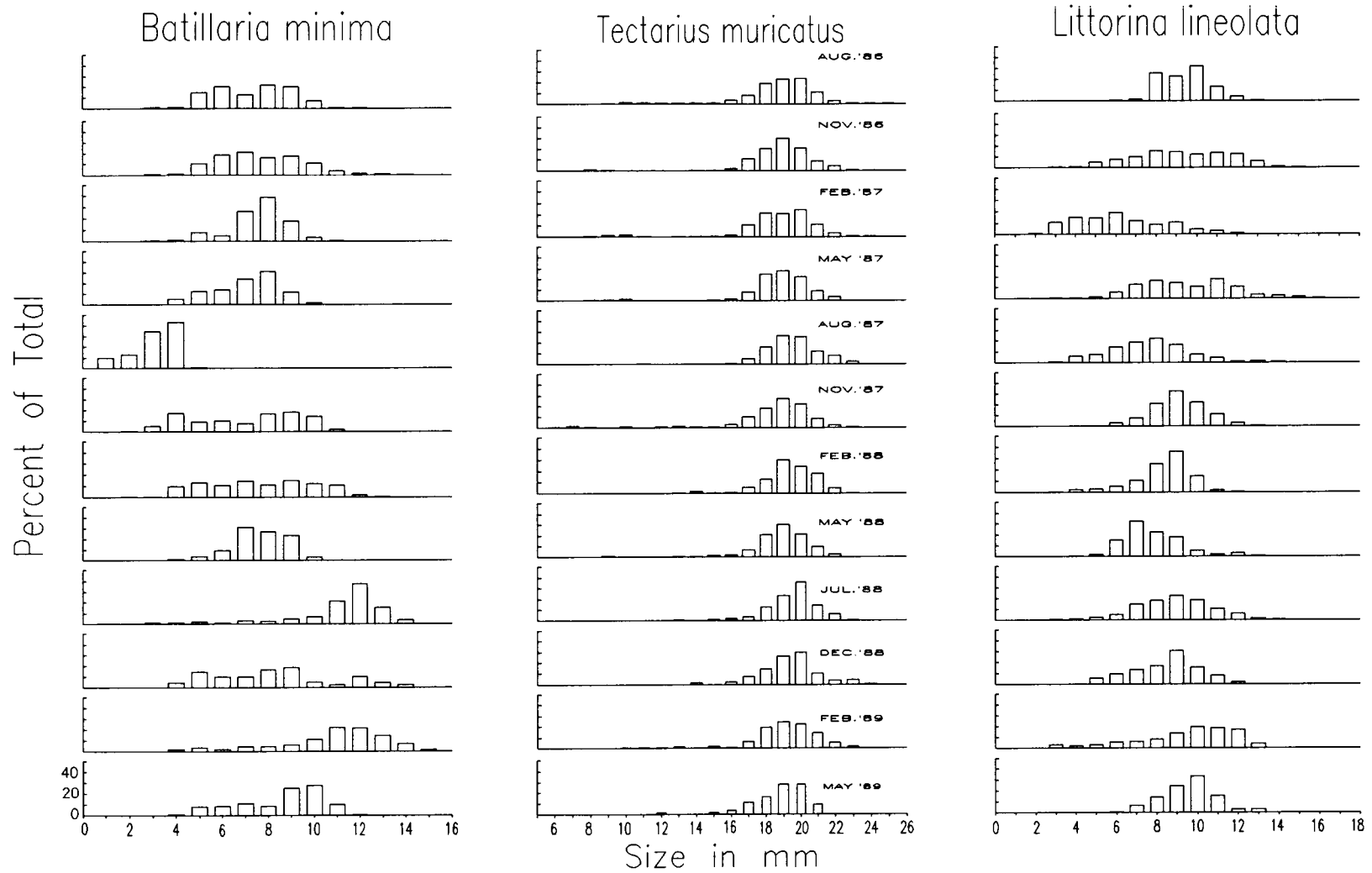


Figure 3.22 Temporal changes in gastropod size-frequency. Examples of recruitment in three species : *Batillaria minima*, *Tectarius muricatus*, and *Littorina lineolata*.

Table 3.8 Episodes of recruitment: 1986-1989

ZONE	DATE:		1986				1987				1988				1989		Total
	No		Fe	My	Au	No	Fe	My	Au	No	Fe	My					
High Rubble																	
Galeta Navy Reef			LL				LL		LZ	NT						4	
Toro Pt.	LZ		LN		LL		LL	LZ		LZ		LL				13	
	LL		NT		NV			LN				NT	TM				
Low Rubble																	
Galeta Navy Reef			LL	PN	LZ			PN	NV	LL		NF	PL			15	
			NS		PL				NS	NT							
			NV						NF								
			NT														
Toro Pt.	LZ		LA		PN			PN	BM	LZ		LL	PN			19	
	LL		LM		NF							NS	BM				
			LN														
			NF														
			NV														
			NS														
			NT														
			PN														
Reef Rock																	
Galeta Navy Reef										LL		NV				4	
												LZ	NS				
Toro Pt.			LA	LM	NS		NV		LZ	NF		LL				15	
			LL	LZ					NV	NS							
			NT	NV													
			NS														
			NF														
Sand																	
Gal. Navy Reef	NE		BM		BM							BM				4	
María Soto	NE		NE	BM						BM						4	

Abbreviations: BM = *Batillaria minima*, LA = *Littorina angustior*, LL = *L. lineolata*, LM = *L. meleagris*, LN = *L. nebulosa*, LZ = *L. ziczac*, NF = *Nerita fulgurans*, NS = *N. tessellata*, NV = *N. versicolor*, NE = *Neritina virginea*, NT = *Nodilittorina tuberculata*, PL = *Planaxis lineatus*, PN = *P. nucleus*.

species varied between sites (Table 3.8). Over the three years, 15 episodes of recruitment occurred at Galeta Navy Reef compared to 19 at Toro Point. One species, *Planaxis lineatus*, recruited only at Galeta Navy Reef, while three species recruited only at Toro Point (Table 3.8).

On reef rock, there was no recruitment at Galeta Navy Reef in either year 1 or 2 following the spill. At Toro Point, eight species recruited during year 1, but only two in the second year. Recruitment was equivalent between sites in the third year, with four species at Galeta Navy Reef and five at Toro Point. Overall, there were four recruitment events in the three years after the spill at Galeta Navy Reef, compared to 15 in the unoiled site's reef rock zone, and four species that recruited at Toro Point during this period did not recruit at Galeta Navy Reef (Table 3.8).

On intertidal sand, there were no differences in the number of recruitment events between sites. Overall, there were four episodes of recruitment at Galeta Navy Reef between August 1986 and May 1989, and four occurred at María Soto (Table 3.8). There were no unique species that recruited to either site.

Recruitment was impossible to follow in *Thalassia* and *Laurencia* zones, due to the low abundance of gastropods in these zones from August 1986 through May 1989. In the former zone, two species of cerithids had been abundant in 1982-3, indicating either a general disappearance of the genus in this region or a very patchy population structure. In the *Laurencia* zone, mobile molluscs are generally rare, and recruitment may be low or variable or both.

3.3.7 Summary of Results

Results of this study suggest the oil spill of 1986 affected the gastropods of the Galeta Navy Reef differentially, primarily as a result of their location on the reef. Effects appear minor in the high rubble zone. Wetted only by wave splash, this zone received only small amounts of oil. Many snails found in the six months after the spill had oil on their shells, and some newly dead individuals were recovered in August and November 1986. However, it is likely that these individuals had moved upwards from the low rubble zone in response to oiling below (*Littorina angulifera*, the mangrove littorinid, was observed to move up to 10 m in trees following oiling in the mangroves; pers. obs.). Activities of a cleanup crew resulted in disturbance to some species, but this appeared temporary. Overall, decreased recruitment relative to that at Toro Point appeared to be the major long-term effect. Species number and abundance have fluctuated at both sites since the spill, but have shown a downward trend, at least partly due to habitat loss unrelated to the spill. If recruitment continues to be below normal at Galeta Navy Reef relative to Toro Point, there will be a gradual reduction in gastropod density, an increase in larger size classes of snails, and eventually a further decrease in species number.

Data from the low rubble zone suggest snails were killed initially by oil at Galeta Navy Reef; some species showed a further reduction in abundance immediately after the activities of a cleanup crew. Since then, despite the complicating factors of habitat loss and snail mortality (or movement) correlated with

dry season storms, data indicate persistent effects of oiling in this zone through May 1988. By August 1988, snails were as abundant as they had been in 1982. In addition to the initial mortality of snails after the spill, there may have been decreased or failed recruitment during the first year after the spill. Recruitment in the second and third years after the spill appeared to be equivalent at Galeta Navy Reef and Toro Point. The diesel spill at Toro Point had strong immediate effects, but was followed by the recruitment of several species and a return to prior patterns of abundance. This suggests few long-term detrimental effects of this small spill in the low rubble zone.

Our data show strong, persistent effects of oiling at Galeta Navy Reef in the reef rock zone, where species number as well as snail abundance was reduced sharply, relative to both conditions prior to the spill and to the pattern of population change at the unoiled site. Recruitment failed completely for two years after the spill, resuming only in November 1988. There was a similar, strong reduction in density (and perhaps recruitment) at Toro Point in May 1988 after a relatively small diesel spill, but populations quickly recovered to previous levels.

In the sand zone, data suggest snail populations may have been initially affected by oiling and further disturbed by the clean-up procedure. Since then, short-lived pulses of recruitment have caused large variations in gastropod density at both oiled and unoiled sites. Oiling had no detectable effects on snails found in *Thalassia*, given the variation in populations over time. We have no explanation for the paucity of gastropods in *Thalassia* beds since 1986 relative to 1982.

In *Laurencia* beds, data from Galeta Navy Reef suggest a reduction in snail populations after the oil spill, followed by a gradual recovery. It is possible that snails moved up from crevices deep in the reef flat or from subtidal populations. The small diesel spill at Toro Point in May 1988 had no detectable effects, because snails had been absent in previous monitorings.

3.4 Discussion

The effects of the oil spill at Bahía Las Minas on gastropod molluscs inhabiting the nearby Galeta reef flat must be evaluated cautiously. Conclusions are speculative, given: (1) the four year time period between the pre- and post-oil spill monitoring, (2) the natural scarcity of gastropods in some habitats, (3) the patchy distribution of many species, (4) the unknown extent of natural variation, and (5) lack of data before the spill from Toro Point and María Soto. Despite this, there is evidence that the Bahía Las Minas oil spill had strong effects upon the molluscs of some zones on the Galeta reef flat. Especially important are (1) observations of dead snails glued to the rock by oil, (2) recovery of large numbers of dead snails at Galeta when none were recorded at Toro Point and (3) failure of recruitment in the most heavily oiled zones.

Of the habitats sampled, oil was most heavily concentrated in the low rubble and the emergent portions of the reef rock zones, where wave action and the rise and fall of tides added layer upon layer of oil to rocky surfaces following the spill

(J. Cubit, pers. comm.). Relatively little oil splashed up onto the high rubble, above the level of the tides. The intertidal sand, *Thalassia* and *Laurencia* zones were also heavily oiled, but the physical evidence quickly disappeared as sand shifted, oiled plants rotted and drifted away, and ephemeral algae bloomed.

The observed pattern of oiling was due to the complicated pattern of tidal emersion at Galeta (Cubit *et al.* 1986, 1989). By April and May, onshore winds, which usually keep water levels high and mask tidal fluctuations on the reef flat, have dropped. This results in much of the surface of the reef flat being periodically emersed. In contrast, an oil spill during the dry season, when water levels and onshore winds are consistently high, would have concentrated oil on or at the edge of the high rubble zone. Lower zones of the reef flat would probably have had little direct contact with surface slicks of oil, and have been affected only by oil mixed into the water column. The latter scenario appears to have occurred when the Galeta reef flat was oiled during dry season by the tanker "Witwater" spill in 1968 (Rützler and Sterrer 1970). Thus the effects of the oil spill are intimately dependent upon the pattern of wind and tides, combined with small differences in elevation across the reef flat (e.g., Connor 1984).

A small diesel spill at Toro Point occurred following the grounding of a 75 foot sailboat on May 14, 1988. Weather conditions were similar to those during the Bahía Las Minas spill. Intertidal reef gastropods were experiencing extended periods of daytime emersion. Extreme daytime exposures (J. Cubit, pers. comm.) occurred for five or more hours each day both before and during the May 1988 monitoring period (4/19-22, 5/2-5, 5/16-22, and 5/29-6/7). Although low water levels occur every year at this time, 1988 had an unusually high number of exposures (J. Cubit, personal communication; Chapter 2). Mobile molluscs behaviorally reduce physical stress from heat and desiccation while emersed by moving lower on the shore, moving to vertical surfaces or crevices, or becoming inactive under cobble, coral rubble, sand or debris (Garrity 1984). Some of these behaviors affected exposure of snails to diesel fuel at Toro Point. Fuel collected in depressions and on sand in the reef rock and low rubble zones (C. González, personal observations).

Physical stress alone does not explain the observed mortality at Toro Point. First, several hundred dead, freshly oiled individuals of six snail species were collected during the May 1988 monitoring at Toro Point; no such dead snails were found either at Galeta Navy Reef or María Soto. Of the 206 *Batillaria minima* counted in quadrats in the low rubble zone at Toro Point, all were dead, while of the 237 individuals counted in quadrats from the sand zone at María Soto, all were alive. Second, snail abundance generally increased at Toro Point from May to August 1988, despite continued daytime reef exposures in June, July and early August. A number of species showed recruitment during these extended exposures, further suggesting adaptation of snail population to such exposures.

As a result of the pattern of oil deposition from the 1986 refinery spill, both reef rock and low rubble zones showed major and persistent changes in gastropod abundance at Galeta Navy Reef. These zones at Toro Point appeared briefly affected by a small diesel spill in May 1988, the same time of year as the spill at

Galeta Navy Reef. At Galeta Navy Reef, there was little evidence of recovery within the reef rock zone until three years after the spill and its resultant mortality; recruitment was strongly affected until late 1988.

The high rubble zone received relatively little oil, and populations of snails have varied since the spill at both oiled and unoiled sites. The presence of large numbers of oiled snails in this zone soon after the spill suggests vertical movement of snails from lower, more heavily oiled zones. The extent and timing of normal snail movement and migration may help explain differential mortality among species and may be a factor in some of the changes in abundance seen at both sites since the spill (e.g., Peckol *et al.* 1989). Activity patterns of gastropods elsewhere have been shown to influence susceptibility to pollutants (Dicks 1976). This aspect remains to be investigated.

In the *Laurencia* zone, several species of gastropods are normally rare. Since the oil spill, few to no molluscs have been found at Galeta Navy Reef. However, the same pattern has been observed at the unoiled site, indicating that oiling alone is not responsible for the reductions. More detailed examination of mollusc populations, perhaps focusing on micromolluscs, might have provided more conclusive results.

In the sand and *Thalassia* zones at Galeta Navy Reef, there were strong differences in snail abundance between 1982 and post-spill monitorings. The oil spill or subsequent cleanup operations (or both) eliminated *Batillaria* from intertidal sand at Galeta Navy Reef. Numbers since have fluctuated, probably depending upon pulses of opportunistic settlement, and suggesting few persistent effects of oiling. In the *Thalassia* bed, two species of *Cerithium* were present at Galeta Navy Reef before the spill and were absent or rare after the spill. However, at the unoiled site both species were also rare in 1986. They have commonly been reported in *Thalassia* beds (e.g., Heck 1977), but have been rare along the entire coast during this study (S. D. Garrity, pers. obs.). We have no explanation for this phenomenon. Although oil may indeed have affected the gastropods of the *Thalassia* bed at Galeta Navy Reef, results could also be explained by unrelated population fluctuations along this coast.

Results from sand and *Thalassia* beds highlight the potential for variation in recruitment among sites (e.g., Caffey 1983) to confound our results. Because only two sites could be monitored for each zone, we cannot separate random variation at each site from effects of oiling. Persistence of effects over time may indicate that recruitment is reduced at oiled sites, or merely that an individual site tends to have low recruitment.

This general problem strongly affects studies of pollution (e.g., Jones 1982; Lewis 1982; Underwood and Peterson 1988). Oil spills are unplanned events that occur rarely in areas where long-term monitoring programs have been in progress for many years or where the life histories of resident organisms are well known. As with many ecological problems, data from tropical habitats are even rarer than data from the temperate zone (but see Birkeland *et al.* 1976; Eisler 1973, 1975 a, b; Chan 1976, 1977; Maynard *et al.* 1977; Maynard 1984; Cubit *et al.* 1986). Despite its imperfections, the present study represents the only long-term data available for tropical molluscs. Interpreted cautiously, the data show thus far that (1) mortality

was extensive in some sections of the reef flat and (2) recovery had not been completed three years after the spill. The separation of natural population fluctuations from those caused by oil and any evaluation of time to final recovery must await further study.

3.5 Acknowledgments

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Chapter 4 Reef Flat Stomatopods

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4.1 Introduction

Gonodactylid stomatopods are benthic, marine crustaceans that occupy and defend cavities in hard substrata as refuges from predators. Gonodactylids are principal components of many intertidal communities due to their position as both important predators and prey. As prey, they serve as an abundant source of food for herons, egrets, fishes, and octopuses (Reaka 1985; Caldwell 1986). As predators, they are capable of structuring local populations of hermit crabs and gastropods, their primary prey (Caldwell *et al.* 1989). In addition, gonodactylids are used often in research on the function and expression of aggressive behavior because of their combativeness and highly developed systems of communication (for reviews: Caldwell and Dingle 1976; Reaka and Manning 1981; Steger 1985).

The population biology, ecology, and behavior of gonodactylids have been studied in great detail around the Galeta Marine Laboratory of the Smithsonian Tropical Research Institute (STRI) on the Caribbean coast of Panama (Figure 4.1). These studies were carried out over a five year period from 1979 to 1983. This area was chosen because gonodactylids were extremely abundant at many accessible sites near the laboratory. Most of the current information on the ecology and behavior of gonodactylid stomatopods, including important findings on the use of aggression, has come from that research program (e.g., Caldwell 1982; Berzins and Caldwell 1983; Steger and Caldwell 1983; Montgomery and Caldwell 1984; Steger 1985; Caldwell *et al.* 1989; Steger 1987).

In April 1986, a major oil spill occurred in Bahía Las Minas near the Galeta Marine Laboratory. We returned to Panama in September 1986 to assess the impact of the spill on our study sites. Although the findings of this initial study have been published (Jackson *et al.* 1989), we will summarize the most important details here. We surveyed gonodactylid communities in intertidal beds of turtle grass, *Thalassia testudinum*, on reef flats at Isla Margarita, Isla Mina, and north- and west-facing reef

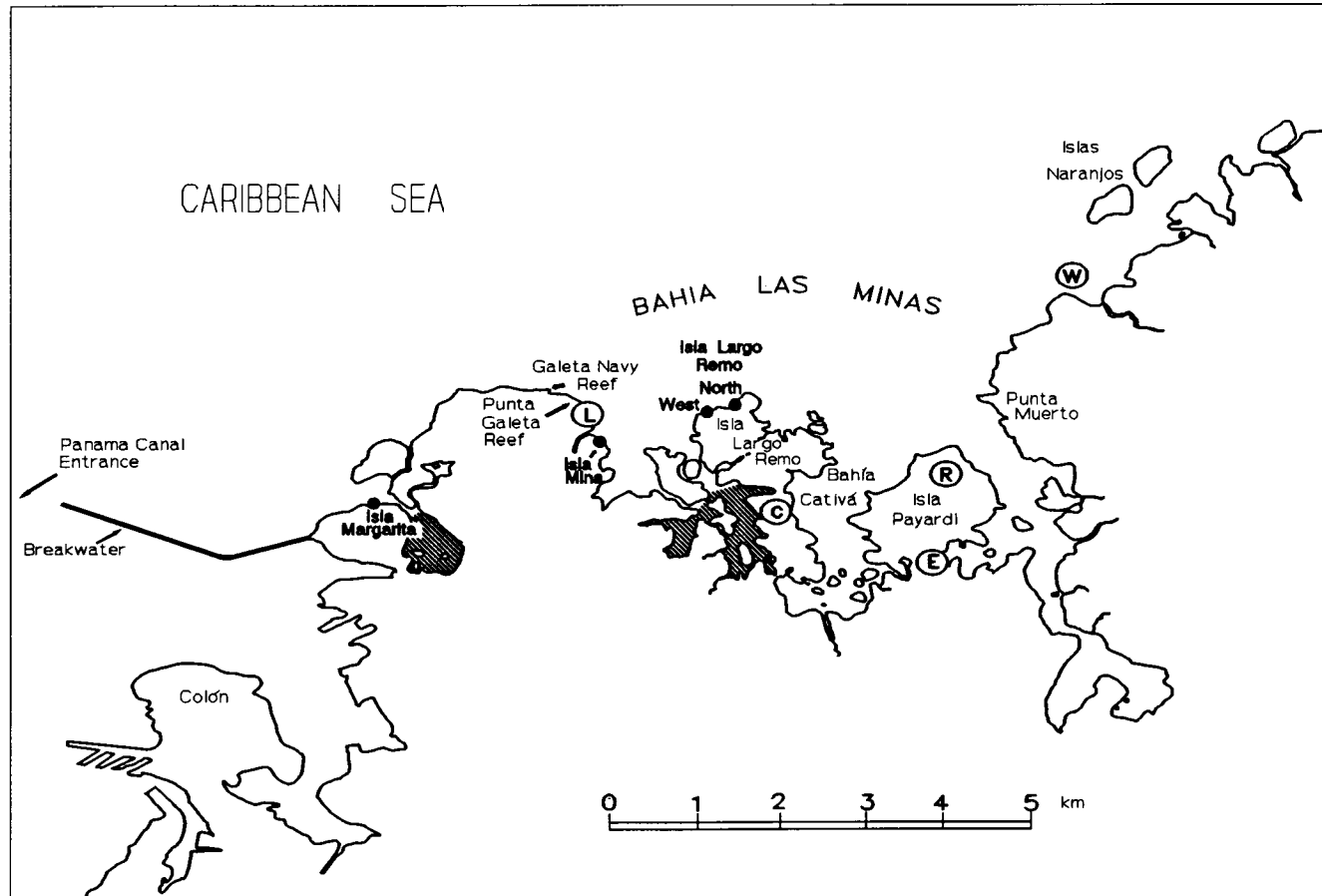


Figure 4.1 Map showing the four study sites. Isla Largo Remo North and Isla Mina were heavily oiled sites; Isla Largo Remo West and Isla Margarita were unoiled or lightly oiled reference sites. Refer to Figure 1.1 for location of the study area in Panama and for further details.

flats at Isla Largo Remo (Figure 4.1). The two Isla Largo Remo sites will be referred to below as Isla Largo Remo North and Isla Largo Remo West (abbreviated as Isla LR-North, Isla LR-West). We focused on intertidal *Thalassia* beds because previous work has shown that this is the primary habitat of gonodactylids throughout the area (Steger 1987).

These particular study sites were chosen for three reasons. First, given the time allocated for field work, we found that we could sample no more than four sites total. Second, these sites were among the sites sampled in previous years before the oil spill. Third, visual inspection of the areas and conversations with the STRI personnel present during the spill indicated that these sites could be separated into two groups: potentially affected sites that were heavily oiled (Isla Largo Remo North and Isla Mina) and reference sites that were lightly oiled to unoiled (Isla Largo Remo West and Isla Margarita).

The results of the initial survey in September 1986 indicated that the preliminary separation of the four sites into affected and reference areas was correct. We had four major findings:

1. There were significantly fewer large gonodactylids, particularly females, at Isla Largo Remo North and Isla Mina, the heavily oiled sites.
2. Individuals occupied significantly larger cavities at Isla Largo Remo North, a heavily oiled site, than previously measured for *Gonodactylus* in 1980.
3. Individuals carried significantly fewer wounds and injuries at Isla Largo Remo North and Isla Mina.
4. Individuals grew more per molt at Isla Largo Remo North and Isla Mina.

These findings were based on comparing data from each site to data collected 1979-83 at the same locations, prior to the oil spill, and by comparing the heavily oiled sites with the lightly oiled to unoiled reference sites.

The results appear to have been caused by the mortality or emigration of larger gonodactylids at heavily oiled sites, which allowed a release from competition for cavities. Competition for cavities by *Gonodactylus* limits the number of individuals, especially larger individuals, in the preferred habitat (Steger 1987) and forces animals of all sizes to live in cavities that are smaller than preferred (Steger 1985). When the number of larger gonodactylids was reduced at heavily oiled sites, their cavities became available and allowed smaller individuals to move into larger, more preferred cavities. The reduced competition for refuges was further reflected in the reduced levels of wounds and injuries among the remaining gonodactylids at heavily oiled sites. The increased growth rate at heavily oiled sites may have been related to reduced competition for both cavities and food. It should be noted, however, that there was an apparent explosion in the numbers of hermit crab prey in the affected sites, which also could have added to the growth rate of gonodactylids at heavily oiled sites.

4.2 Sampling Methods

Our goal for monitoring was to repeat the sampling regime established in September 1986. The procedures for three types of samples (area quadrats, rubble, and cavity volumes) established in the initial survey were continued at the same sites. Based on the population biology and ecology of gonodactylids in these areas, sampling was conducted at each site every six months, in September and February. The resulting data were minimal but sufficient to track any changes at the locations and to detect any differences in seasonal effects between heavily oiled and reference sites.

We continued to remove thirty 0.5 m² quadrats from a 400 m² area reserved at each site for quadrat samples. The position of quadrats was determined by laying down x,y coordinate axes and using pairs of random numbers to locate the corner of each quadrat. We recorded the water height and physical characteristics within the quadrat and removed all hard substrata. Pieces of hard substrata were measured and then broken in order to collect all of the stomatopods. The quadrat samples were used to identify changes in the habitat and to estimate densities of rubble and gonodactylids per unit area.

Pieces of coral rubble were collected from areas adjacent to the quadrat areas at each site. Each piece was selected haphazardly, examined for cavities, and placed in a plastic bag. The criteria for selecting a piece were that it contain at least one hole that a stomatopod could use as a refuge and that it be representative of rubble at the site (i.e., that it be neither extremely large nor extremely small when compared to the majority of rubble at the site). Each piece was then measured, broken, and all stomatopods were removed. Rubble samples were used to calculate densities of *Gonodactylus* per unit volume of rubble and to generate sufficient numbers of individuals for estimating growth, reproduction, recruitment, and the frequency of injuries.

We monitored the pattern of cavity utilization by bringing intact pieces of coral rubble into the laboratory. Each resident was harrassed into leaving its cavity and then the volume of the cavity was determined by the volume of lead shot required to fill it. The relationship between the size of a gonodactylid and the volume of its cavity was measured at Isla Largo Remo North, a heavily oiled site, during the initial impact study in September 1986. Due to limited time, we were not able to collect these data at the other sites. However, as these data turned out to be critical to our interpretation of the non-lethal effects of the oil spill on gonodactylids, we measured the relationship between gonodactylids and their cavities at three sites starting February 1987. Cavities were measured at Isla Largo Remo North (a heavily oiled site), Isla Largo Remo West (a reference site most similar physically to Isla Largo Remo North), and Isla Margarita (a reference site where cavity data had been collected in 1980 prior to the oil spill). The relationship between the size of the resident and the volume of its cavity was used to assess the intensity of competition.

All gonodactylids collected in the field were brought to the laboratory and examined under a dissecting microscope to determine species, sex, size, injuries, and

the reproductive state of females. We then maintained each animal in an individual container outside of the laboratory under ambient conditions for three days. If animals molted during this period or were collected in the field with their molt skins, the exuviae were used to calculate growth per molt by measuring the change in carapace length.

4.3 Results

The densities of *Gonodactylus* per unit area from quadrat samples were analyzed for four size categories: all sizes combined, small (5-15 mm total length), medium (16-40 mm total length), and large (>40 mm total length). Statistical analyses were carried out by Kruskal-Wallis one-way analysis of variance (ANOVA) (Wilkinson 1986) run on data from each site across the four sampling periods covered by this report (Tables 4.1 - 4.4). Significance levels were set at 0.05. For brevity, lightly oiled to unoiled reference sites will be referred to simply as reference sites.

The mean densities of *Gonodactylus* of all sizes decreased significantly at Isla Largo Remo North, a heavily oiled site, and Isla Margarita, a reference site, between September 1986 and February 1988 (Table 4.1). The mean densities of *Gonodactylus* of all sizes did not vary significantly at Isla Mina, a heavily oiled site, and Isla Largo Remo West, a reference site, during the same period (Table 4.1).

The densities of large *Gonodactylus*, individuals greater than 40 mm in total length, did not vary significantly at both reference sites, Isla Largo Remo West and Isla Margarita, and at one heavily oiled site, Isla Largo Remo North, between September 1986 and February 1988 (Table 4.2). In contrast, the density of large *Gonodactylus* increased significantly at Isla Mina, a heavily oiled site (Table 4.2). The increase in density of large *Gonodactylus* at Isla Mina took place between September 1986, when no gonodactylids larger than 40 mm were found in quadrat samples, and February 1987, when the density of large gonodactylids was 0.5 individuals per 0.5 m². The density of large *Gonodactylus* did not vary significantly at Isla Mina between February 1987 and February 1988, after that initial increase (Kruskal-Wallis one-way ANOVA performed on Isla Mina data in Table 4.2 excluding the September 1986 sample, $P = 0.64$).

The densities of medium-sized *Gonodactylus*, 16-40 mm in total length, decreased significantly at Isla Largo Remo North, a heavily oiled site, and at Isla Margarita, a reference site, between September 1986 and February 1988 (Table 4.3). The densities of medium-sized *Gonodactylus* did not vary significantly at Isla Mina, a heavily oiled site, and Isla Largo Remo West, a reference site, during this period (Table 4.3).

Table 4.1 Mean densities of *Gonodactylus* per 0.5 m². Means are for 30 quadrats taken at each site in September (S) and February (F) of the years indicated. Standard errors are in parentheses. Reference sites were lightly oiled to unoiled.

		S 86	F 87	S 87	F 88
Heavily Oiled	Isla Mina	2.5 (0.42)	2.8 (0.44)	2.2 (0.43)	2.8 (0.45)
	Isla LR-North	* 0.9 (0.20)	0.6 (0.31)	0.7 (0.21)	0.2 (0.10)
Reference	Isla LR-West	3.0 (0.75)	2.2 (0.51)	1.4 (0.37)	2.4 (0.86)
	Isla Margarita	* 5.0 (0.73)	3.6 (0.66)	3.0 (0.66)	2.8 (0.56)

* Kruskal - Wallis ANOVA : Isla LR-North, P=0.005; Isla Margarita, P=0.038 .

Table 4.2 Mean densities of large *Gonodactylus* per 0.5 m². Means are for gonodactylids greater than 40 mm total length from 30 quadrats taken at each site in September (S) and February (F) of the years indicated. Standard errors are in parentheses. Reference sites were lightly oiled to unoiled.

		S 86	F 87	S 87	F 88
Heavily Oiled	Isla Mina	* 0	0.5 (0.17)	0.4 (0.12)	0.3 (0.11)
	Isla LR-North	0.03 (0.033)	0.2 (0.11)	0.07 (0.046)	0.03 (0.033)
Reference	Isla LR-West	0.5 (0.26)	0.6 (0.18)	0.5 (0.17)	0.6 (0.25)
	Isla Margarita	0.4 (0.12)	0.1 (0.063)	0.3 (0.095)	0.2 (0.11)

* Kruskal - Wallis ANOVA : Isla Mina, P=0.014 .

Table 4.3 Mean densities of medium-sized *Gonodactylus* per 0.5 m². Means are for gonodactylids 16-40 mm in total length from 30 quadrats taken at each site in September (S) and February (F) of the years indicated. Standard errors are in parentheses. Reference sites were lightly oiled to unoiled.

		S 86	F 87	S 87	F 88
Heavily Oiled	Isla Mina	2.2 (0.37)	2.0 (0.28)	1.7 (0.36)	1.5 (0.30)
	Isla LR-North	* 0.9 (0.20)	0.4 (0.19)	0.6 (0.19)	0.1 (0.074)
Reference	Isla LR-West	2.1 (0.47)	1.2 (0.33)	1.0 (0.25)	1.2 (0.48)
	Isla Margarita	* 4.0 (0.57)	2.3 (0.46)	2.5 (0.57)	1.3 (0.33)

* Kruskal - Wallis ANOVA : Isla LR-North, P=0.001; Isla Margarita, P<0.001 .

Table 4.4 Mean densities of small *Gonodactylus* per 0.5 m². Means are for gonodactylids 5-15 mm in total length from 30 quadrats taken at each site in September (S) and February (F) of the years indicated. Standard errors are in parentheses. Reference sites were lightly oiled to unoiled.

		S 86	F 87	S 87	F 88
Heavily Oiled	Isla Mina	* 0.2 (0.092)	0.3 (0.10)	0.1 (0.056)	1.0 (0.22)
	Isla LR-North	0	0.03 (0.033)	0	0.07 (0.046)
Reference	Isla LR-West	* 0.4 (0.16)	0.3 (0.15)	0	0.6 (0.22)
	Isla Margarita	* 0.6 (0.16)	1.2 (0.33)	0.2 (0.092)	1.3 (0.24)

* Kruskal - Wallis ANOVA : Isla Mina, P<0.001; Isla LR-West, P=0.006; Isla Margarita, P<0.001 .

The densities of small *Gonodactylus*, 6-15 mm in total length, varied significantly at both reference sites, Isla Margarita and Isla Largo Remo West, and at Isla Mina, a heavily oiled site, between September 1986 and February 1988 (Table 4.4). However, the density of small *Gonodactylus* did not vary significantly at Isla Largo Remo North (Table 4.4).

The relationship between the size of a gonodactylid and the volume of its cavity was measured during September 1986, February 1987, September 1987, and February 1988 at Isla Largo Remo North and during February 1987, September 1987, and February 1988 at Isla Largo Remo West and Isla Margarita. The same data were taken prior to the oil spill during 1980 at Isla Margarita. An exponential model ($y = ae^{bx}$) best fit the data where the logarithmic transforms of the cavity volumes were regressed on the total length of *Gonodactylus* residents. There was a significant, linear relationship between the logarithm of cavity volume and the total length of its resident during all sampling periods at all sites (Table 4.5).

Table 4.5 Summary of Model I regression parameters. The logarithmic transforms of the cavity volumes (cc) were regressed on the total length (mm) of *Gonodactylus* residents. An exponential model ($y = ae^{bx}$) best fit the data.

	N	b*	a	Pearson's Correlation
Isla Largo Remo North				
September 1986	46	.081	.68	0.70
February 1987	53	.070	.82	0.78
September 1987	56	.113	.13	0.87
February 1988	54	.097	.29	0.80
Isla Largo Remo West				
February 1987	54	.084	.30	0.90
September 1987	59	.093	.23	0.94
February 1988	56	.089	.27	0.90
Isla Margarita				
Jan - Dec 1980	148	.086	.26	0.89
February 1987	54	.077	.40	0.89
September 1987	41	.114	.10	0.86
February 1988	51	.100	.16	0.82

* Analysis of variance, $H_0: b = 0$, all $P < 0.001$.

Cavity volume regressions were analyzed in three groups of comparisons by analysis of covariance (ANCOVA) (Wilkinson 1986). The three types of comparisons were (1) pairwise comparisons of post-spill regression statistics between samples within each site (Table 4.6); (2) pairwise comparisons of post-spill regression statistics between sites within the same sampling period (Table 4.7); and (3) pairwise comparisons of pre-spill regression statistics from Isla Margarita (1980) with post-spill statistics from each site (Table 4.8). The ANCOVA analyses were made by first testing for the homogeneity of slopes by analysis of variance (ANOVA). If there was no significant difference between the slopes, the regressions were compared by testing for differences among the intercepts by ANOVA. Considering the number of comparisons made, a conservative approach was adopted by setting the significance level at 0.01 for each test.

Table 4.6 Significance of ANCOVAs of cavity volume vs. stomatopod size for post-spill comparisons between dates. Regression statistics were obtained from the regressions of cavity volume (cc) on the total length (mm) of *Gonodactylus* residents during September (S) and February (F) of the years indicated.

		Slopes Homogeneous	Intercepts Equal
Isla Largo Remo West			
	F87 vs S87	0.2	0.1
	S87 vs F88	0.6	0.9
	F87 vs F88	0.5	0.2
Isla Margarita			
	F87 vs S87	0.001 *	
	S87 vs F88	0.2	0.5
	F87 vs F88	0.005 *	
Isla Largo Remo North			
	S86 vs F87	0.5	0.1
	F87 vs S87	< 0.001 **	
	S87 vs F88	0.2	0.03
	S86 vs S87	0.04	< 0.001 **
	F87 vs F88	0.04	0.8
	S86 vs F88	0.3	0.04

* P<0.01, ** P<0.001

Starting with the first set of comparisons (within site, between post-spill samples), we found no significant differences between any of the cavity volume regressions measured at Isla Largo Remo West (Table 4.6). At Isla Margarita, the slope of the cavity volume regression for February 1987 was significantly different from the slopes for September 1987 and February 1988, but was not for the September 1987 - February 1988 comparison (Table 4.6). At Isla Largo Remo North, the slope of the cavity volume regression for February 1987 was significantly different from the slope for September 1987. Although there were no significant differences between the slopes for the rest of the regressions measured there, the intercept of the cavity volume regression for September 1986 was significantly different from the intercept for September 1987 (Table 4.6).

Table 4.7 Significance of ANCOVAs of cavity volume vs. stomatopod size for post-spill comparisons between sites. Regression statistics were obtained from the regressions of cavity volume (cc) on the total length (mm) of *Gonodactylus* residents during September (S) and February (F) of the years indicated.

	Slopes Homogeneous	Intercepts Equal
Isla Largo Remo West vs Isla Margarita		
F87	0.4	0.8
S87	0.05	0.05
F88	0.2	0.1
Isla Largo Remo North vs Isla Largo Remo West		
F87	0.2	< 0.001**
S87	0.04	0.05
F88	0.5	< 0.001**
Isla Largo Remo North vs Isla Margarita		
F87	0.5	< 0.001**
S87	0.96	0.004*
F88	0.8	< 0.001**

* P<0.01, ** P<0.001

In the next group of comparisons (between sites, within sampling periods), there were no significant differences in the regressions for the reference sites, Isla Largo Remo West and Isla Margarita (Table 4.7). Comparing Isla Largo Remo North, a heavily oiled site, and Isla Largo Remo West, a reference site, we found no significant differences between the slopes of the cavity volume regressions. However, the intercepts of the regressions for February 1987 and February 1988 were significantly different. There were no significant differences between the slopes or intercepts for Isla Largo Remo North and Isla Largo Remo West during September 1987 (Table 4.7). Comparing Isla Largo Remo North to the second reference site, Isla Margarita, we found no significant differences between the slopes of the regressions during each sampling period. However, all of the intercepts of the cavity volume regressions were significantly different (Table 4.7).

Table 4.8 Significance of ANCOVAs of cavity volume vs. stomatopod size for comparisons between post-spill regressions at each site and a pre-spill regression at Isla Margarita. Regression statistics were obtained from the regressions of cavity volume (cc) on the total length (mm) of *Gonodactylus* residents during September (S) and February (F) of the years indicated. Pre-spill data from 1980 were taken throughout the year.

	Slopes Homogeneous	Intercepts Equal
Isla Largo Remo West vs Isla Margarita 1980		
F87	0.7	0.6
S87	0.3	0.06
F88	0.7	0.05
Isla Margarita vs Isla Margarita 1980		
F87	0.2	0.2
S87	0.01	0.2
F88	0.07	0.8
Isla Largo Remo North vs Isla Margarita 1980		
S86	0.6	< 0.001 **
F87	0.04	< 0.001 **
S87	0.004 *	
F88	0.3	< 0.001 **

* P<0.01, ** P<0.001

In the last group of comparisons (pre-spill versus post-spill), there were no significant differences between the cavity volume regression measured at Isla Margarita in 1980 and the cavity volume regressions measured at Isla Margarita and Isla Largo Remo West during the sampling periods after the 1986 oil spill (Table 4.8). In contrast, all of the cavity volume regressions measured at Isla Largo Remo North, a heavily oiled site, were significantly different from the regression measured at Isla Margarita before the oil spill. In September 1987, the slopes were significantly different; the intercepts were significantly different for the other sampling periods (Table 4.8).

Since wounds and injuries tend to be concentrated in larger gonodactylids, we examined the amount of injury among individuals greater than 35 mm total length collected at the study sites from all post-spill samples and those collected at the same sites in years prior to the oil spill (Table 4.9). Injuries were analyzed by first examining the number of individuals with injuries at each site (Table 4.10) and then by examining the level of injuries across sites, factoring in the amount of oiling at the sites (Table 4.11).

Injuries were analyzed within sites by tests of independence in contingency tables of the numbers of injured and not injured (Table 4.9). Comparisons were made to determine differences among pre-spill samples, among post-spill samples, and between pre-spill and post-spill samples. Post-spill samples were also examined for seasonal effects between the wet season (September samples) and the dry season (February samples).

At Isla Margarita, the number of gonodactylids injured and not injured was independent of date sampled among pre-spill samples, among post-spill samples, and between pre-spill and post-spill samples (Table 4.10). The number of injured gonodactylids at Isla Margarita, a lightly oiled to unoiled reference site, was not associated with the date sampled or with the oil spill.

At Isla Largo Remo West, the number injured was not independent of date sampled for pre-spill and post-spill samples (Table 4.10). For post-spill samples, injuries were independent of season, but were associated with the year sampled for each season (Table 4.10). However, injuries were independent of date sampled when comparing pre-spill and post-spill samples. Thus, the number of injured gonodactylids at Isla Largo Remo West, a lightly oiled to unoiled reference site, varied significantly between many of the samples, but the differences in the number injured were not associated with the oil spill.

Table 4.9 Number of injured *Gonodactylus* greater than 35 mm total length. Gonodactylids were collected during September (S) and February (F) of the years indicated. Samples are listed by site, when collected relative to the oil spill (Pre-spill or Post-spill), and season.

	Season	Date	Number Injured	Number Not Injured	Percent Injured
Isla LR-North					
Pre-Spill	Wet	S81	24	94	20
		S86	12	113	10
Post-Spill	Wet	S87	10	62	14
		S87	12	113	10
	Dry	F87	21	112	16
		F88	11	77	13
Isla Mina					
Pre-Spill	Wet	S81	22	41	35
		S83	47	155	23
Post-Spill	Wet	S86	18	142	11
		S87	29	100	22
	Dry	F87	50	141	26
		F88	26	71	27
Isla LR-West					
Pre-Spill	Wet	S81	21	37	36
		S82	27	101	21
Post-Spill	Wet	S86	49	79	38
		S87	21	78	21
	Dry	F87	42	140	23
		F88	47	49	49
Isla Margarita					
Pre-Spill	Wet	S80	17	53	25
		S81	16	35	31
Post-Spill	Wet	S86	42	70	38
		S87	21	61	26
	Dry	F87	27	54	33
		F88	33	49	40

Table 4.10 Significance of log likelihood ratio tests (chi-square) of the numbers of gonodactylids injured and not injured for the samples indicated at each site.

	Isla Margarita	Isla LR-West	Isla LR-North	Isla Mina
Pre-Spill Samples (Wet Season only)	0.4	0.03 *	--	0.07
Post-Spill Samples				
Among all Samples	0.2	< 0.001 ***	0.5	0.001 **
Wet vs Dry Season	0.4	0.8	0.3	0.003 **
Wet Season (S86 vs S87)	0.08	0.005 **	0.4	0.01 *
Dry Season (F87 vs F88)	0.4	< 0.001 ***	0.5	0.9
Pre-Spill vs Post-Spill Samples (Wet Season only)	0.3	0.3	0.03 *	0.005 **

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$, --: only one pre-spill sample.

At Isla Mina, the number of injured gonodactylids was independent of date sampled for pre-spill samples (Table 4.10). For post-spill samples, the number injured was not independent of date sampled and season. The number of injured was independent of date sampled for dry season samples, but not for wet season samples. The number injured also was not independent of date sampled comparing pre-spill and post-spill samples at Isla Mina (Table 4.10). The association between the number injured and date sampled for post-spill samples and between pre-spill and post-spill samples was caused by the low level of injuries found in a single sample, September 1986 (Table 4.9). The oil spill was associated with a decreased number of injured gonodactylids at Isla Mina, a heavily oiled site, in the initial sample following the spill, but the level of injuries returned to pre-spill levels by February 1987.

At Isla Largo Remo North, the number injured was not associated with date sampled for post-spill samples (Table 4.10). Comparing a single wet season, pre-spill sample with the wet season, post-spill samples, the number of injured was not independent of date sampled. The oil spill appeared to be associated with a decrease in the number of injured at Isla Largo Remo North, but this conclusion is tentative. The analysis was hampered by the availability of only one pre-spill sample.

Table 4.11 Significance of log likelihood ratio tests (chi-square) for the frequency of injured gonodactylids, pre- and post-spill and between sites. Reference sites were lightly oiled to unoled. All samples were collected during September, the wet season.

Level of Analysis: within each Site, Pre-Spill vs Post-Spill number injured.	
	<u>P-value</u>
Heavily oiled sites	
Isla Mina	0.005 **
Isla LR-North	0.03 *
Reference Sites	
Isla Margarita	0.3
Isla LR-West	0.3
Level of Analysis: between Sites, Pre-Spill vs Pre-Spill number injured.	
	<u>P-value</u>
Reference Sites	
Isla Margarita vs Isla LR-West	0.8
Reference vs heavily oiled	
Isla Margarita vs Isla Mina	0.8
Isla Margarita vs Isla LR-North	0.2
Isla LR-West vs Isla LR-North	0.3
Isla LR-West vs Isla Mina	0.96
Heavily oiled	
Isla Mina vs Isla LR-North	0.2
Level of Analysis: between Sites, Post-Spill vs Post-Spill number injured.	
	<u>P-value</u>
Reference Sites	
Isla Margarita vs Isla LR-West	0.7
Reference vs heavily oiled	
Isla Margarita vs Isla Mina	< 0.001 ***
Isla Margarita vs Isla LR-North	< 0.001 ***
Isla LR-West vs Isla LR-North	< 0.001 ***
Isla LR-West vs Isla Mina	< 0.001 ***
Heavily oiled	
Isla Mina vs Isla LR-North	0.1

* P<0.05, ** P<0.01, *** P<0.001

Injuries were also analyzed across sites to factor in the amount of oiling at the sites. The analysis was carried out only on wet season data for gonodactylids larger than 35 mm total length (Table 4.9) by a test of independence using a log-linear model in a three-way contingency table (Wilkinson 1986). The main effects in the model were the site, date of sampling relative to the oil spill (pre-spill and post-spill), and the injury state of the gonodactylids (injured and not injured). Sites were not simply pooled into heavily oiled versus reference sites because known differences between the two heavily oiled sites and between the two reference sites would be ignored. The test for a three factor interaction was significant (log likelihood ratio chi-square = 13.23, D.F. = 3, P = 0.004). Thus, the degree of association between pre-spill and post-spill levels of injuries depended on the site. This could be interpreted as an effect of the oil spill if the site differences were consistent with the amount of oiling. However, since it was a three factor interaction, it was also possible that the degree of association between the level of injury and site depended on pre-spill levels of injury. That is, differences among sites were more important than the oil spill.

To examine these possibilities, we followed the recommended procedure to make separate, two-way tests of independence within each level of the main factors (Sokal and Rohlf 1981). This involved two-way tests at each level of site for pre-spill and post-spill injuries, at the pre-spill level of injury examining pair-wise comparisons of sites, and at the post-spill level of injury examining pair-wise comparisons of sites.

The number injured was not associated with pre-spill versus post-spill sampling at the reference sites of Isla Margarita and Isla Largo Remo West (Table 4.11). However, the number injured was associated with pre-spill versus post-spill sampling at both heavily oiled sites (Table 4.11). For pre-spill samples, the number injured was not associated with site for any pair-wise comparisons of sites (Table 4.11). That is, the amount of injury was independent of site in pre-spill samples. For post-spill samples, the number injured was not associated with site for the comparison of reference sites (Isla Margarita versus Isla Largo Remo West, Table 4.11) and the comparison of heavily oiled sites (Isla Mina versus Isla Largo Remo North). In contrast, the number injured was associated with site for all pair-wise comparisons of reference versus heavily oiled sites (all $P < 0.001$). These results indicate that the number of injured gonodactylids was associated with the amount of oiling at the sites rather than inherent differences among the sites. Heavily oiled sites had fewer injured gonodactylids after the oil spill than reference sites.

Physical characteristics of the habitat were recorded from quadrat samples taken at each site. It is important to remember that quadrat areas were selected in areas we had sampled prior to the oil spill and that these areas were primary habitat (i.e., intertidal beds of turtle grass) for gonodactylids prior to the oil spill. Based on that point, we divided the physical characteristics found in the quadrats into three categories:

1. *Thalassia* Blades. Quadrats in this category were located in lush stands of *Thalassia* where blades of the turtle grass were abundant throughout the quadrat.
2. *Thalassia* Rhizomes. Quadrats in this category contained no turtle grass blades above the sandy bottom, but the sand contained turtle grass roots and rhizomes.
3. Coral Bench. Hard substratum in the form of dead coral bench covered at least 10% of the quadrat.

The data were analyzed within sites by a test of independence using a log-linear model for a 4 x 3 contingency table. The main effects entered in the model were the date sampled (four post-spill samples) and the number of quadrats with the physical characteristics described above. The physical characteristics of the habitat were associated with the date sampled at Isla Largo Remo North during post-spill sampling (Table 4.12). None of the 120 quadrat samples taken at Isla Largo Remo North contained live blades of *Thalassia*. All samples contained roots and rhizomes in the sand, evidence of the previous *Thalassia* bed that had existed at the site; all these roots and rhizomes were dead. By February 1988, the matrix of dead roots and rhizomes began to break up and the soft substratum had started to wash away. This exposed the underlying hard substrata in 30% of the quadrats at Isla Largo Remo North. The other sites showed no association between the date sampled and characteristics of the habitats (all $P > 0.8$). These sites, including both reference sites and Isla Mina, a heavily oiled site, continued as thriving beds of turtle grass with widely scattered patches of bench and sand through February 1988.

We also recorded oil observed at the sites. At Isla Largo Remo North, an oil sheen appeared on the surface of the water whenever we walked through the area during each sampling from September 1986 - February 1988. Oil was also found in some of the coral rubble at Isla Largo Remo North during every visit. At Isla Mina, we observed oil released from the sediments during September 1986 - September 1987, but not in February 1988. Oil was found in rubble at Isla Mina in September 1986 and February 1987, but not in September 1987 and February 1988. We did not see oil released from the sediments and we did not find oil in the coral rubble at Isla Largo Remo West and Isla Margarita. These post-spill observations reaffirm the original classifications of heavily oiled and reference sites and support data implying that Isla Largo Remo North was oiled more heavily than Isla Mina.

Table 4.12 Physical characteristics of the habitats. Data (number of quadrats) were recorded from 30 quadrats of 0.5 m² at each site during September (S) and February (F) of the years indicated. Reference sites were lightly oiled to unoiled.

		<i>Thalassia</i> Blades	<i>Thalassia</i> Rhizomes	Coral Bench
Heavy Oiled Sites				
Isla LR-North *	S86	0	29	1
	F87	0	29	1
	S87	0	30	0
	F88	0	21	9
Isla Mina	S86	30	0	0
	F87	28	2	0
	S87	27	3	0
	F88	29	1	0
Reference Sites				
Isla Margarita	S86	30	0	0
	F87	29	0	1
	S87	30	0	0
	F88	30	0	0
Isla LR-West	S86	30	0	0
	F87	30	0	0
	S87	30	0	0
	F88	30	0	0

* Log-Likelihood Ratio Chi-square = 15.88, D.F.=6, P=0.014.

4.4 Discussion

The densities of *Gonodactylus* have changed in a complex pattern between September 1986 and February 1988 following the oil spill. The total density of gonodactylids has decreased significantly at one of two reference sites and at one of two heavily oiled sites (Table 4.1). The variation in total gonodactylid densities following the oil spill, therefore, cannot be interpreted as a simple effect of the amount of oiling.

The changes in gonodactylid densities have been driven by a steady decline in the abundance of medium-sized individuals, 16-40 mm in total length. Medium-sized individuals decreased significantly at Isla Margarita and Isla Largo Remo North (Table 4.3), the reference and heavily oiled sites where total densities declined. Even at sites where no significant change in total density occurred, there was a steady trend of declining density in medium-sized individuals.

In addition to the post-spill data given here, mean gonodactylid densities from quadrat samples taken at Isla Margarita prior to the oil spill in 1979 and 1982 are available (Steger 1985, 1987). Although the raw data from these samples have not yet been compiled with the post-spill data for statistical analysis, the published means are useful for a tentative and qualitative discussion (Table 4.13).

The most striking difference between the mean densities measured at Isla Margarita before the oil spill in 1979 and 1982 and the densities measured after the oil spill is the general low level of postlarval recruitment (small gonodactylids) at all sites after the oil spill (Table 4.4). In dry season samples, comparing March-April 1982 at Isla Margarita with February 1987 and 1988 at all sites, the density of recent recruits (6-15 mm total length) in 1982 at Isla Margarita was about 1.5 times the density at Isla Margarita in 1987 and 1988, 2 to 6 times the density at Isla Mina, 3 to 6 times the density at Isla Largo Remo West, and 25 to 60 times the density at Isla Largo Remo North. In wet season samples, the differences were even greater.

Table 4.13 Mean densities of *Gonodactylus* per 0.5 m² (from Steger 1985,1987). Means are for 100 quadrats of 1 m² taken at Isla Margarita in July-August, 1979 and from 20 quadrats of 0.5 m² taken at Isla Margarita in March-April of 1982.

	July-August 1979	March-April 1982
Large (> 40 mm total length)	0.35	0.35
Medium (16-40 mm total length)	3.52	1.80
Small (6-15 mm total length)	0.95	1.75
Total Density (All sizes)	4.82	3.90

The decline in gonodactylid densities after the oil spill, therefore, appears to be caused by low levels of postlarval recruitment, which led to declines in the abundances of medium-sized individuals. The number of gonodactylids recruiting at two of the sites has not been sufficient to maintain populations at 1986 levels and, unless recruitment increases, the other sites may also show declines. The low recruitment is especially noteworthy in February samples. Ordinarily, large numbers of larvae settle on the reefs around Punta Galeta from January through June every year, while recruitment during the rest of the year is lower and highly variable from year to year (Steger 1987). The abundances of large gonodactylids, which are all over two years old, have not been affected by the lack of recruitment.

Since recruitment is apparently low at both heavily oiled and lightly oiled to unoiled reference sites, oiling may not have caused the reduction. However, there are two points worth noting. First, the initial post-spill survey showed that the abundances of large females were reduced greatly by the oil spill. Large females normally produce vast numbers of larvae compared to smaller females. Second, the level of recruitment in post-spill samples increased away from Bahía Las Minas, the origin of the oil spill. This includes the case of Isla Largo Remo West where the existing populations were not affected directly by the oil spill. Nevertheless, the site is surrounded by heavily oiled areas and recruitment levels appear to be very low. During five years of studies prior to the oil spill, we never witnessed such a general lack of postlarval recruitment. Considering the large number of reefs that were oiled, it is possible that the larval pool of gonodactylids in the area of Bahía Las Minas has been reduced by the mortality of large females. Alternatively, low levels of chronic oiling may be inhibiting the return of larvae. However, until we identify the mechanism behind the reduced recruitment, it remains possible that this is a natural, albeit unusual, phenomenon triggered by events unrelated to the oil spill.

The intensity of competition for cavities is expected to be a function of the abundance of individuals relative to the abundance of cavities. A good indicator of the intensity of competition is the relationship between the size of a gonodactylid and the size of its cavity. When cavities are limiting the populations, the intensity of competition for cavities is high and individuals are forced to occupy cavities smaller than they prefer (Steger 1985). This leads to the prediction that as densities of gonodactylids change, the cavity volume relationship should change also.

At Isla Largo Remo North and Isla Margarita, changes in densities of gonodactylids have occurred in a complex pattern since 1986. Densities of large individuals have not changed (Table 4.2) while densities of medium-sized animals have decreased (Tables 4.3). Cavity volumes were not measured for any individuals smaller than 18 mm total length. The changes in densities appear to have caused the relationship between cavity volume and size of resident to change at these sites. However, both slopes and intercepts have varied significantly and no simple pattern has emerged. A thorough analysis of changes in cavity use patterns as they are related to shifts in densities has not yet been completed. We hope that such an analysis will better show how shifts in the relative densities of different sizes of individuals affect overall patterns of competition.

At Isla Largo Remo North, Isla Margarita, and Isla Mina, the number of injured among large gonodactylids was closely associated with density in post-spill samples. At Isla Largo Remo North and Isla Margarita, there were no significant changes in the density of large gonodactylids (Table 4.2) and there were no significant changes in the number injured (Table 4.9).

Events at Isla Mina following the oil spill have been especially interesting. In September 1986, there were few large individuals, especially females, at Isla Mina (Jackson *et al.* 1989). In fact, we found no gonodactylids larger than 40 mm total length in the quadrat samples. In February 1987, the density of large *Gonodactylus* increased to 0.5 individuals per 0.5 m² and has remained unchanged through February 1988 (Table 4.2). Since these individuals are over two years old, they must have migrated into the site from nearby areas. The number of large gonodactylids carrying injuries mirrored the changes in density completely. As density increased, the number of injured increased. Unfortunately, the total picture of competition and the effects of density is incomplete since this was the one site where we chose not to measure cavity volumes due to time constraints.

At Isla Largo Remo West, there was no agreement between shifts in the number injured and densities. The density of large gonodactylids did not vary significantly (Table 4.2) while the number of injured varied greatly from sample to sample (Table 4.9). In fact, the density of large gonodactylids at Isla Largo Remo West showed the least variation among samples for any site, while the number of injured ranged between 21% and 49%, the greatest variation in injuries for any site. However, the intensity of competition, as indicated by the cavity volume relationship, has not changed significantly (Table 4.6). Isla Largo Remo West was also the only site to show significant variation in the number injured among samples taken prior to the oil spill (Table 4.10). There is no easy explanation for the fluctuation in the number of injured gonodactylids at this site.

The discussion of densities, competition, and injuries has focused on events at each site up to this point. Comparisons among sites have also shown that the amount of oiling had a significant effect overall by lowering the number of injured gonodactylids found at heavily oiled sites. A thorough discussion of this effect, however, must be postponed until more samples taken prior to the oil spill are prepared for analysis.

Comparisons among sites and with data collected at Isla Margarita prior to the oil spill also indicated that the cavity utilization pattern was significantly different at Isla Largo Remo North, the heavily oiled site (Tables 4.7 and 4.8). Gonodactylids occupied larger cavities at Isla Largo Remo North than at Isla Margarita and Isla Largo Remo West. Given that densities and the number injured were also lower, it appears that competition for cavities is less intense at Isla Largo Remo North, a heavily oiled site, than at the reference sites. This appears to be an effect of the oil spill, but further analysis of the data will be necessary to conclude decisively.

It is important to note that our discussion has concentrated on large individuals. We have done this because the initial survey indicated that the major impact of the oil spill was on this size class. This report establishes that many

important changes are now affecting smaller individuals. Although we are expanding our analyses to include the cavity use patterns, wounds, and growth of all sizes of individuals, those analyses are not yet complete.

The deterioration of the habitat at Largo Remo North is an extremely important event. The habitat at Largo Remo North is changing dramatically as the former *Thalassia* bed is breaking down (Table 4.12). The soft sediment that was stabilized by the root and rhizome system is being washed away. If this pattern continues, the habitat will be lost as primary habitat for gonodactylid stomatopods and their associated community. The site is currently shifting from an intertidal *Thalassia* bed to an area of reef bench and unconsolidated chunks of coral that had previously been buried. After the soft sediments are gone, there will be little chance in the near future for this site to "recover" to anything that resembles the area prior to the oil spill.

It appears that Isla Mina was not as heavily oiled or, at least, not as heavily affected by the oil as Isla Largo Remo North. The *Thalassia* bed at Isla Mina was not killed by the oil spill and there has been no overall deterioration of the habitat. In addition, large gonodactylids have migrated into the site (Table 4.2). Except for a few patches of dead mangroves along the shore, Isla Mina appears today much as it did before the oil spill.

In summary, three significant patterns are apparent. First, stomatopod densities have been declining at all four sites, apparently due to low levels of postlarval recruitment. Local populations cannot be maintained unless recruitment increases. Second, with the exception of one site, competition for cavities and the levels of injuries associated with competition for cavities remained associated with stomatopod densities. Third, the habitat at Isla Largo Remo North, a heavily oiled site, has been changing dramatically since the oil spill killed the *Thalassia* bed there.

4.5 Future Research and Analyses

We plan to continue the sampling regime as presented here. It is especially critical to continue to monitor the densities of individuals and follow the pattern of recruitment because these sites are in such an unusual state of flux, compared with earlier studies. The fate of these populations is uncertain, given the relatively low post-spill recruitment measured so far.

Our ability to analyze the data has been greatly enhanced because a computer has just been acquired in French Polynesia. We plan to assemble all data from before the oil spill and analyze in greater detail the possible decrease in recruitment and changes in densities. Furthermore, as indicated above, the pattern of competition and shifts in competition associated with changes in densities will be examined by size class. The compilation and analysis of growth data and density data from rock samples should provide a much clearer view of the long-term impact of the oil spill on gonodactylids.

We plan to expand our samples of coral rubble to include areas that are outside Bahía Las Minas. This is necessary to determine whether the oil spill has

had an effect on levels on postlarval recruitment or a much larger scale depression has occurred. The reefs around Portobelo may be suitable for additional sites and will be sampled as much as possible.

Chapter 5 Subtidal Reef Corals

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5.1 Introduction

The extent of the effects of oil on reef corals in their natural environment is controversial and poorly understood (Loya and Rinkevich 1980; Brown and Howard 1985). On one hand, a few field studies suggest harmful effects and lasting damage in oiled populations as measured by abundance, mortality, reproduction, and recruitment (Loya 1976; Loya and Rinkevich 1980). Field studies supporting this view are for intertidal corals or lack observations before pollution began. On the other hand, results of experiments on the effects of oil and dispersants in the laboratory or field contradict this view and suggest little or no mortality or persistent sub-lethal effects of oil on corals (Dodge *et al.* 1984; Knap 1987).

Subtidal reefs along the Caribbean coast of Panama are mostly fringing reefs that give way to plains of sediments at a depth of 10 to 25 m. Despite heavy sedimentation and runoff, coverage of living organisms is high, averaging about 83% (Weil and Jackson, unpublished data). The most abundant organisms are macroalgae (45%), scleractinian corals (27%), sponges (5%), and crustose algae (3%). The most abundant coral species are the scleractinians *Siderastrea siderea*, *Porites astreoides*, *Diploria clivosa*, *Agaricia agaricites* and *D. strigosa*, and the hydrocoral *Millepora complanata*. Other, more patchily abundant, species are *Agaricia tenuifolia*, *Montastrea annularis*, *Acropora palmata* and *Porites* spp. All these species are widely distributed and occur in similar habitats at shallow depths (Weil and Jackson, unpublished data; Guzmán *et al.* in press). Because of this, and the likely greater effects of the spill in shallow water, all work to examine the effects of oil on corals has been done in shallow areas (less than 10 m).

The amount of oil overlying the reefs was visually assessed during the three months following the spill by air, boat, and underwater observations. Reef sites were ranked as heavily oiled (reefs 2-7), moderately to lightly oiled (reefs 1 and 8), or unoiled (reefs 9-12) (Figure 5.1). Hydrocarbon concentrations in tissues of the corals *Siderastrea siderea* and *Agaricia tenuifolia* and in the reef sediments correspond well to the visual classification of the degree of oiling that was used to define the "control" and the "polluted" reefs (Jackson *et al.* 1989; Burns and Knap 1989). We have also continued to record the presence or absence of oil slicks every time the reefs are visited. These observations have suggested that oil pollution is chronic throughout much of Bahía Las Minas where partially degraded oil from the 1986 spill continues

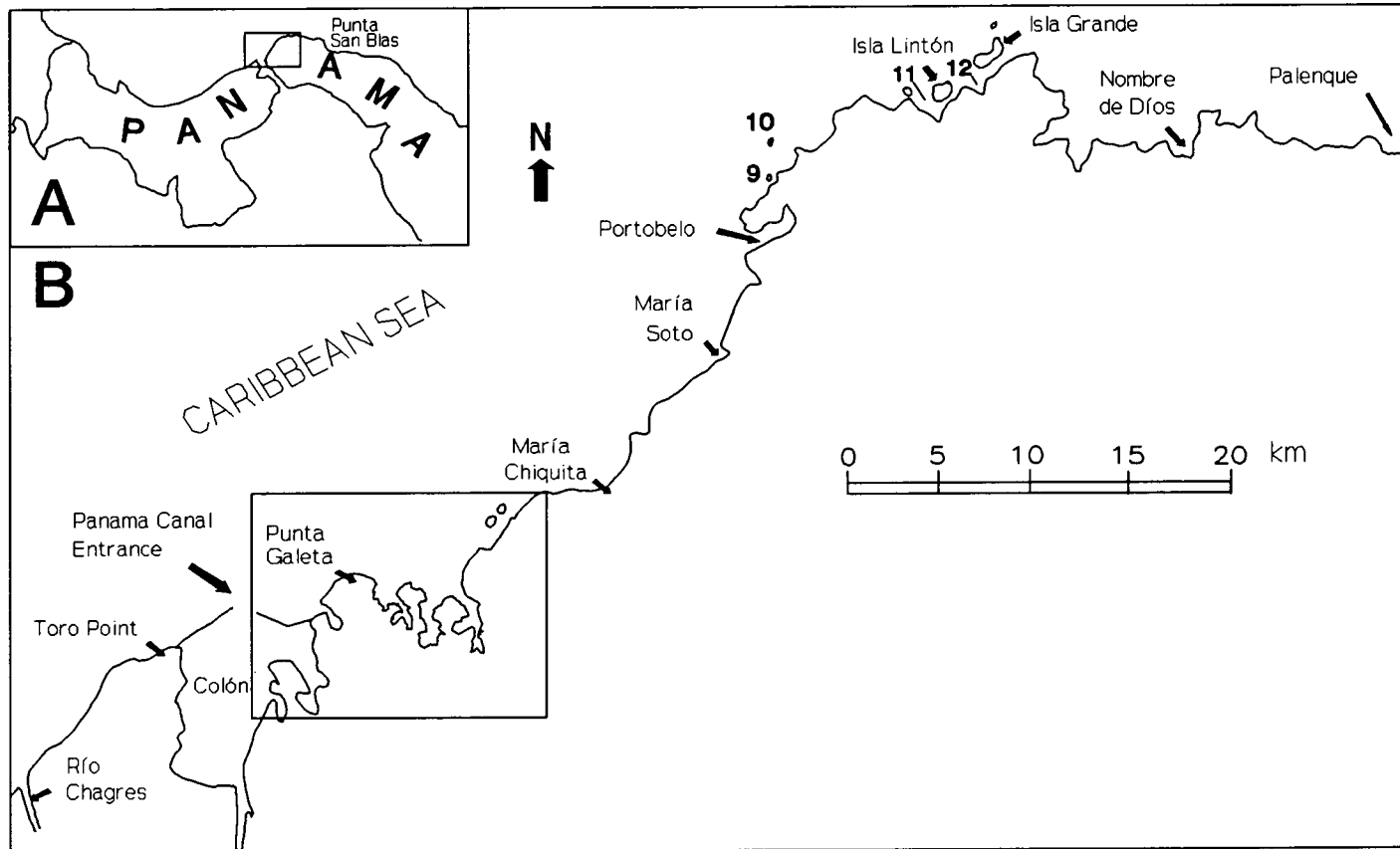


Figure 5.1 Map of the Republic of Panama showing the 12 reefs and the refinery. The boxed area in (B) includes the oiled reefs shown in greater detail in (C). Unoiled "control" reefs (9-12) are northeast of the oiled reefs. Refinery marked as encircled "R." See Figure 1.1 for further details.

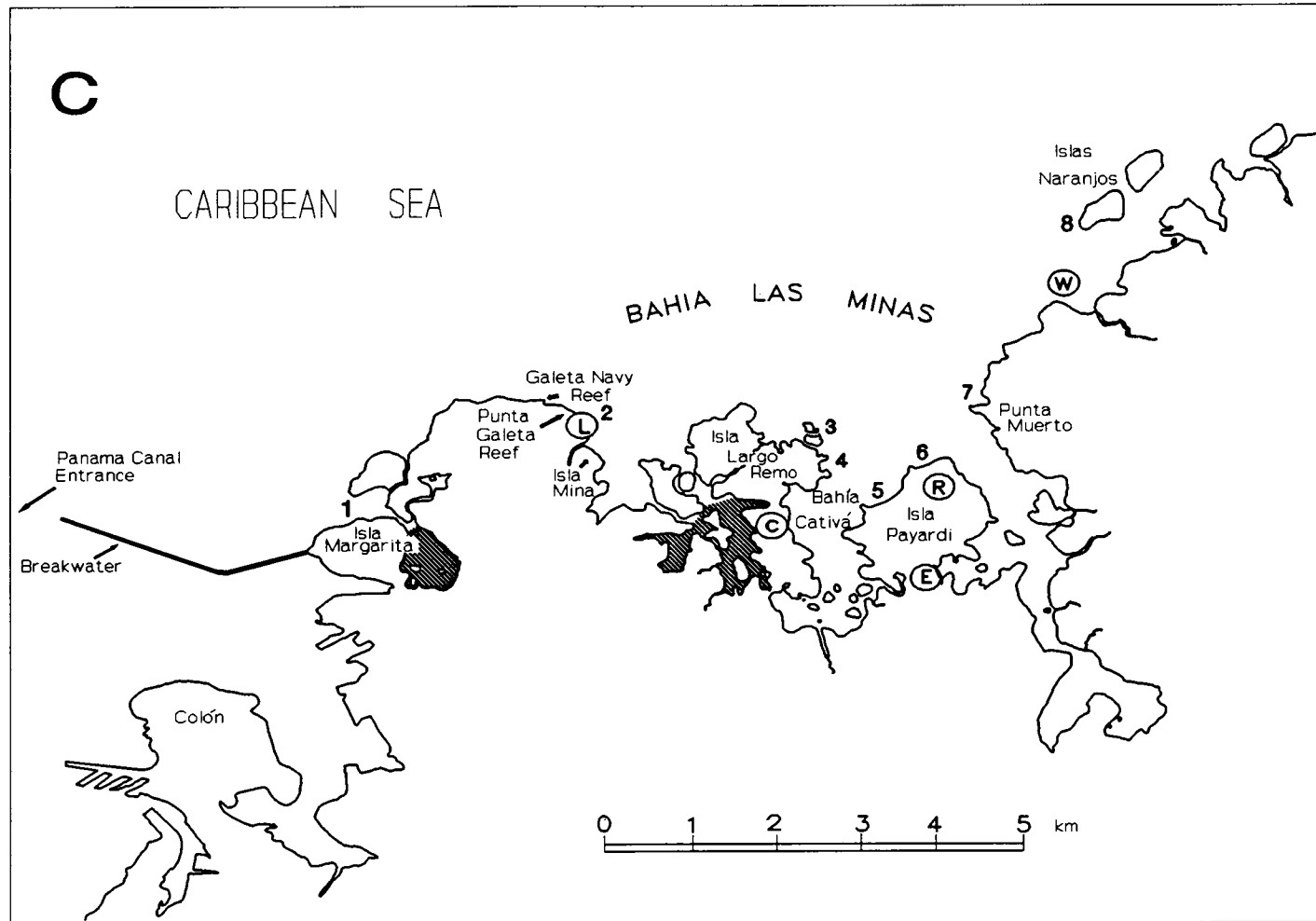


Figure 5.1 Map of the Republic of Panama showing the 12 reefs and the refinery (continued).

to be released from mangrove sediments and landfill, especially after heavy rains (Table 5.1). According to the Instituto de Recursos Hidráulicos y Electrificación (IRHE) and refinery personnel, during December 1988 an unestimated amount of diesel spilled into the Bahía Las Minas area from a storage tank at the IRHE electrical generating station (Figure 5.1, encircled "R"), about 1 km from the refinery (S. Garrity, pers. comm.). Three of our study sites (reefs 3, 4, and 5) may have been affected by this additional spill.

5.2 Tasks

This study included surveys of coverage of all sessile organisms, periodic monitoring of four coral species that showed much recent injury just after the spill, and assays of physiological state (regeneration of injuries and rates of growth) of two common coral species. Temperature, salinity, suspended sediments, and resuspended sediments were also measured at all sites.

Different tasks were, of necessity, performed on different subsets of reefs for reasons of availability of corals, logistics, etc. These are listed in Table 5.2.

Table 5.1 Percent days when oil slicks were observed. Numbers of observation days are in parentheses. The periods are August-December 1987, 1988, and January-August 1989. Reefs are listed from east to west (see Figure 5.1 for details).

No.	Reef		Percent days with oil slicks ^a		
	Acronym ^b	Oiling	1987	1988	1989
12	JUG	None	25(4) ^c	0(17)	0(13)
11	PALW	None	0(4)	0(16)	0(12)
10	DMA	None	0(6)	0(15)	0(12)
9	DONR	None	0(4)	0(14)	0(13)
8	NARS	Moderate	9(12)	32(25)	5(19)
7	PM	Heavy	30(10)	65(17)	32(19)
6	PAYN	Heavy	60(10)	89(38)	67(24)
5	PAYW	Heavy	25(4)	73(11)	74(27)
4	LRE1	Heavy	17(6)	100(7)	90(10)
3	LRE2	Heavy	23(9)	86(36)	63(27)
2	GALC	Heavy	80(10)	80(20)	30(20)
1	MAR3	Moderate	0(2)	0(7)	0(13)

^a This includes only slicks passing over the reefs. Oil is commonly observed every day, the entire year, in different areas in Bahía Las Minas.

^b Site acronyms are listed in the preface.

^c A small gasoline slick was observed probably from an outboard engine.

Table 5.2 Subtidal reefs studied for the different tasks. (COV=percent cover of sessile organisms; INJ=coral injury; REG=regeneration experiments; SCL=sclerochronology; MON=physical monitoring; HYD1=hydrocarbon samples, 1986; HYD2=hydrocarbon samples, 1988-89 and 1990).

Reef		Tasks							
No.	Acronym	Oiling	COV	INJ	REG	SCL	MON	HYD1 ^a	HYD2 ^b
1	MAR3	Moderate	X	X		X	X		X
2	GALC	Heavy	X	X		X	X	X	X
3	LRE1	Heavy	X	X					X
4	LRE2	Heavy	X	X	X	X	X		X
5	PAYW	Heavy	X	X	X	X	X		X
6	PAYN	Heavy	X	X		X	X	X	X
7	PM	Heavy	X	X		X	X		X
8	NARS	Moderate	X	X		X	X	X	X
9	DONR	None	X	X		X	X		X
10	DMA	None	X	X		X	X		X
11	PALW	None	X	X	X	X	X	X	X
12	JUG	None	X	X	X	X	X		X

^a Reefs sampled by BBSR for hydrocarbons in 1986.

^b *Siderastrea siderea*, *Porites astreoides* and reef sediments were collected at all reefs.

Acropora palmata, *Agaricia tenuifolia* and *Diploria strigosa* collected only at PAYN and PALW reefs.

5.2.1 Coverage of Sessile Organisms

Annual surveys are made to measure the percent cover of different dominant groups of organisms (fleshy algae, sponges, corals, etc.) and of species of scleractinian corals at different depths on each of the twelve reefs under study (Table 5.2).

Methods

Surveys of reefs for coral cover, abundance, size, and diversity were done using 1 m² quadrats in order to use the same methods that had been employed in a survey of reefs before the oil spill (Weil and Jackson, unpubl. data). Six reefs within the study area had been surveyed between July and October 1985, including four that were subsequently unoiled (reefs 9-12), one that was lightly to moderately oiled (reef 1), and one that was heavily oiled (reef 2; Table 5.2). These reefs were surveyed again after the spill during July and August 1986 and in May and June 1988.

At each reef, four or five line transects were extended perpendicularly to the shore from haphazardly chosen points at the shoreward edge of the reef to the bottom of the reef. Where a reef flat occurred (see Chapter 2), the transects were extended from the seaward edge of the reef flat. Along each line, 1 m² quadrats

were placed contiguously or spaced at regular intervals up to 3 m apart, depending on the length of the transect and profile of the reef. All quadrats were placed subtidally and were not exposed, even during seasonal periods of extreme low water. The quadrats were divided by strings into one-hundred 100-cm² cells; coral size (area of live tissue) was estimated visually using these cells. For purposes of the analyses, quadrats were pooled into two depth intervals: 0.5-3 m (shallow) and >3-6 m (deep). The typical number of quadrats per reef was 120.

Data analysis for coral cover, number and sizes of colonies, numbers of species, and Shannon-Wiener diversity H' (based on percent cover of corals and on number of coral colonies) was done using single mean values for each depth interval on each reef. All percentage data were transformed using the arcsin function. Differences in these variables between the 1985 and 1986 censuses were compared in order to assess the immediate effects of the oil spill. These analyses were done by calculating the natural logarithm of the value for 1985 divided by the value for 1986, i.e., $\ln(\text{mean cover } 1985/\text{mean cover } 1986)$, and testing for the effect of oil by analysis of variance (ANOVA) with one heavily oiled, one lightly oiled, and four unoiled reefs.

Variation in the same variables (cover, number and sizes of colonies, and diversity) over all three censuses (1985, 1986, 1988) were evaluated by repeated measures MANOVA (BMDP Programs 2V and 4V, 1987) with the amount of oil and depth as between-factor variables and time (year of census) a within-factor variable. A repeated measures design was necessary because the same reefs were repeatedly sampled (Underwood 1981).

Results

The data from the six reefs that were surveyed both before and after the oil spill (pre-spill: October 1985; post-spill: August 1986 and May-October 1988) are presented here. Differences between 1985 and 1986, 3 months after the oil spill, revealed extensive effects of oil (Table 5.3; Figures 5.2 and 5.3). Total coral cover decreased by 76% at 0.5-3 m and by 56% between >3-6 m on the heavily oiled Punta Galeta reef, decreased less on the moderately oiled reef at Isla Margarita, and generally increased or stayed the same on the four unoiled reefs (2-way ANOVA, $F=11.7$, d.f.=3,8, $P=.004$). *Acropora palmata* was restricted almost entirely to depths <3 m where it was typically the most abundant coral before the oil spill. This species was practically eliminated from the heavily oiled Punta Galeta reef, whereas its abundance increased by an average of 38% on the four unoiled reefs during the same year (Table 5.3; 1-way ANOVA, $F=21.0$, d.f.=2,3, $P=.017$). No other species showed a significant reduction on the oiled reefs except marginally for *Porites astreoides* (2-way ANOVA, $F=4.14$, d.f.=3,8, $P=.058$). However, there was a general decrease in cover for almost all the species between 0.5-3 m depth (Table 5.3).

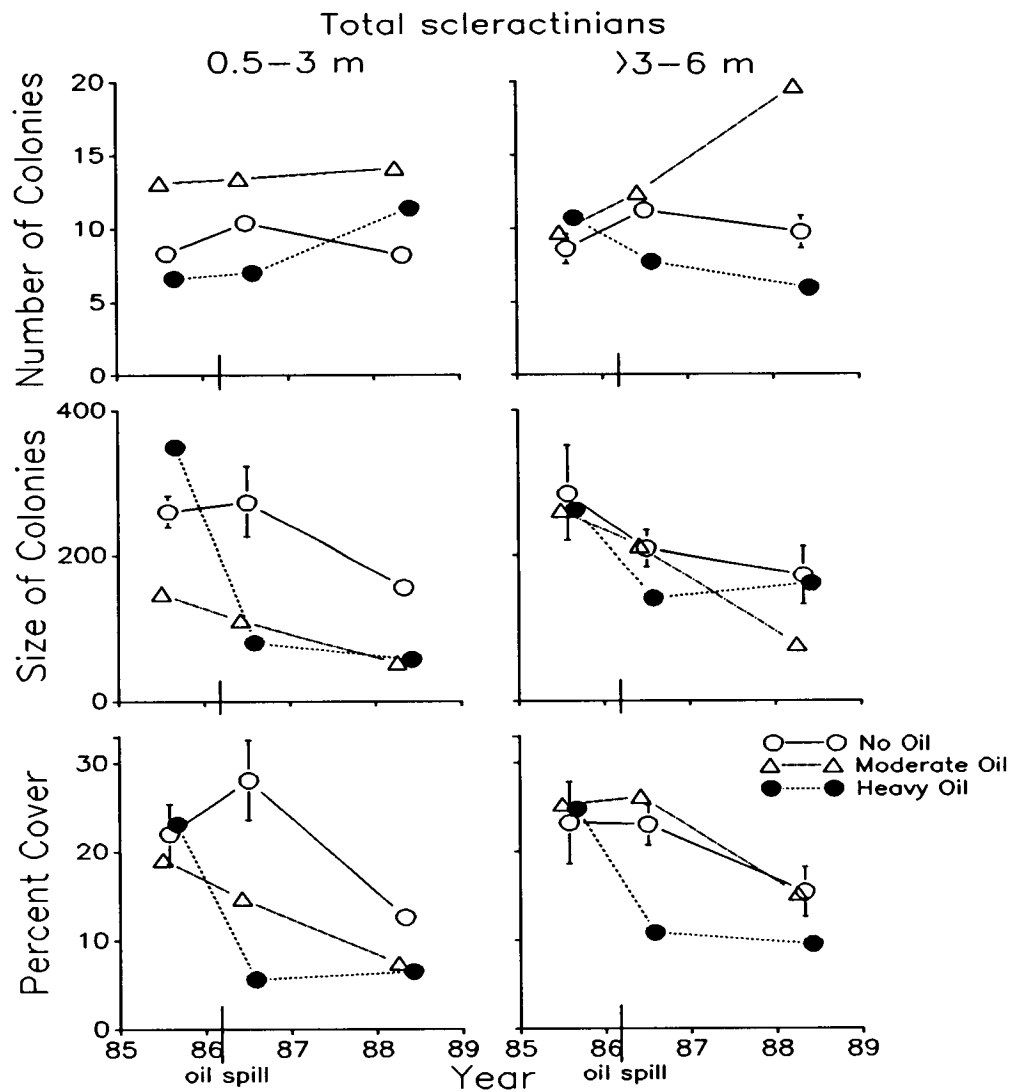


Figure 5.2 Changes in mean numbers and sizes of coral colonies per m² and percent cover for all coral species combined in relation to amount of oil and depth. Time of oil spill indicated by vertical bar. Standard errors shown for the four unoled reefs except when smaller than the symbols used. There is no error value for the two types of oiled reefs as they are each represented by only a single reef.

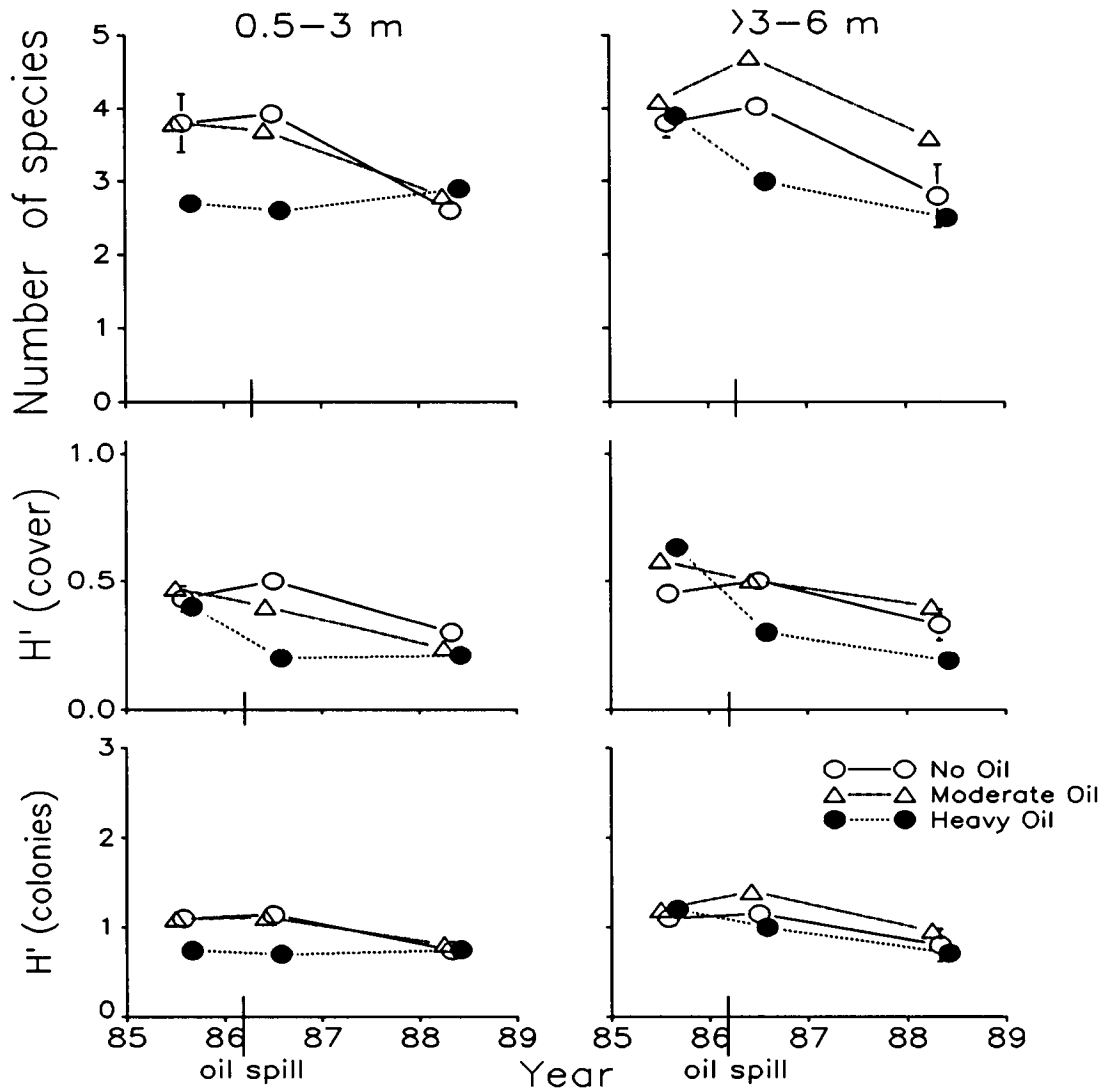


Figure 5.3 Changes in average numbers of coral species per m^2 and Shannon-Wiener diversity per m^2 in relation to amount of oil and depth. Time of oil spill indicated by vertical bar.

Table 5.3 Percent cover of common coral species, total coral cover, and number of coral species by depth, reef, and date of census. Data are means and standard errors in parentheses, except for number of species. Blanks in table are species not observed.

Species	Depth (m)	Year	Number of Reef					
			1	2	9	10	11	12
<i>Acropora cervicornis</i>	0.5-3	85						1.29 (0.20)
		86						1.86 (0.27)
		88						0.28 (0.03)
	>3-6	85						3.26 (0.41)
		86						3.19 (0.45)
		88				0.03 (0.00)		0.67 (0.11)
<i>Acropora palmata</i>	0.5-3	85	0.59 (0.07)	6.56 (0.76)	4.86 (0.72)	4.75 (1.68)	7.18 (1.36)	10.4 (1.65)
		86	0.01 (0.00)	0.26 (0.04)	8.04 (1.07)	8.38 (2.03)	4.54 (0.91)	15.1 (2.21)
		88	0.03 (0.00)		3.72 (0.47)	0.16 (0.02)	0.18 (0.02)	5.00 (0.55)
	>3-6	85			2.02 (0.27)		1.87 (0.28)	
		86			1.89 (0.26)	5.33 (0.97)	1.72 (0.26)	2.22 (0.31)
		88			0.57 (0.07)	0.51 (0.09)		
<i>Agaricia agaricites</i>	0.5-3	85	0.17 (0.02)	0.12 (0.01)	0.20 (0.03)	0.09 (0.03)	0.08 (0.01)	0.43 (0.07)
		86	0.28 (0.03)	0.02 (0.00)	0.17 (0.02)	0.09 (0.02)	0.23 (0.05)	0.69 (0.10)
		88	0.29 (0.03)	0.27 (0.03)	0.04 (0.00)	0.12 (0.01)	0.04 (0.01)	0.11 (0.01)
	>3-6	85	0.00 (0.00)	0.15 (0.02)	0.41 (0.05)	0.19 (0.06)	0.04 (0.01)	0.58 (0.07)
		86	0.64 (0.12)	0.00 (0.00)	0.12 (0.02)	0.20 (0.04)	0.06 (0.01)	0.21 (0.03)
		88	0.22 (0.05)	0.04 (0.01)	0.10 (0.01)	0.00 (0.00)	0.15 (0.03)	0.08 (0.01)
<i>Agaricia tenuifolia</i>	0.5-3	85						0.43 (0.07)
		86	0.04 (0.00)	0.20 (0.03)	1.79 (0.24)	0.28 (0.07)		0.12 (0.02)
		88	0.49 (0.05)				0.02 (0.00)	0.19 (0.02)
	>3-6	85		0.02 (0.00)	0.13 (0.02)		0.02 (0.00)	3.46 (0.44)
		86	1.49 (0.29)	0.56 (0.10)	1.15 (0.16)	0.23 (0.04)	0.43 (0.07)	6.66 (0.93)
		88	1.81 (0.44)	0.10 (0.02)	0.03 (0.00)	0.34 (0.06)		7.56 (1.26)
<i>Colpophyllia natans</i>	0.5-3	85		0.04 (0.00)				0.19 (0.03)
		86						0.43 (0.06)
		88						
	>3-6	85		0.65 (0.10)	0.14 (0.02)	1.14 (0.34)	0.03(0.01)	0.75 (0.10)
		86	4.36 (0.84)	0.12 (0.02)	2.40 (0.33)	0.61 (0.11)	0.71 (0.11)	0.32 (0.04)
		88	0.24 (0.06)		0.04 (0.01)	0.29 (0.05)	1.32 (0.28)	0.14 (0.02)

Table 5.3 Percent cover of common coral species, total coral cover, and number of coral species by depth, reef, and date of census (continued).

Species	Depth (m)	Year	Number of Reef					
			1	2	9	10	11	12
<i>Diploria clivosa</i>	0.5-3	85	0.72 (0.08)	4.96 (0.58)	3.50 (0.52)	0.44 (0.15)	1.90 (0.36)	0.50 (0.08)
		86	1.55 (0.18)	0.95 (0.15)	2.54 (0.34)	1.75 (0.43)	4.34 (0.87)	0.57 (0.08)
		88	0.02 (0.00)	1.48 (0.15)	0.15 (0.02)	0.89 (0.10)	2.81 (0.34)	0.04 (0.00)
	>3-6	85					0.10 (0.01)	0.13 (0.02)
		86		0.67 (0.12)	0.16 (0.02)	0.15 (0.03)	0.24 (0.04)	0.05 (0.01)
		88			0.03 (0.00)	0.46 (0.08)		
<i>Diploria strigosa</i>	0.5-3	85	0.04 (0.00)	3.39 (0.39)	3.63 (0.54)	1.69 (0.60)	0.79 (0.15)	1.04 (0.16)
		86	0.04 (0.01)	0.46 (0.07)	2.21 (0.30)	0.99 (0.24)	0.71 (0.14)	0.39 (0.06)
		88		0.38 (0.04)	1.85 (0.23)	2.08 (0.23)	1.51 (0.18)	0.54 (0.06)
	>3-6	85		0.50 (0.07)	2.81 (0.37)	0.20 (0.06)	0.47 (0.07)	0.20 (0.02)
		86	0.35 (0.07)		0.40 (0.06)	0.47 (0.09)	1.51 (0.23)	0.15 (0.02)
		88		0.12 (0.02)	2.07 (0.27)	0.98 (0.17)	1.27 (0.27)	0.04 (0.01)
<i>Montastrea annularis</i>	0.5-3	85			2.24 (0.33)			7.52 (1.19)
		86			1.58 (0.21)	0.66 (0.16)	0.50 (0.10)	10.59 (1.55)
		88			0.37 (0.05)	0.04 (0.00)		3.31 (0.37)
	>3-6	85		0.01 (0.00)	12.66 (1.66)	0.07 (0.02)	0.33 (0.05)	3.91 (0.50)
		86			8.12 (1.12)	3.44 (0.63)	1.81 (0.28)	0.77 (0.11)
		88			1.65 (0.22)	0.04 (0.01)	1.16 (0.25)	
<i>Porites astreoides</i>	0.5-3	85	3.45 (0.38)	3.96 (0.46)	3.13 (0.46)	4.19 (1.48)	1.09 (0.21)	0.78 (0.12)
		86	2.71 (0.31)	1.39 (0.22)	2.12 (0.28)	2.87 (0.70)	3.32 (0.66)	1.66 (0.24)
		88	1.79 (0.18)	2.07 (0.20)	2.41 (0.31)	1.70 (0.18)	1.27 (0.15)	1.34 (0.15)
	>3-6	85	1.35 (0.27)	3.09 (0.46)	2.13 (0.28)	2.00 (0.60)	1.03 (0.15)	0.78 (0.10)
		86	0.77 (0.15)	0.90 (0.17)	1.34 (0.18)	1.90 (0.35)	1.53 (0.23)	1.09 (0.15)
		88	0.04 (0.01)	0.42 (0.08)	2.67 (0.35)	1.79 (0.30)	0.41 (0.09)	0.14 (0.02)
<i>Porites furcata</i>	0.5-3	85	2.27 (0.25)	0.11 (0.01)		0.24 (0.09)	0.19 (0.04)	0.01 (0.00)
		86	1.59 (0.18)	0.24 (0.04)	0.09 (0.01)		0.36 (0.07)	0.27 (0.04)
		88	0.66 (0.07)	0.46 (0.05)	0.21 (0.03)	0.08 (0.01)	0.06 (0.01)	0.04 (0.00)
	>3-6	85	7.28 (1.46)	4.30 (0.63)		0.19 (0.06)	0.11 (0.02)	0.22 (0.03)
		86	2.36 (0.45)	2.13 (0.40)	0.14 (0.02)		0.64 (0.10)	0.16 (0.02)
		88	2.35 (0.57)	1.17 (0.21)	0.32 (0.04)	0.21 (0.04)	0.08 (0.02)	0.30 (0.05)

Table 5.3 Percent cover of common coral species, total coral cover, and number of coral species by depth, reef, and date of census (continued).

Species	Depth (m)	Year	Number of Reef					
			1	2	9	10	11	12
<i>Porites porites</i>	0.5-3	85	2.67 (0.30)	0.27 (0.03)	0.55 (0.08)	0.25 (0.09)	0.10 (0.02)	0.07 (0.01)
		86	0.93 (0.11)	0.15 (0.02)	0.97 (0.13)	1.61 (0.39)	0.56 (0.11)	0.04 (0.01)
		88				0.06 (0.01)		
	>3-6	85	0.10 (0.02)	0.13 (0.02)	2.47 (0.32)		0.13 (0.02)	0.03 (0.00)
		86			1.28 (0.18)	0.38 (0.07)		
		88	0.06 (0.01)			0.07 (0.01)		
<i>Siderastrea siderea</i>	0.5-3	85	4.68 (0.52)	1.62 (0.19)	3.12 (0.46)	2.50 (0.88)	1.08 (0.20)	1.14 (0.18)
		86	4.62 (0.53)	0.25 (0.04)	2.69 (0.36)	5.30 (1.29)	0.80 (0.16)	3.03 (0.44)
		88	1.58 (0.16)	0.94 (0.09)	2.04 (0.26)	3.19 (0.35)	4.02 (0.49)	3.08 (0.34)
	>3-6	85	6.81 (1.36)	7.04 (1.04)	5.10 (0.67)	6.55 (1.97)	5.75 (0.85)	1.14 (0.15)
		86	10.40 (2.00)	4.22 (0.78)	2.78 (0.38)	5.21 (0.95)	6.46 (0.99)	1.78 (0.25)
		88	4.03 (0.98)	5.81 (1.04)	3.71 (0.49)	6.54 (1.10)	6.89 (1.47)	0.13 (0.02)
Total Coral Cover	0.5-3	85	18.46 (2.05)	22.77 (2.65)	27.72 (4.09)	17.69 (6.26)	15.12 (2.86)	27.25 (4.31)
		86	14.54 (1.67)	4.85 (0.77)	24.59 (3.29)	25.04 (6.07)	20.07 (4.01)	41.47 (6.05)
		88	7.16 (0.73)	6.27 (0.62)	11.42 (1.45)	10.56 (1.14)	11.79 (1.43)	14.79 (1.63)
	>3-6	85	26.26 (5.25)	28.00 (4.13)	37.77 (4.96)	19.90 (6.00)	16.86 (2.49)	17.93 (2.28)
		86	24.91 (4.79)	10.82 (2.01)	26.68 (3.66)	27.50 (5.02)	21.35 (3.26)	20.27 (2.84)
		88	14.40 (3.49)	9.82 (1.76)	13.37 (1.76)	18.12 (3.06)	19.76 (4.21)	9.79 (1.63)
Number of Species	0.5-3	85	15	15	17	13	16	23
		86	17	16	20	15	18	27
		88	12	12	17	19	15	16
	>3-6	85	12	22	27	11	26	33
		86	19	17	31	23	28	25
		88	16	16	23	19	18	14

The principal change in pattern by the third (1988) census was a striking decrease in cover, size, and diversity (cover) on unoiled reefs (Figures 5.2 and 5.3). Average total coral cover decreased from 28 to 13% on the four unoiled reefs between 0-3 m, and from 23 to 18% between 3-6 m, with more than half of the decrease due to the marked decline of *Acropora palmata* and *Montastrea annularis* (Table 5.3). This unexplained decrease may complicate assessment of the effects of oil or possible recovery using repeated measures MANOVA for all three years. The variability added by the decrease in coral cover between the 1986 and the 1988 surveys may affect the ability of the test to detect a significant difference. For example, the amount of oiling was significant in only three cases (total number of corals, $F=22.0$, d.f.=2,6, $P<.01$; size of *Diploria clivosa*, $F=27.4$, d.f.=2,3, $P=.01$; and marginally for diversity based on cover, $F=3.8$, d.f.=2,6, $P=0.08$). Also, the interaction of oil x year was significant for the cover and abundance of all corals combined ($F=4.0$, d.f.=4,12, $P=.02$ and $F=4.68$, d.f.=4,12, $P=0.01$ respectively), for diversity based on percent cover ($F=5.0$, d.f.=4,12, $P=.01$), for size of *D. clivosa* ($F=18.8$, d.f.=4,6, $P<.01$), and marginally for cover of *Siderastrea siderea* ($F=2.8$, d.f.=4,12, $P=0.07$). In contrast, the numbers of species and species diversity based on colonies showed no relation to oiling.

5.2.2 Recent Injury of Corals (Partial Mortality)

Patches of tissue of colonial animals like corals are frequently injured or harmed by natural enemies or physical processes (Loya 1976; Bak *et al.* 1977; Jackson and Palumbi 1979; Bak and van Eys 1980; Palumbi and Jackson 1982, 1983; Jackson 1983; Wahle 1983; Guzmán 1986; Guzmán and Cortés 1989b). Such injuries expose the white skeleton of coral colonies. After a few weeks, the exposed skeleton is overgrown by algae and other organisms (see references cited above). Eventually the coral may regenerate the injury by overgrowing these invaders, or the lesion may persist.

Quarterly surveys were made on twelve reefs (Table 5.2) for *Siderastrea siderea*, *Diploria strigosa*, *D. clivosa* and *Porites astreoides*. The proportion of corals showing recent injury was estimated from the percentage of colony surface that is recently bare (white), and the apparent causes of mortality (physical or biological) recorded when possible (e.g., salinity, temperature, abrasion, sedimentation, predation, overgrowth, diseases, etc.). It is important to note, however, that the validity of the census of injuries is not dependent on knowing the causes of injuries observed.

Methods

At each census, two 50 m transects were run on each reef parallel to the reef crest: one shallow transect in 0.5-1 m water depth, the other between >1 and 2 m depth. The two transects were a minimum of 10 m apart, depending on the horizontal profile of the reef. All colonies of the four species in a 1 m band to either

side of the transect were recorded for a total area of 100 m² per transect. The sizes of corals were estimated in five size classes (1-100 cm², 100-200 cm², 200-400 cm², 400-800 cm², and > 800 cm²) and the proportion of recent injury estimated visually (none, < 10% injury, > 10%).

Variations in amounts of injury at any census were examined by fitting a nonparametric hierarchical loglinear model (Norusis 1988) to a 4-dimensional contingency table with the following variables: coral species (3; *D. strigosa* was not common enough for this analysis), depths (2), level of injury (none, some), and level of oiling (none, moderate, heavy). Significance of association between oiling and injury was calculated for each census using the likelihood chi square ratio. Temporal variations in injury were analyzed by repeated measures MANOVA with amount of oil and depth as between-factor variables and time of census as the within-factor variable.

Results

Both the frequency and size of recent injuries increased substantially with the amount of oiling, particularly at the shallower depth (Figure 5.4). *Siderastrea siderea* was affected more than *Diploria clivosa* and *Porites astreoides*. There were too few data for *Diploria strigosa* to merit analysis, but the trend suggested an oil effect. Likelihood ratios were highly significant for all censuses except November 1987 for *S. siderea* and *P. astreoides* and only during the first census for *D. clivosa*.

Variation through time in total recent injury indicates that colonies were most affected at heavily and moderate sites during the first year after the spill (Figure 5.5). Repeated measures MANOVAs incorporating oil and depth as factors showed significant effects of oil for *S. siderea* ($F = 8.7$, d.f.=2,13, $P=.003$) and *Diploria clivosa* ($F=4.0$, d.f.=2,10, $P=.05$, but not for *Porites astreoides*, and marginally the interaction of oil x time (month) for *S. siderea* ($F= 1.8$, d.f.=10,65, $P=0.07$). Separate analysis of variance for each survey on the data presented in Figure 5.5 showed a significant increase of total injuries (regardless of depth) at oiled sites in all surveys but November 1986 and 1987 for *S. siderea* and only during August 1986 for *D. clivosa* and marginally for *P. astreoides*.

5.2.3 Resistance of Corals to Stress

Organisms may be weakened by exposure to oil in their natural environment in a complex, time-dependent fashion (Loya and Rinkevich 1980; Brown and Howard 1985). This is particularly likely for very shallow-water corals that suffered high levels of injury in oiled areas. Without moving corals to the laboratory, we were able to study their responses to stress on oiled reefs and to compare their performance with that of corals on unoiled reefs.

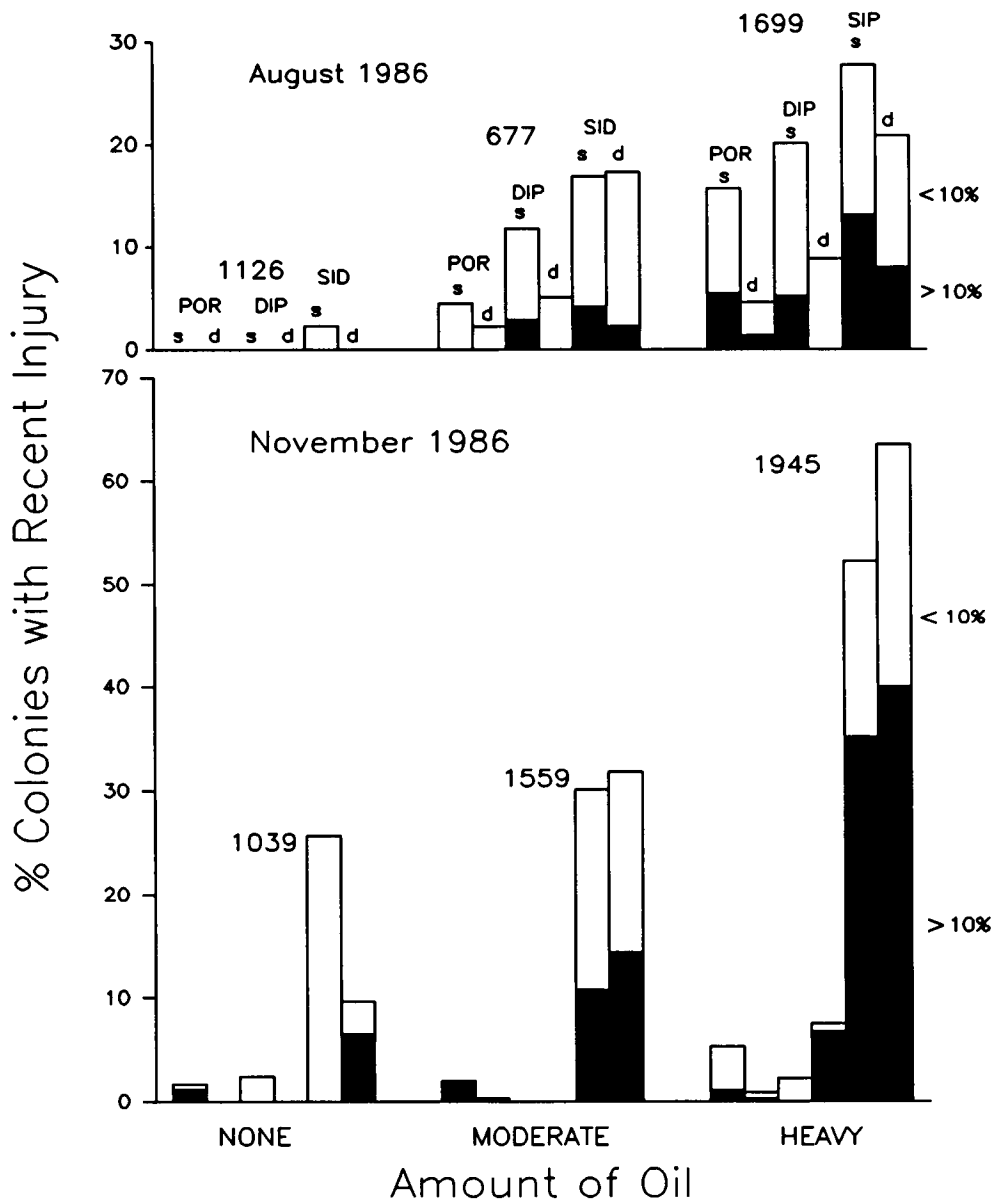


Figure 5.4 Frequency of colonies with recent injury (partial mortality) for the three most common species of massive corals in relation to the amount of oiling at 12 reefs from August 1986 to May 1988. There were four unoiled, two moderately oiled, and six heavily oiled reefs. Open bars represent coral colonies with recent injury that did not exceed 10% of the surface area of the coral; filled bars represent recent injury greater than 10%. POR = *Porites astreoides*, DIP = *Diploria clivosa*, SID = *Siderastrea siderea*, s = shallow (0.5-1 m), d = deep (>1-2 m).

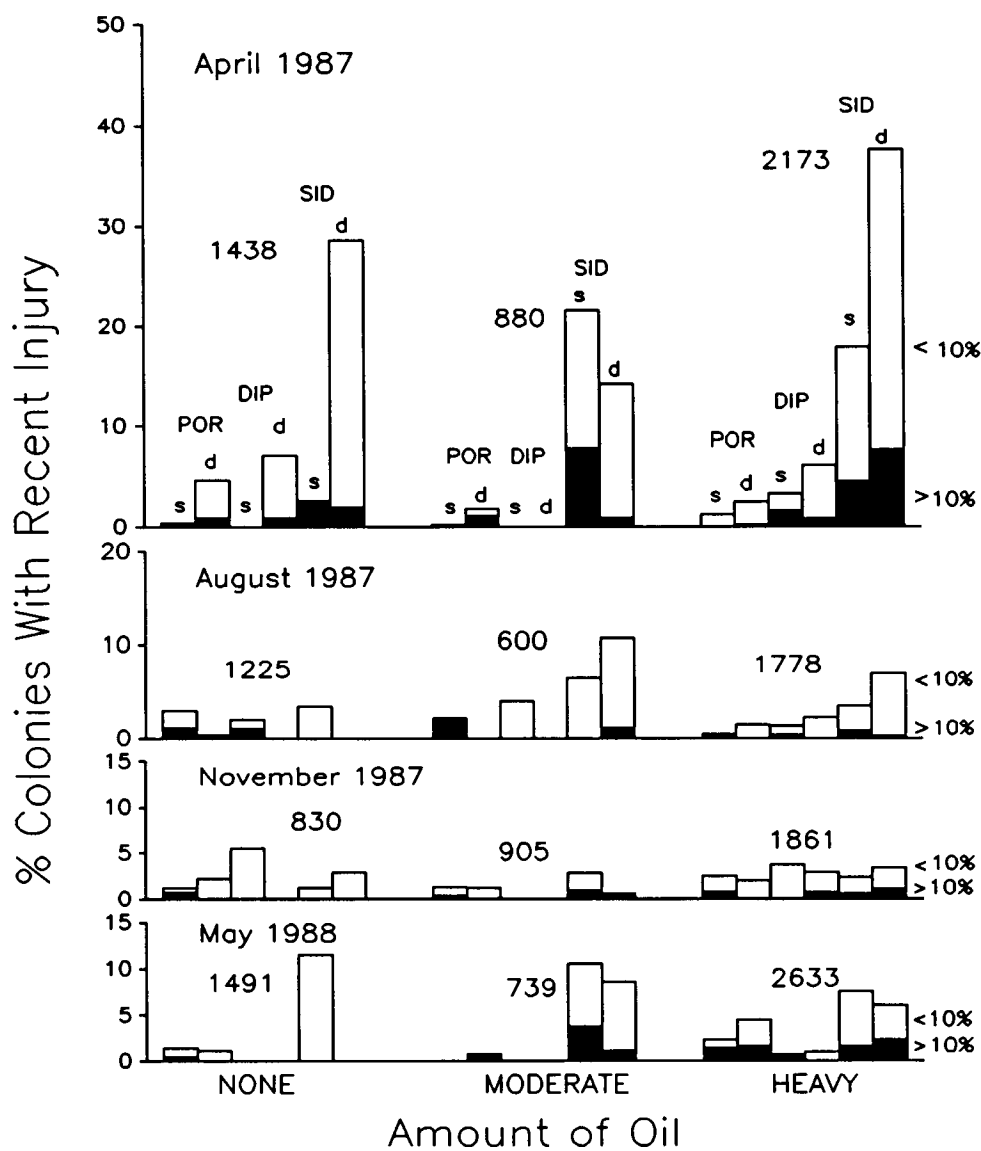


Figure 5.4 Frequency of colonies with recent injury (partial mortality) for the three most common species of massive corals in relation to the amount of oiling at 12 reefs from August 1986 to May 1988 (continued).

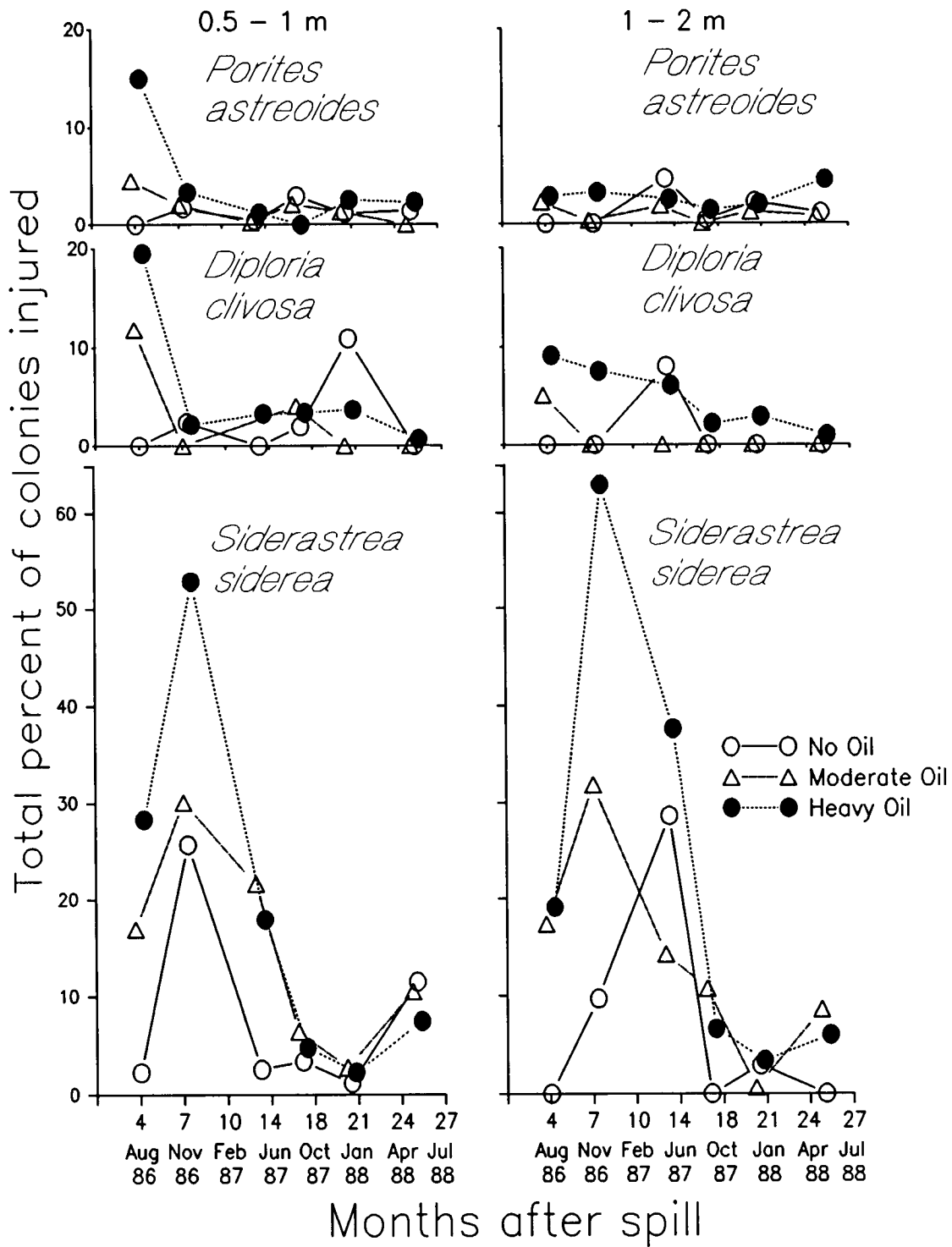


Figure 5.5 Percent of colonies that showed recent injury for the three most abundant coral species as a function of the amount of oiling and depth. The oil spill occurred in April 1986.

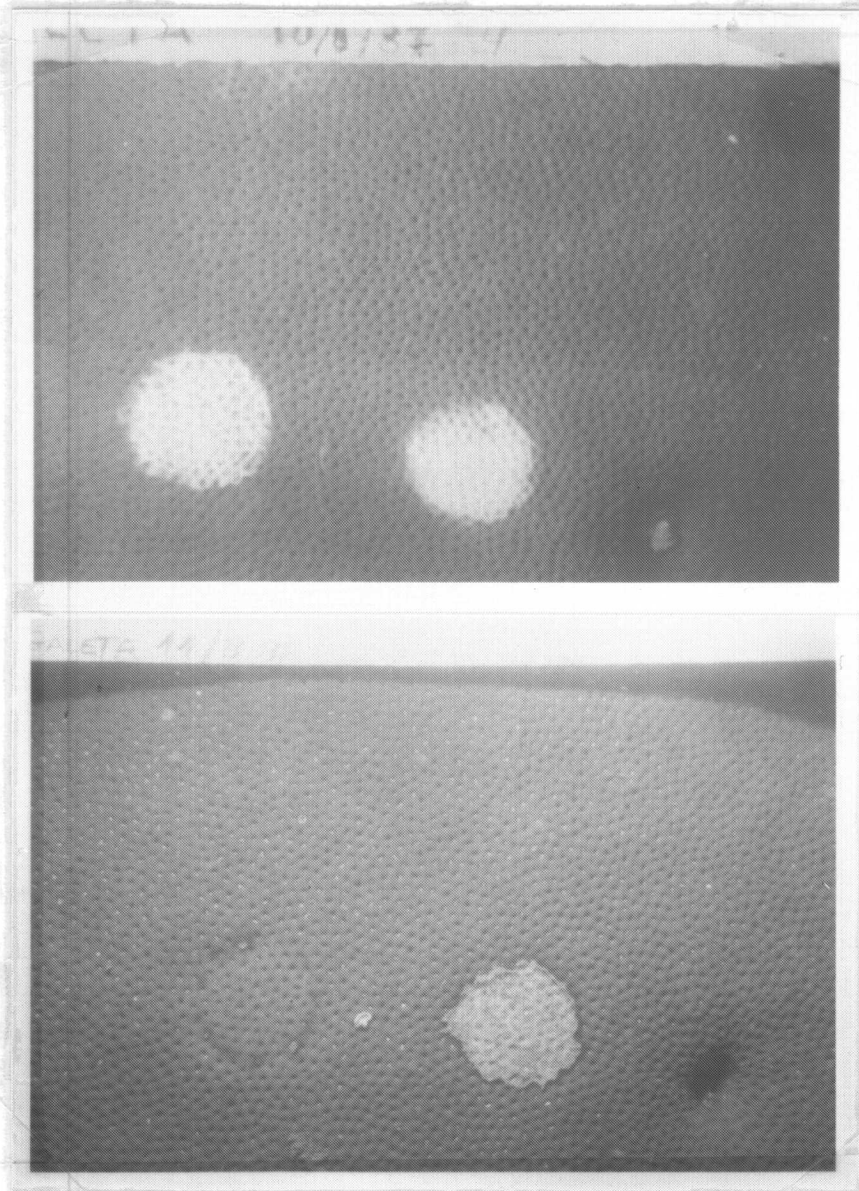


Figure 5.6 *Siderastrea siderea* after one month of recovery (bottom photo). Injury to the left was made by rasping and that to the right by blowing away the tissue using compressed air.

Methods

We developed a technique for measuring regeneration of injuries and tested the time intervals necessary to follow the recovery of injured corals using close-up photography. The procedures are based on those used in Curaçao for different species (Bak *et al.* 1977). Two different types of injuries, each 5.7 cm², are inflicted on a single colony. One type is made by rasping the skeleton with a rotating tool (skeleton and tissue lesions), the other by blasting off the coral tissue with air (tissue lesion). Photographs are taken just before and after the injuries, and at bi-monthly intervals thereafter for one year (Figure 5.6). They are being carried out on two reefs heavily exposed to oil (reefs 4 and 5) and two that were not exposed (reefs 11 and 12) at 1 m depth, using five replicates per species.

Results

Tissue removal (blasting) is more similar to the injuries observed on heavily oiled reefs after the spill. The first set of experiments was completed in September 1989.

Figure 5.7 shows the percent regeneration (after 180 days) for *Siderastrea siderea* and *Porites astreoides* subjected to the two kinds of experimental injuries (blasting and rasping). *Porites* recovered faster than *Siderastrea* on all reefs, and regeneration was faster at oiled reefs than at unoiled reefs.

5.2.4 Sclerochronology

Coral sections contain a wealth of information regarding rates of growth, past and present incidents of stress, and, to some extent, the history of the environment in which they live (Knutson *et al.* 1972; Dodge *et al.* 1974; MacIntyre and Smith 1974; Hudson *et al.* 1976; Dodge and Vaisnys 1980; Dodge and Lang 1983; Jackson 1983; Brown and Howard 1985). Growth rate (i.e., skeletal extension) can be determined from the width of annual growth bands using X-ray techniques (see references cited above). Furthermore, the history of colony injury and the amount of terrigenous sediments and runoff may be recorded and dated from such growth bands (see references cited above).

Alizarine staining of ten corals (five *S. siderea* and five *P. astreoides*) was done in November 1988 at Galeta reef to confirm annual banding. We have, however, confirmed that growth bands are annual using other means. This was done by collecting several specimens at different times during the year (i.e., beginning and ending of dry and rainy seasons) and comparing their band structure.

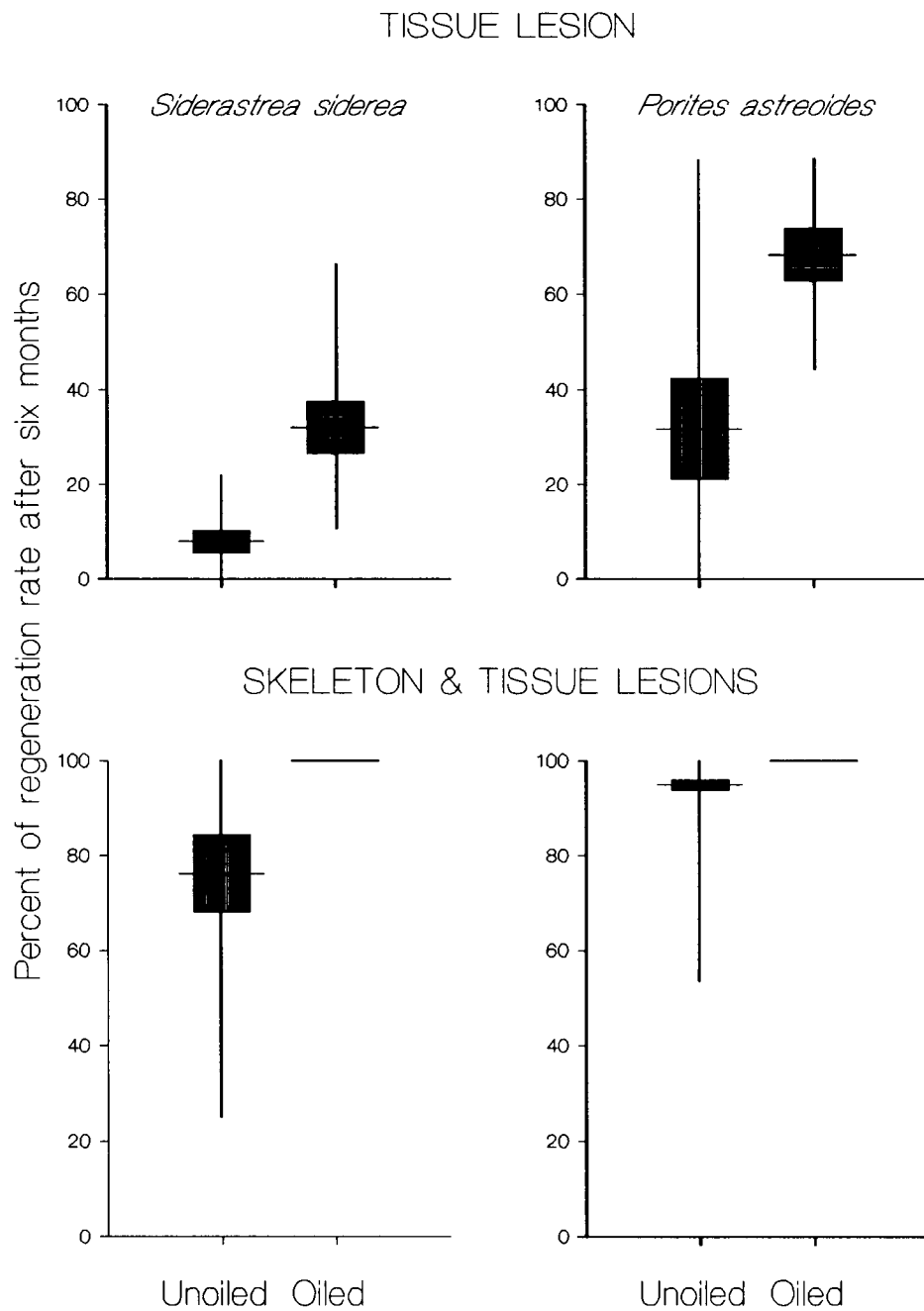


Figure 5.7 Percent regeneration (after 180 days) for *S. siderea* and *P. astreoides*. The colonies were subjected to two kinds of experimental injury (blasting and rasping) at two unooled and two oiled reefs.

Methods

The techniques are standard (Dodge 1980; Hughes and Jackson 1985; Guzmán and Cortés 1989a). Four coral species were studied (*Montastrea annularis*, *Diploria strigosa*, *Porites astreoides* and *Siderastrea siderea*). All coral colonies (five replicates per species, where present), of similar size, were collected from shallow water (1-3 m) at each reef and rinsed with fresh water. In the laboratory, slabs 5-7 mm thick were cut, using a rock saw (Figure 5.8), parallel to the axis of growth. The corals slabs were X-rayed on a Universal X-ray unit (model Little Giant 30) using Kodak Industrex AA film (Figure 5.9). Exposures were made at 60 KVp, 30 ma for 15 seconds with a source-to-film distance of 1 m. Contact X-radiograph prints were made on high-contrast paper for growth rate analyses. Growth bands were measured (Figure 5.10) using a fine ruler (resolution 0.5 mm). One transect along the axis of maximum growth was measured for each coral. Growth bands were measured inward along the transect starting at the outer (distal) surface of the most distal high-density band to the distal surface of the next oldest high-density band. Comparison of corals collected during mid-to late-1987 and early 1988 shows that the beginning of the low density band coincides with the onset of the "dry season" in late December so that each primary couplet of light and dense bands approximately coincides with the calendar year.

In order to understand the yearly variation patterns in growth before the year of the spill for each of the four coral species, the nine years previous to the spill (1977-1985) were examined by two-way repeated measures ANOVA using the three groups of reefs that were subsequently affected differently by the spill (unoiled, moderately oiled, or heavily oiled) and time (years) as factors. In this case, repeated measures analysis is required because successive growth bands are measured in the same corals. To examine the effect of the 1986 oil spill we compared growth in the year of the oil spill to the nine previous years by calculating the natural logarithm of the ratio of mean width in 1986 divided by the mean width for 1977-1985 and by comparing these values in a 2-way ANOVA with species and amount of oil as factors.

Preliminary results based on colonies of *S. siderea* and *P. astreoides* (reefs 2, 4, 6, 7, and 9-12) showed very high variance in growth between years and between reef sites independent of possible effects of the 1986 oil spill. To try to reduce effects of high variability, three more reef sites were added. Moreover, so little previous work had been done on these two species (Hubbard and Scaturro 1985; Huston 1985) that it seemed prudent to compare these results to published standards for better-studied taxa. Accordingly, a similar number of replicate colonies of two additional common Caribbean species whose growth patterns are well known were collected. These additional species, *Montastrea annularis* and *Diploria strigosa*, are not as abundant as *S. siderea* and *P. astreoides*, but could be found on most of the



Figure 5.8 Rock saw (right side) used to cut coral slabs 5-7 mm thick, and coral collections.

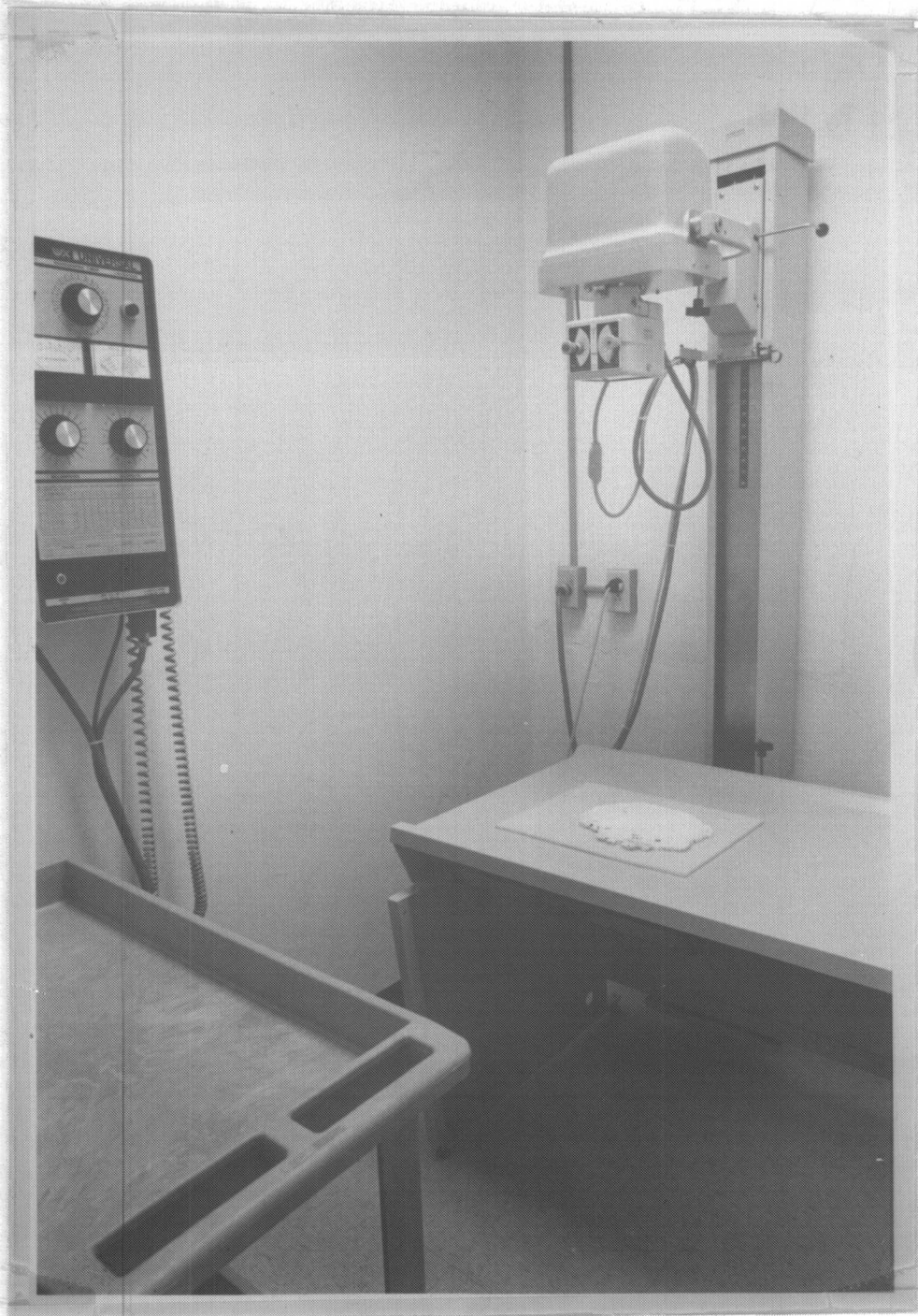


Figure 5.9 X-ray unit used for the sclerochronology study of corals.

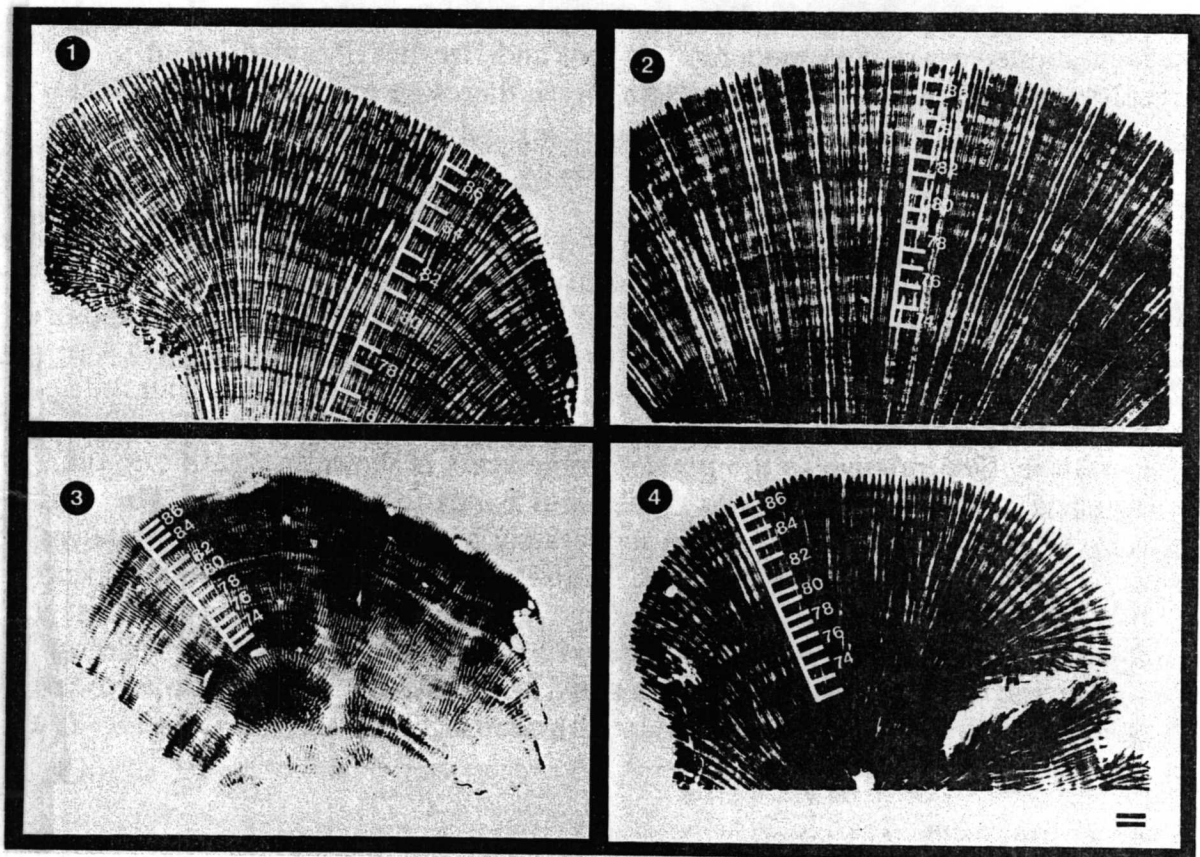


Figure 5.10 Positive radiographs for the four coral species examined for coral growth showing bands for the last 10 years. 1- *M. annularis*, 2- *D. strigosa*, 3- *P. astreoides*, and 4- *S. siderea*. Scale bar = 1 cm.

reefs. Growth of *Montastrea annularis* has been studied more than any other Caribbean coral (Aller and Dodge 1974; Dodge *et al.* 1977; Gladfelter *et al.* 1978; Dodge and Brass 1984; Hubbard and Scaturro 1985; Huston 1985). *Diploria strigosa* was chosen because of the detailed and extensive experimental work done at the Bermuda Biological Station for Research (BBSR) on the effects of oil on this species (Dodge *et al.* 1984, 1985; Knap 1987).

A total of 190 colonies (four coral species) were collected at eleven reef sites: four unoiled reefs, two moderately oiled, and five heavily oiled. Ten specimens collected did not show clear bands and were therefore excluded from the analyses.

Results

Yearly mean growth rates were determined for each of the four species measured from 1977 to 1986 (Appendix Tables D.1-D.4). Results showed not significant nine-year variations in growth between reefs grouped by their exposure to oil in 1986 for any of the four coral species (Figure 5.11).

Most of the lowest mean annual growth rates measured for all four species on oiled reefs occurred in 1986, the year of the spill (Figure 5.11). This change in growth in 1986 relative to the previous nine years is shown in relation to oiling in Figure 5.12 using data only for corals with bands for all 10 years. There was a substantial reduction in growth on moderately and heavily oiled reefs relative to unoiled reefs for all species except *Siderastrea siderea*. Two-way ANOVA using the data from Figure 5.12, and level of oiling (reef groups) and species as factors gave a nearly significant result for oiling ($F=2.93$, $P=.057$), but not for species or the interaction of species and oil. Subsequent 1-way ANOVA for oiling alone, to test all species combined (general coral growth), was significant ($F=3.35$, $d.f.= 2, 154$, $P=.038$).

5.2.5 Physical Monitoring

The importance of physical monitoring is to assess circumstantially the possible effects of factors other than oil on coral abundance and condition and, conversely, to better interpret possible indirect biological consequences of the spill. There has already been extensive mortality of red mangrove trees due to the spill (Jackson *et al.* 1989; Chapter 6), and some of these trees have fallen or rotted away (Chapter 6). In a few years, coastal erosion is likely to increase considerably, with net transport of sediments (perhaps rich in petroleum hydrocarbons) out onto the reefs. Thus, corals could suffer considerable, indirect sediment stress years after the spill.

Measurements of suspended and resuspended sediments were made at monthly intervals on 11 of the reefs under study (Table 5.2). In addition, salinity and sea surface temperature were monitored every time we visited any of the study reefs. All these physical variables are known from experiments to affect corals or to be strongly correlated with their distribution (Dodge *et al.* 1974; Cortes and Risk 1985).

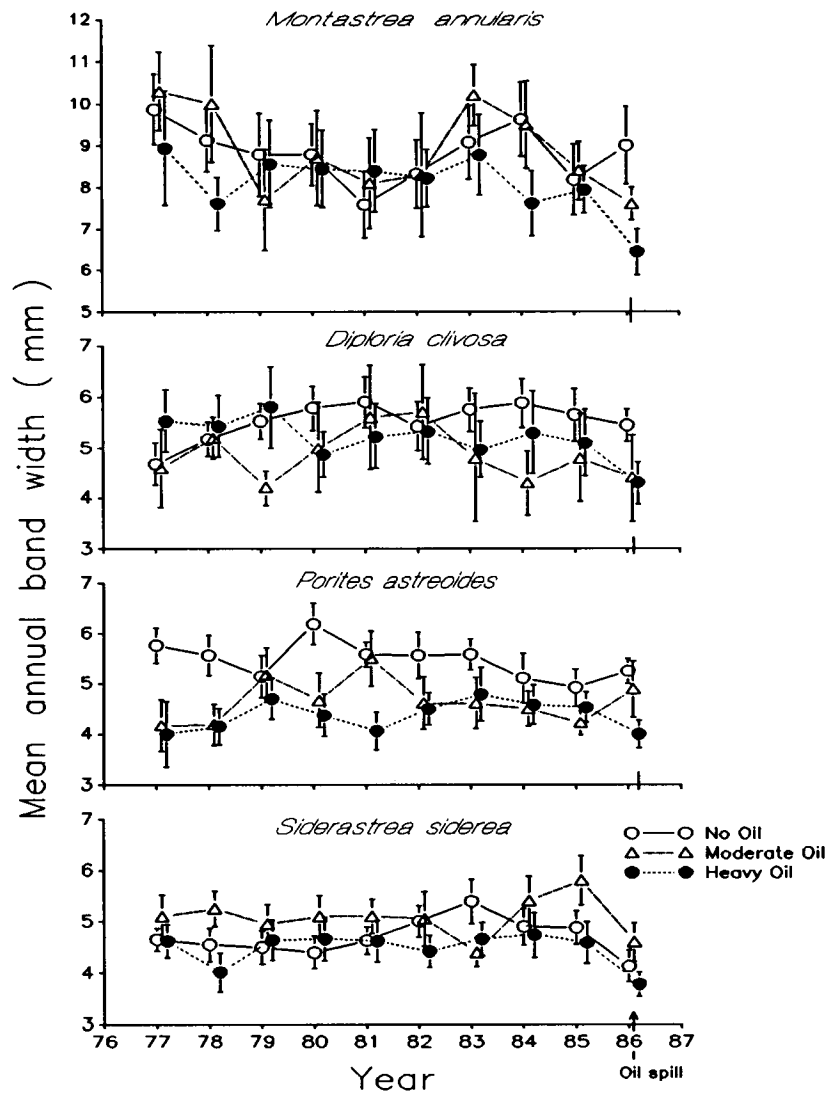


Figure 5.11 Mean annual band width (mm) from 1977 to 1986 for the four species at 11 reefs in relation to the amount of oiling in 1986. Four reefs were unoiled, two moderately oiled, and five heavily oiled.

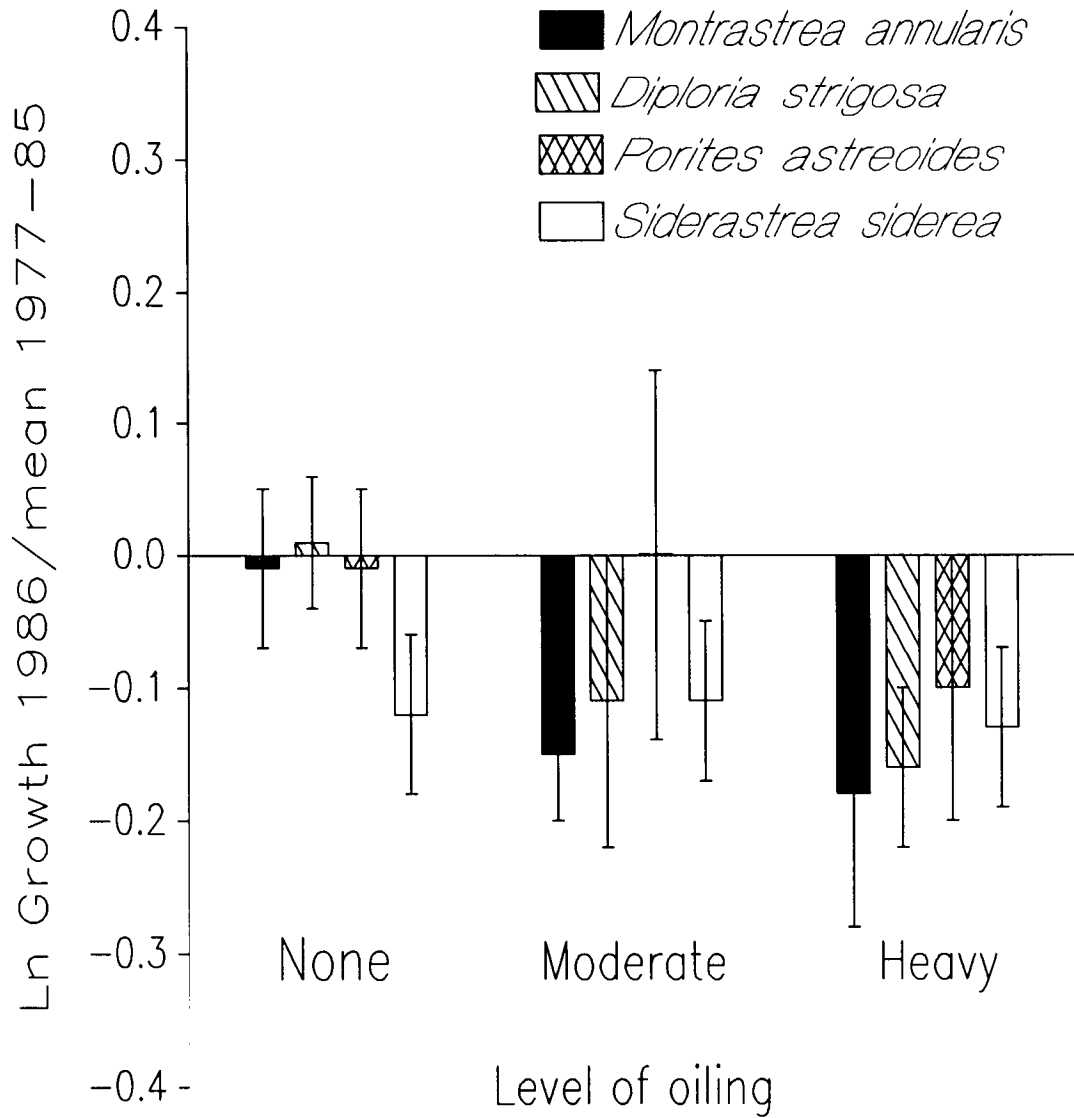


Figure 5.12 Coral growth for four coral species versus their mean growth over the previous nine years (1977-1985) as a function of level of oiling in 1986.

Methods

Monthly observations of particulate matter and resuspended sediments began in October 1987; observations of salinity and temperature began in May 1987, weather permitting. Oil slicks were recorded during every visit to the reef sites. Approximate values of salinity (+/- 1 o/oo) were determined using a hand-held refractometer (American Optical Corporation). Temperature was measured using a mercury thermometer with 0.5°C divisions. Suspended particulate matter (SPM) was determined by filtration (Cortés and Risk 1985; Tomascik and Sander 1985; Guzmán 1986). Six replicate 1 liter samples of seawater were collected at each reef. Samples were filtered using pre-weighed Millipore filters (47 mm). A few drops of sodium azide (NaN₃) were used to poison the samples to prevent decomposition of organic matter. Filters were oven dried at 45°C for 24 hours, cooled and reweighed to the nearest 0.01 mg.

Resuspended sediments were sampled using six sediment traps per reef (Young and Rhoads 1971; Cortés and Risk 1985; Guzmán 1986). The traps were constructed of PVC pipe 8 cm by 24 cm (height/width ratio of three) and were set on iron stakes 20 cm above the bottom. The traps were collected 2-5 days after placement on the reef, and the sediments trapped were recovered by filtering the water on pre-weighed paper filters (Whatman # 1, 12.5 cm). Samples were then processed following the same routine described for SPM.

Results

Table 5.4 is a summary of sea surface temperature and salinity data. Water temperature averaged 29°C and varied little during the year.

Table 5.5 shows the mean values for SPM from October 1987 to March 1988. In general, values were less on control reefs than on oiled reefs; statistics have not yet been calculated.

Chronic oil slicks (see Table 5.1) or adverse weather have occasionally affected the setting up and collecting of sediment traps, so there are missing data.

In general, there appear to be more resuspended sediments on the oiled reefs (Table 5.6). The long-term study may provide a better view on the effects of the measured variables on the reefs.

Table 5.4 Sea surface temperature (T, °C) and salinity (S, o/oo) at four oiled reefs and four unoiled reefs recorded from May 1987 to August 1988 (see Figure 5.1 for reef sites).

		Oiled Reefs				Unoiled Reefs			
		4	6	7	8	9	10	11	12
May 87	T (°C)	28	28	28	27	28	29	29	29
	S (o/oo)	34	24	24	28	37	35	36	36
Jun	T	28	28	29	29	29	28	29	29
	S	30	28	30	32	32	32	33	32
Jul	T	28	29	29	29	28	27	28	29
	S	26	26	28	30	28	34	30	30
Aug	T	30	30	30	30	29	29	30	29
	S	32	36	30	31	34	34	35	34
Sep	T	30	29	29	30	29	29	29	30
	S	32	33	32	32	30	34	32	32
Oct	T	30	29	29	28	30	29	29	29
	S	33	33	33	33	33	33	34	33
Nov	T	29	29	28	28	29	28	28	28
	S	34	25	27	31	36	37	37	36
Dec	T	28	28	28	28	28	28	28	28
	S	32	34	34	33	34	35	32	32
Jan 88	T	NO DATA				NO DATA			
	S	NO DATA				NO DATA			
Feb	T	30	29	29	29	29	28	28	29
	S	37	37	27	27	27	27	27	27
Mar	T	NO DATA				NO DATA			
	S	NO DATA				NO DATA			
Apr	T	30	29	29	29	28	28	28	28
	S	37	37	37	37	37	37	37	37
May	T	31	30	31	31	29	29	29	29
	S	38	38	38	38	39	38	38	39
Jun	T	29	29	29	29	29	29	29	29
	S	37	37	37	37	37	37	37	37
Jul	T	28	29	29	29	28	28	28	28
	S	33	34	34	35	35	36	37	35
Aug	T	NO DATA				NO DATA			
	S	NO DATA				NO DATA			

Table 5.6 Mean values for resuspended sediments (mg/cm² /day) on seven oiled (2 moderately*, 5 heavily) and four unoiled reefs, from October 1987 to March 1989. N.D.= No data collected due to bad sea conditions; none of the reefs could be sampled during December 1987-April 1988 and December 1988-March 1989. Values below the means are the standard error/sample size.

Month-yr	Oiled Reefs							Unoiled Reefs			
	1*	2	4	5	6	7	8*	9	10	11	12
Oct 1987	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
Nov 1987	N.D.	N.D.	206.3 30.4/6	N.D.	492.1 46.6/5	254.6 34.4/6	N.D.	218.2 83.5/6	324.3 47.0/6	40.1 6.2/6	95.5 40.1/6
May 1988	N.D.	N.D.	117.2 46.5/6	N.D.	159.9 29.3/6	50.0 11.8/6	53.0 13.3/6	64.2 11.7/3	23.6 5.3/6	19.4 3.7/6	14.7 1.8/3
Jun 1988	N.D.	N.D.	4.8 0.5/6	N.D.	N.D.	5.4 0.6/6	21.7 7.6/6	6.6 2.2/4	5.7 0.5/6	3.7 0.2/6	3.2 0.4/6
Jul 1988	N.D.	N.D.	238.6 44.2/3	N.D.	N.D.	211.5 78.6/6	277.3 80.6/6	16.0 13.9/4	25.6 3.2/6	11.0 1.0/6	11.3 3.8/6
Sep 1988	N.D.	N.D.	17.8 2.0/5	N.D.	N.D.	15.6 5.0/6	11.8 2.6/5	10.0 3.9/6	6.9 0.8/6	12.3 1.9/6	4.5 1.1/6
Oct 1988	N.D.	10.0 1.5/6	9.4 0.9/6	N.D.	N.D.	31.9 5.2/6	52.2 18.7/6	2.7 0.1/3	4.9 0.9/6	3.2 0.7/6	2.9 0.4/6
Nov 1988	4.8 0.7/6	15.4 3.6/6	15.4 2.3/6	22.4 2.1/6	N.D.	9.2 1.3/6	4.9 1.0/6	1.3 0.3/3	2.4 0.2/6	7.6 0.7/6	2.0 0.5/6

5.3 Preliminary Conclusions

We studied the short-term effects of the spill on common shallow subtidal reef corals. Numbers of corals, total coral cover, and species diversity based on cover decreased significantly with increased amounts of oiling. Cover of the large branching coral *Acropora palmata* decreased most. Frequency and size of recent injuries on massive corals increased with level of oiling, particularly for *Siderastrea siderea*. Growth of three massive species (*Porites astreoides*, *Diploria strigosa*, and *Montastrea annularis*, but not *S. siderea*) was less on oiled reefs in the year of the spill (1986) than the average of the nine previous years.

Heavy slicks continue to emerge from mangroves and the landfill beneath the refinery, most frequently following heavy rains, but also after very high tides. The pattern of this chronic oiling is consistent with that observed immediately after the

oil spill. In 1988, reefs that were heavily oiled by the spill still had oil over them about 82% of the 122 days we visited them (see Table 5.1). In contrast, we observed oil on the moderately oiled reefs on only 15% of the 30 days, and found no oil in 61 days of observations for reefs unoiled in 1986. These differences are highly significant (chi square=48.2, d.f.=2, $P < 0.001$). Some of this oil is almost certainly reaching the bottom adsorbed to suspended and resuspended sediments. Sedimentation rates average 5.3 mg/L and 99.3 mg/cm²/day, respectively, over reefs in Bahía Las Minas. Given the enormous amounts of oil apparently still locked up in mangrove sediments, this chronic pollution due to the original spill is likely to last many years.

Chapter 6 Mangrove Forests

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6.1 Introduction

Since March 1989, this project has been investigating long-term effects of the Refinería Panamá oil spill of April 1986 (Chapter 1) on mangrove forests in the region of Bahía Las Minas, Atlantic coast of Panama. This large bay is characterized by wide, dense bands of mangrove trees bordering tidal channels and more riverine parts, as well as backing exposed reef flats. The shoreline is generally convoluted with numerous small islets, and the entire bay is divided into three parts by two larger islands, Isla Largo Remo and Isla Payardi (Figure 6.1).

These divisions of the bay were reinforced over the past 15 years by human activities, notably with the oil refinery on Isla Payardi and a cement factory on the island between Isla Largo Remo and the mainland. Changes included joining of the larger islands to the mainland with causeways and port construction. However, while this work was relatively minor, it appears that patterns of water flow were altered considerably. For instance, causeways are generally solid, without bridges. In addition, previous water flow patterns were apparently altered further by the water cooling process of an electricity generating plant positioned on the causeway linking Isla Payardi to the mainland (Figure 6.1). Water is pumped from one embayment (Eastern) into another (Central), where there is a chronic impact on mangrove forests surrounding the outlet channel and nearby (personal observations). This impact presumably results not only from altered water flow, but also from heated water (measured at 35°C) and occasional smaller oil spills. Furthermore, watershed lands were largely deforested for both urban development and an expanding grazing industry. This deforestation has presumably resulted in massive topsoil erosion, manifest in the bay as increased turbidity and decreased depths. In this case, field observations are supported by those from aerial photography and maps spanning at least twenty years.

In April 1986, this situation was further aggravated when the refinery spill killed a large portion of fringing mangrove (Jackson *et al.* 1989; Teas *et al.* 1989). Despite efforts to contain oil to the central embayment, it eventually spread throughout the bay, except for two small enclaves on the western side and the inner larger embayment on the eastern side (Figure 6.1).

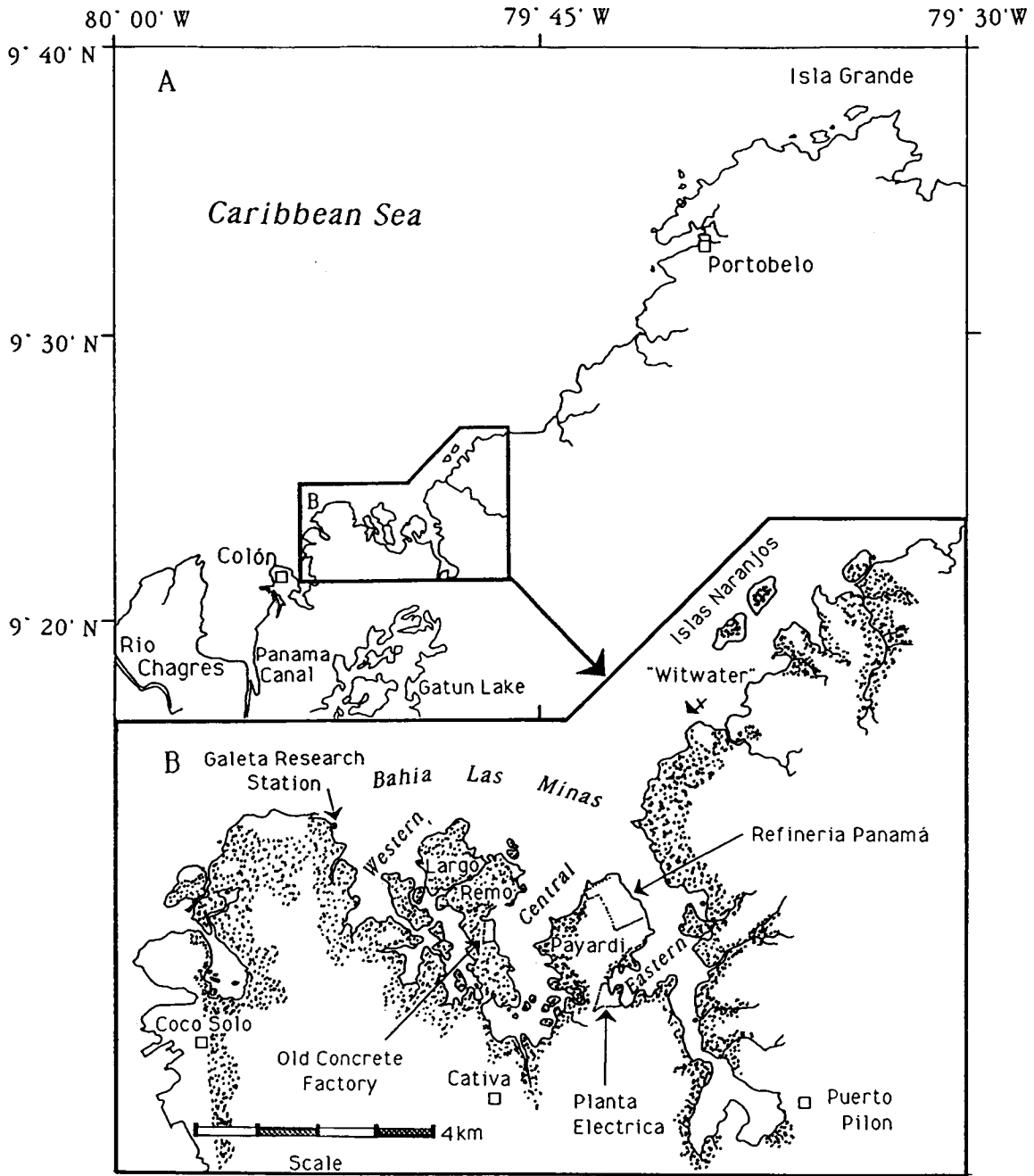


Figure 6.1 Map of the study area for the Mangrove Forest Project. Stippled areas (Inset B) denote mangrove forests around Bahía Las Minas. Open squares denote major population centers.

6.2 Immediate Impact of Oil on Mangroves

The severity of the impact of the spill on mangroves became readily apparent soon afterwards, when large patches of mature trees died after dropping their leaves. Oil thickly coated exposed roots and covered the surrounding substratum. Tree mortality presumably resulted from oil toxicity or the physical blocking of the vital respiratory function of the roots or both. Whatever the mechanism, the spilled oil resulted directly in the death of approximately 75 hectares of mangrove forest (Teas *et al.* 1989) along 11 km of coastline (Jackson *et al.* 1989), representing the most extensive impact of oiling ever recorded for mangroves. Other spills have been much less devastating to mangroves. For example, when the *Zoe Colocotronis* grounded off Puerto Rico in 1973 (note the review by Wardrop 1987), only one hectare of forest was destroyed. However, this example provides an important consideration that was not adequately quantified for the refinery spill in Panama: the precise extent of oiling compared with that of deforestation. In Puerto Rico, eight hectares of mangroves were reportedly oiled, indicating that only 15% of the area oiled became deforested. If a similar ratio applies to Bahía Las Minas, then possibly 400 hectares were oiled. Especially since fatalities were not limited to trees, this estimate provides a better quantification of the total area impacted in mangroves. In places where trees survived, faunal mortality was reportedly high. For instance, STRI personnel observed massive numbers of dead mangrove-inhabiting animals, particularly crustaceans and molluscs, in mangroves adjacent to the Galeta field station. Except for epibiota on fringing mangrove roots (Chapter 7), none of this die-off was quantified.

Three years post-spill, the most obvious evidence was the weathered and rotting remains of dead trees, but these were fast disappearing as they decayed. In many areas, they were also becoming submerged in forests of young seedlings. The seedlings included those planted by the Refinería Panamá (Teas *et al.* 1989) and an unknown number of natural recruits of various ages. In the mud substratum, however, oil was still present in sufficient amounts to come oozing out whenever it rained, or whenever the sediments were disturbed. While trees and animals did not appear to be still dying from this residual oil, questions remained about how recovery may be affected, and whether distribution and growth patterns have been altered.

In general, the impact of oil on mangroves is divided into two distinct effects depending on the amount of oiling and site characteristics. The first effect is deforestation, where the presence and quantity of oil were sufficient to kill mature trees, hence destroying the main structure of the habitat. Secondly, where the habitat remained intact and trees survived because oiling was presumably less, there may be sublethal consequences for survivors. In sites of deforestation, recovery chiefly involves processes of recruitment, seedling growth, and possibly asexual or congenetic sprawling of mature survivors. In sublethally affected areas, mature trees could have altered patterns of growth and development. In each case, recovery is expected to be mainly a function of time and the initial concentration of oil. The time period,

however, differs markedly in each case. While oil impact for mature survivors may be reduced within three-and-a-half years (see Chapter 6.6.1), this period is only the beginning for seedlings growing in deforested areas because complete recovery may take up to 50 or 70 years (e.g., Sandrasegaran 1971).

Post-spill surveys have made important generalizations regarding responses of mangroves to oiling (Wardrop 1987). These however, all relate to impact immediately following oiling and rarely apply to long-term recovery. The 1986 Refinería Panamá spill provides a rare opportunity to assess such recovery.

6.3 The Mangrove Forest Ecosystem and Long-term Recovery

The best way to assess such long-term recovery in a few years is to extrapolate from existing processes in the ecosystem, comparing oil-affected and unoiled areas. This, however, presupposes some knowledge about the processes. Unfortunately, this was not the case in Panama, and the choice of studies required for this project initially depended on evidence from elsewhere. Some recent studies in Australia provided important insights, since they dealt with mangroves of the same genus growing under similar climatic conditions (e.g., Duke *et al.* 1984; Robertson 1986; Robertson and Duke 1987; Smith *et al.* 1989; Robertson and Daniel, in press). These highlight the importance and close relationship between primary production and a small number of macroscopic primary consumers.

Three major components of primary production, leaves, reproductive parts, and wood, flow rapidly to higher trophic levels (Figure 6.2). First, canopy herbivores in Australia removed around 10% of standing leaf biomass (Robertson and Duke 1987). Second, nearby crabs removed 40% of the leaves (Robertson 1986) and 50-100% of the propagules (Smith *et al.* 1989) from the forest floor. Third, wood breakdown was rapid, with 50% loss in dry mass after about seven years (Robertson and Daniel, in press). These results clearly indicate that a few processes could account for a large portion of primary production (note the heavier arrows in Figure 6.2). In Bahía Las Minas, preliminary field observations suggest that similar mechanisms apply normally in unoiled sites. However, in oiled locations there may be disruption of these processes. For example, there are often excessive accumulations of leaf litter, which could be the result of depleted crab numbers. One of the important tasks of this project, therefore, will be to determine levels of primary consumption, establishing the relationship between crabs and mangrove litter, and assessing changes in these processes at oiled locations.

This understanding of trophic inter-relationships will be coupled with other studies focused on the recovery of deforested sites: recruitment, survival, and growth of seedlings. These will be compared in oiled and unoiled sites, providing the basis for some estimation of the time required for recovery of Panamanian mangrove forests.

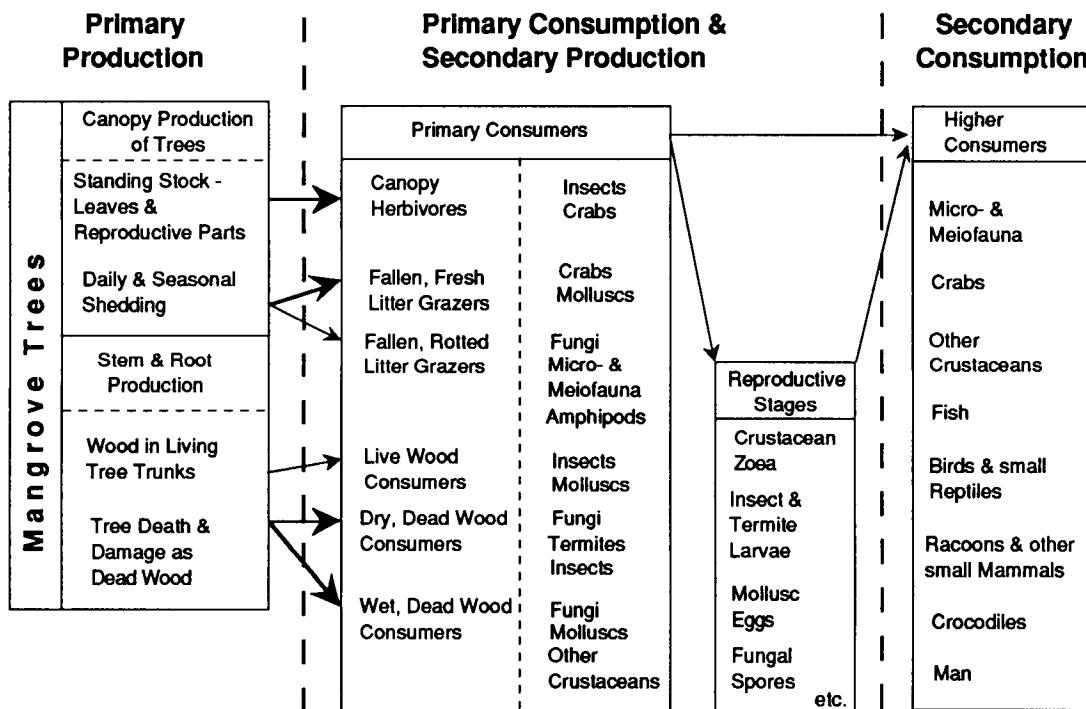


Figure 6.2 Major trophic pathways in tropical mangrove forests. Arrows indicate the major processes, and the greater thickness of lines displays those of greatest importance (see text).

Table 6.1 List of studies in the Mangrove Forest Project.

SECTION 1. Extent of deforestation and surrounding mangrove
(a) Mapping study
(b) Profile study
SECTION 2. Effect on surviving biota
(a) Tree and forest study
(b) Primary consumer study
(c) Primary production study
(d) Primary consumption study
SECTION 3. Effect and recovery in deforested areas
(a) Seedling recruitment and demography study
(b) Seedling growth study
(c) Forest gap primary consumer study

6.4 Description of Studies

This project is comprised of three sections, each including a number of studies (Table 6.1) based on the different biotic scales and major impacts of oiling. The first section examines the precise extent of mangroves and post-spill deforestation. The second and third sections look at the processes and biota of selected sites in the study area (Table 6.2, Figure 6.3).

Most study sites were chosen to coordinate with the Mangrove Root Project (Chapter 7), and to minimize the number of samples for hydrocarbon analyses. Accordingly, the two projects consider the same habitats for impact and effect of oiling. There are two treatments (oiled and unoled), three habitats (open, channel and river), and four or five replicates (generally four).

All sections were scheduled to begin approximately at the same time (Table 6.3), although individual studies have focused initially on primary production and growth, and later will progress to primary consumption.

Section one is primarily a mapping exercise, plotting vegetation boundaries in plan and profile views. Resulting maps will, for the first time, provide a visual display of the extent of the impact of the 1986 spill on mangrove forests. These studies will provide data for considering how tides and wind at the time of the spill influenced the observed impact. The maps will also display the areas of planting by Refinería Panamá (Teas *et al.* 1989).

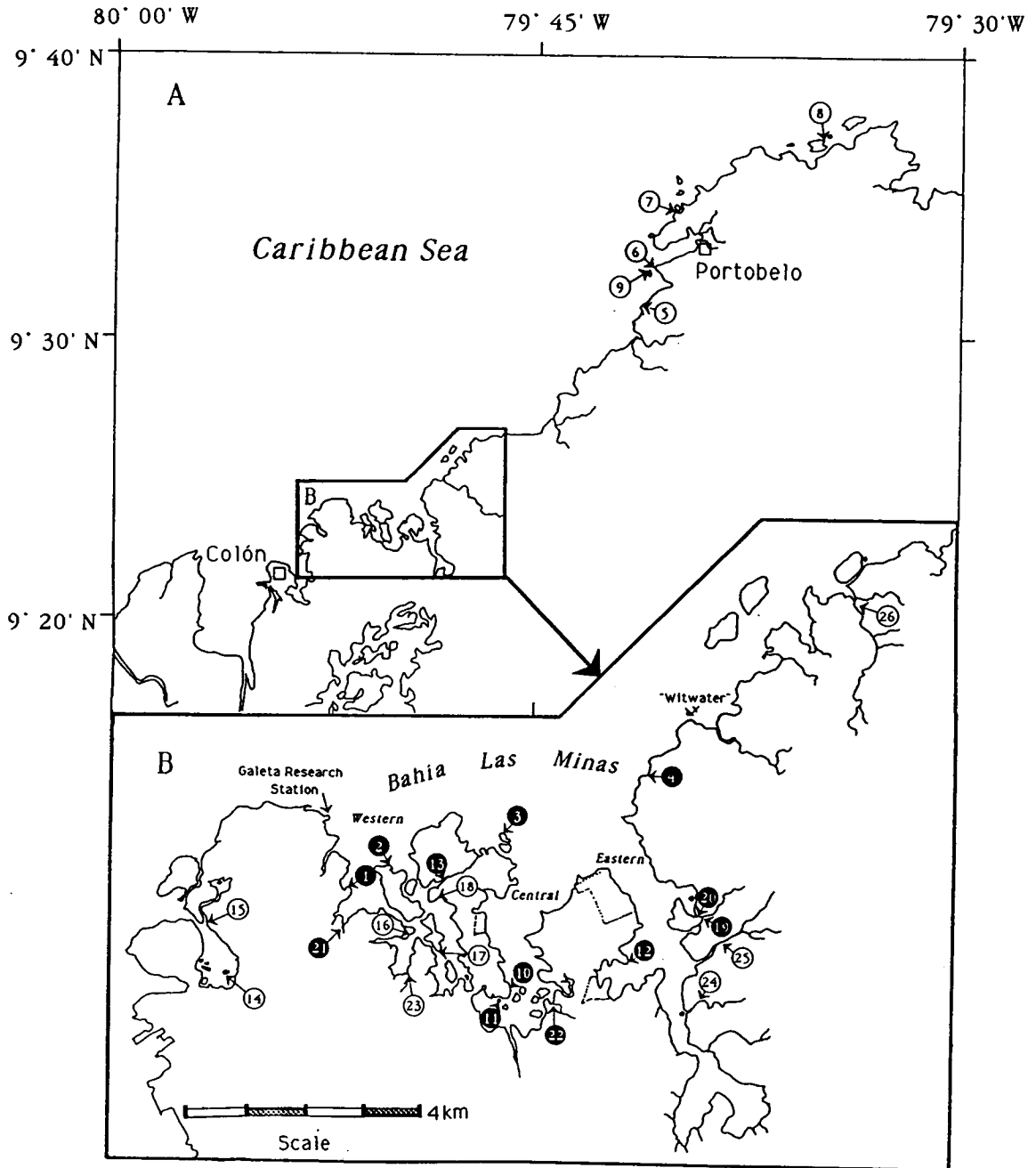


Figure 6.3 Map of the twenty-six study sites used by the Mangrove Forest Project. Site numbers from Table 6.2. Open circles denote unoiled control sites, and closed circles denote oiled sites. Open squares denote major population centers.

Table 6.2 Mangrove Forest Project sites. Descriptions, replicate numbers codes, and corresponding Mangrove Root Project site codes (Chapter 7) . Letters listed with replicate numbers refer to habitats (open=O, channel=C and riverine=R) and treatments (oiled=O and unoiled=U). The first letter of the Forest Code refers to either locations in Bahía Las Minas (i.e., in BLM, as western=W, central=C and eastern=E), towards Isla Grande(=G), near Isla Margarita (=M), or Islas Naranjos (=N). The second letter refers to habitats already listed for replicate number. Third and fourth letters of Forest Code relate to site description names. - = not matching site.

Site No.	Replic. Description	Site	Forest Code	Root Code
1	OO1	Mina - West BLM	WOMI	MINM
2	OO2	Peña Guapa - West BLM	WOPG	PGM
3	OO3	Droque - Central BLM	CODR	DROM
4	OO4	Punta Muerto - East BLM	EOPM	PMM
5	OU1	María Soto - I. Grande	GOMS	MSM
6	OU2	Portobelo - I. Grande	GOPB	PBM
7	OU3	Padre - I. Grande	GOPA	PADM
8	OU4	Lintón - I. Grande	GOLI	LINM
9	OU5	Magoté Sur - I. Grande	GOMG	-
10	CO1	Samba Bonita - Central BLM	CCSB	SBCE
11	CO2	Pequeña - Central BLM	CCPQ	SBCS
12	CO4	Payardi - East BLM	ECPY	PCS
13	CO5	Largo Remo - West BLM	WCLR	LRCW
14	CU1	Sur - Margarita	MCSU	MACS
15	CU2	Norte - Margarita	MCNO	MACN
16	CU3	Norte - West BLM	WCNO	HIDC
17	CU4	Sur - West BLM	WCSU	SBCW
18	CU5	Largo Remo Sur - West BLM	WCLS	LRCS
19	RO3	Puerto Sur - East BLM	ERPS	PMRE
20	RO4	Puerto Norte - East BLM	ERPNO	PMRW
21	RO5	Rabinowitz - West BLM	WRRB	-
22	RO6	Kuna - Central BLM	CRKU	-
23	RU1	Hidden - West BLM	WRHD	HIDR
24	RU2	Unnamed - East BLM	ERUN	UNR
25	RU3	Alejandro - East BLM	ERJA	ALER
26	RU4	Las Mercedes - Naranjos	NRME	MERR

Table 6.3 Monitoring schedule of the Mangrove Forest Project, 1989 to 1992. Symbols denote full effort (+), or part effort (-), and beginning (<) and completion (>) of data collection.

Study	1989				1990				1991				1992				
	Jun	Aug	Oct	Dec	Feb	Apr	Jun	Aug	Oct	Dec	Feb	Apr	Jun	Aug	Oct	Dec	Feb
1(a) Mapping	<-	-	+	-	->	?
1(b) Profile	<+	->
2(a) Tree and Forest	.	.	.	<-	.	.	+	+	.	->	.	.
2(b) Primary Consumer	<+	+	.	>	.	.
2(c) Primary Production	<+	+	+	+	+	+	+	+	+	+	>
2(d) Primary Consumption	.	<-	.	+	.	.	+	+	.	->	.	.
3(a) Seedling Recruitment and Demography	<-	+	.	.	+	.
3(b) Seedling Growth	.	<-	+	.	.	.	+	+	.	+	.	.
3(c) Forest Gap Primary Consumer	<+	+	.	+	.	.

The studies in section two will examine surviving biota, particularly the major primary producers and consumers. Studies of primary production focus on litter fall, shoot observations, and trunk measurements of *Rhizophora mangle*. The impact of oiling will be assessed by comparing gross estimates of production and phenology for oiled and unoled sites in the study area. Additional parameters of growth will also be examined. For instance, Gill and Tomlinson (1971) reported that different leaf production rates in mature trees corresponded with different kinds of axillary growth, ranging from leaves only, to leaves and reproductive parts, to leaves and vegetative shoots. Therefore, if oil has altered leaf production rates, it may also have changed tree canopy structure and reproductive success. Studies of primary consumption will focus on canopy herbivory and removal of fallen leaves and fruit from the forest floor by crabs. These studies will identify where the major herbivores occur in these mangrove forests, although estimates of abundance based on quadrat counts are expected to underestimate actual numbers because of the difficulty in counting and capturing crabs among mangrove roots. Instead, estimates of relative population densities based on leaf removal rates may be used (e.g., Robertson 1986).

Section three looks at recovering biota in areas where mature trees were killed. Studies include recruitment by mangrove trees, seedling growth, and censuses of primary consumers. This study will examine natural recruitment, but it will also include an assessment of the Refinería Panamá planting of 86,000 seedlings (Teas *et al.* 1989). Refinery personnel will be consulted, where appropriate. Surprisingly, they apparently made no assessment of natural recruits, and without this information, it is impossible to determine the success and future value of their planting effort. In

addition, some seedlings appear to have survived the oiling, which killed surrounding adults. This novel finding will be looked at closely during this study. Furthermore, since a method to age seedlings has been discovered, a more detailed assessment of seedling demography and growth will be made. This study will also partly quantify primary consumption in sites of deforestation. In this way, comparisons will be made of relative numbers of crabs between oiled and unoiled forest gaps and between unoiled mature forests and oil-deforested gaps. In the latter case, this procedure will provide the best quantification of animals lost because of oiling. Furthermore, successive monitoring will reveal rates of recovery, if any.

In assessing the structural character of forests, any determination of tree age would be extremely beneficial. However, aging mature trees by using their physical characteristics is not possible at present. For instance, tree biomass measurements and trunk-wood ring counts have proven to be unsatisfactory toward aging mangrove trees (Tomlinson 1986). In further consideration of this question, evidence will be gathered from the various studies, notably from mapping, primary productivity and seedling growth, in an attempt to identify seedlings or trees of known age, and to describe age-related parameters for them. Most efforts will be focused on one species, *R. mangle*, since it is the one commonly missing from the deforested areas.

However, there are at least three other species in the study area and these are included in the studies, as appropriate (Table 6.4). For example, seedlings of non-*Rhizophora* species are common in some deforested areas previously monotypic for *Rhizophora*. This suggests that oil-deforestation gaps may alter forest succession patterns by allowing the introduction of species previously denied access for one reason or another. One possibility is that crab predation of mangrove propagules (Smith *et al.* 1989) was disrupted by oiling, which caused a reduction in crab numbers. Dr. Smith collaborated in an attempt to test this hypothesis.

6.4.1 Mapping Study

Objectives

The main objective of the mapping study is to delineate the extent of deforestation in mangrove forests in and near the oil spill area (see Table 6.1). Secondary objectives include quantification of general changes in mangrove forest boundaries, aging of trees in certain extended mangrove forest fringes, quantification of changes in watershed vegetation cover, observations on sediment discharge and current flow patterns, observations on extent and success of the Refinería Panamá planting experiment (Teas *et al.* 1989), and comparison of changes in associated habitats (coral reefs, reef flats, and seagrass beds).

Table 6.4 Mangrove tree species in the vicinity of the Refinería Panamá oil spill, notably in tidal forests from Isla Margarita, Bahía Las Minas, to Isla Grande. Species are ordered by dominance in estuarine fringing habitats, where most oil-deforestation occurred. These range from fringing forests behind coral reef lagoons toward the mouth of estuaries to those along major tidal channels and more riverine tidal channels.

Species	Dominance	Habitat occurrence
<i>Rhizophora mangle</i>	dominant	all habitats
<i>Laguncularia racemosa</i>	common	all habitats
<i>Avicennia germinans</i>	less common	all habitats
<i>Pelliciera rhizophorae</i>	uncommon	only riverine habitats

Methodology

The study area is defined as the coastal margin from Río Chagres in the southwest to Isla Grande in the northeast, near the Atlantic mouth of the Panama Canal (Figure 6.1). Existing aerial photography of this area will be obtained from government mapping agencies, such as the Instituto Geográfico Nacional "Tommy Guardia" of the Republic of Panama. Other remote sensing media were assessed and found to be less suitable, considering image definition and cost. Selected photographs and field observations will be used to draw vegetation boundaries for mangroves. These outlines will be combined with accurate map images into final vegetation maps. The mapping will require the assistance of a map drafting agency.

6.4.2 Profile Study

Objectives

The purpose of the profile study is to define topographic profiles in a range of mangrove habitats, noting where trees died and oil accumulated (see Table 6.1). Further objectives include quantification of profile shapes and their correspondence with associated physical and biological factors, and quantification of tidal range and inundation frequency in specific sites within the study area.

Methodology

Profile transects will be chosen at sites of biotic studies referred to in Sections 2 and 3 (Table 6.1). The transects will be approximately perpendicular to tidal isohytes, minimizing their length. It is the intent of this study to define accurately areas of deforestation and oiling, not the entire intertidal profile. Reference datum points will be established with respect to the Galeta Bench Mark, from which accurate referral can be made to the local port of Cristóbal, adjacent to Colón, Atlantic mouth of the Panama Canal.

6.4.3 Tree and Forest Study

Objectives

The tree and forest study will define tree species of mangrove forests in Bahía Las Minas and nearby and describe structural characteristics of the forests (see Table 6.1). Secondary objectives include assessment of the comparability of these species with those in other areas, description of demographic patterns for mangrove forests of Bahía Las Minas, development of a method to age mangrove trees, and establishment of long-term study sites assessing changes in structure and composition.

Methodology

Species descriptions are based on reference materials and the literature. A reference herbarium of voucher specimens gathered during the study will be sent to appropriate regional herbaria and the U.S. National Museum of Natural History at the completion of the project. Data scored within forest plots chiefly include species, height, girth, and inter-tree distance. Other methodologies for characterizing forest structure will be used as appropriate. For example, determination of basal area by point scanning may not be possible in more tangled mangrove areas where trunks are twisted, rather than straight and vertical. Furthermore, trunks in these areas may not be singular, and there is the additional problem of aerial roots from the upper canopy. Changes will be monitored during the course of this study at six-or twelve-month intervals. However, because longer term observations will be necessary to follow full recovery, permanent markers will be installed at all sites.

6.4.4 Primary Consumer Study

Objectives

The main objective of the primary consumer study is to identify the major primary consumers in mangrove forests, particularly canopy herbivores and litter (chiefly leaves and fruits) and wood consumers (see Table 6.1). The secondary

objective is to define specific distributional ranges of certain species across the intertidal zone and between major estuarine habitats in the study area.

Methodology

Quadrat sampling (with three replicates) will be conducted at selected intervals along profile transects (Chapter 6.4.2). The size and number have not yet been determined because these will depend on field trials roughly establishing population densities.

6.4.5 Primary Production Study

Objectives

The primary production study quantifies major components of primary production by mangrove trees (chiefly *Rhizophora mangle*) and show their phenology (see Table 6.1). Secondary objectives include quantification of reproductive success in seasonal cycles of the canopy, assessment of morphological changes in leaves and propagules because of oiling, and quantification of leaf longevity and turnover.

Methodology

Preliminary observations showed that *Rhizophora mangle* was the main mangrove species affected by oil-deforestation. Primary production in the forest canopy of this species is studied using two techniques, including litter fall collection and shoot observations. At most sites (Table 6.2), three litter traps (each 1 m²) were suspended under the canopy, above the highest tide, alongside 21 tagged leafy shoots positioned in the upper 1-2 m of canopy. Collection and monitoring are approximately monthly and will continue for at least 18 months, having started in June 1989 (Table 6.3). Contents from litter traps are sorted (and counted, as appropriate) by component (leaves, interpetiolar stipules, twigs and bark, and reproductive parts) and species (Table 6.4), then dried for at least three days at 80°C and weighed. For shoots, scores are made of structural status in relation to a fixed reference point within the shoot. This method will provide monthly measures of leaf number per shoot (a conveniently quantifiable entity in *Rhizophora* species), new leaves, falling leaves, axillary shoots, and status of reproductive parts. These procedures are well established from studies in Australia (e.g., Duke *et al.* 1984; Duke 1988). Another aspect of primary production, concerning trunk wood growth in mature trees, will be monitored by marking outer wood layers with dye so that additional layers can be measured after about two years. An attempt will also be made to re-assess tree girths measured by Rabinowitz (1975). These measurements may provide estimates of longer term trunk growth in this region.

6.4.6 Primary Consumption Study

Objectives

The main purpose of the primary consumption study is to quantify primary consumption of canopy leaves, litter fall, and wood (see Table 6.1). A secondary objective is to investigate the relationship between crab predation of propagules and tree species composition of mangrove forests.

Methodology

At least three sub-studies will be conducted. First, canopy herbivory will be assessed by quantification of leaf loss in the canopy (Robertson and Duke 1987). Exclusion devices will be installed in the field to help determine respective proportions removed by arboreal crabs (Beever *et al.* 1979) and insects. In a pilot study, excluders were installed in July 1989; monitoring at six-month intervals will commence next year (Table 6.3). The second sub-study will assess litter fall consumption on the forest floor by tethering various components (leaves or propagules) with one meter strings tied to roots. These will be laid in replicate sets of quadrats where concurrent litter fall traps will monitor day-to-day fall rates. In this way, numbers of tethered components will reflect actual falls. Each consumption session will last about five hours, determined by the period of low tide and forest floor exposure. In Australia, Robertson (1986) has shown that these consumers do not move about when the forest floor is submerged; crabs close their burrows prior to flooding. The third sub-study will score wood consumption by determining periodic losses in volume and weight of wooden blocks. These will be sawn from mangrove trees (chiefly *Rhizophora mangle*) to a standard dimension, weighed, and then tethered on the forest floor. Recovery of blocks is planned at six-month intervals for up to two years.

6.4.7 Recruitment and Seedling Demography Study

Objectives

The main objective of the recruitment and seedling demography study is to determine patterns of natural recruitment and the affect of oiling (see Table 6.1). Secondary objectives include plotting recruitment across the intertidal profile and between habitats, and assessment of potentially beneficial silvicultural practices following similar mangrove deforestation.

Methodology

Replicated quadrats will be established in areas of oil-deforestation and cleared unoiled control sites. These latter sites may not all correspond with the

standard monitoring sites of this project. They will also be about the same age as oil-deforested sites. In all quadrats, existing seedlings will be permanently marked. Plots will then be monitored annually for new recruits until the completion of the project. The technique to age seedlings allowed this study to be expanded considerably into a study of seedling demography.

6.4.8 Seedling Growth Study

Objectives

The main objective of the seedling growth study is to determine the natural rate of seedling growth and seasonal changes in oiled and unoled forest gap sites (see Table 6.1). Secondary purposes include finding methods to age seedlings, comparing seedling growth with that in mature trees (notably with shoot observations; Chapter 6.4.5), and further assessment of potentially beneficial silvicultural practices following mangrove deforestation.

Methodology

Seedlings were tagged and measured between September 1989 and January 1990. Their growth will be monitored for the duration of the project at six-month intervals (Table 6.3). Parameters scored include not only standard measures, such as tree height, but also several used in shoot observations (Chapter 6.4.5). Tagged individuals include a range of existing plants, from the smallest to the largest. These will be important in assessing age by a technique discovered when examining seedlings planted by C. Getter and C. González. Furthermore, such findings will also provide the basis for assessing the success of the extensive plantings by Refinería Panamá personnel; collaboration with staff from the refinery will be sought (Chapter 6.4.7).

6.4.9 Forest Gap Primary Consumer Study

Objectives

The main purpose of the forest gap primary consumer study is to determine the type and relative abundances of primary consumers in oiled and unoled deforestation gaps (see Table 6.1). Secondary objectives include comparison with mature tree forests and primary consumption in gaps (e.g., herbivory of seedling leaves, or leaf litter removal).

Methodology

Distribution and relative abundances will be monitored by techniques similar to those developed for the primary consumer study of mature trees (Chapter 6.4.4).

Timing also will be the same (Table 6.3). Similarly, monitoring of consumption will follow the earlier study (Chapter 6.4.6), as appropriate, and as time permits.

6.5 Studies in Progress

Major field work commenced in June 1989. By November, four main studies were in progress. The status of each is briefly noted for the respective studies listed in Table 6.1.

6.5.1 Plan Mapping

A. Aerial photography (black and white) for the study area has been obtained for the years available from 1966 to 1987. The scale and quality of this photography vary considerably but, overall, the pre-spill coverage is adequate. However, additional post-spill material is required.

B. New aerial photography is being obtained through the Instituto Geográfico Nacional "Tommy Guardia" of the Republic of Panama.

C. Drafting of vegetation maps from traced outlines can also be done with the Instituto Geográfico Nacional, but this will not be pursued formally until the new aerial photography has been completed.

6.5.2 Primary Productivity

A. Initial surveys (field and literature) of all earlier and on-going mangrove-related studies in the study area around Bahía Las Minas were completed in May 1989. These efforts culminated in the discovery of two additional mangrove species for the area. One, *Nypa fruticans*, was previously unknown in the neotropics (Duke, prep.). It was probably introduced forty or fifty years ago and occurs in just one location outside the influence of the 1986 oiling. The other species, *Pelliciera rhizophorae*, occurs in sites of greater freshwater influence. However, while it does occur within Bahía Las Minas, the sites where it occurs were not reached by the spill. In addition, there are three other common mangrove species in the study area (Table 6.4). However, just one, *Rhizophora mangle*, received the major impact of oiling in areas of deforestation. Therefore, most studies of primary production and seedling growth are based on it.

B. Twenty-one leafy shoots of *Rhizophora mangle* were tagged on single trees at 26 separate sites (the two treatments and three habitats) in June 1989. A second overlapping sub-study started in August; twenty-one shoots were tagged on three separate trees at each of six sites (the two treatments). Shoots were generally chosen within the top 2 m of canopy, and the distance for each one was measured from the top (Table 6.5). All have been monitored monthly. A small number of shoots were lost due to natural causes and immediately replaced.

Table 6.5 Height frequency table for leafy shoots (n=21) measured from the top of the canopy in 26 sites. Site descriptions (OO, OU, etc.) listed according to codes for habitat and treatment in Table 6.2.

Habitat/ Treatment	Site n.	Height Classes (m, <=)						
		-0.5	-1.0	-1.5	-2.0	-2.5	-3.0	-3.5
OO	1	10	8	3				
	2	7	12	2				
	3A	15	6					
	3B	5	7	5	4			
	3C	15	1	5				
	4	12	6	3				
OU	5	13	6	2				
	6	4	11	2	4			
	7A	10	4	7				
	7B	6	5	7	3			
	7C	6	5	8	2			
	8	5	9	5	2			
CO	9	11	9	1				
	10	2	12	7				
	11	6	6	3	6			
	12A	4	13	3	1			
	12B	12	3	4	2			
	12C	8	9	4				
CU	13	6	6	9				
	14	5	8	8				
	15	1	8	8	3	-	-	1
	16	12	6	3				
	17	9	11	1				
	18A	1	12	8				
RO	18B	10	7	4				
	18C	8	6	2	5			
	19A	8	4	4	4	1		
	19B	8	11	2				
	19C	4	6	8	3			
	20	1	9	11				
RU	21	1	12	8				
	22	2	11	8				
	23A	8	10	2	1			
	23B	8	9	4				
	23C	3	9	6	3			
	24	6	7	8				
	25	4	12	3	2			
	26	6	8	5	2			

C. Three litter fall traps, each 1 m² square, were installed under mature *Rhizophora mangle* trees in each of 25 sites (one was too exposed to theft), corresponding with the shoot sub-study, above. Seventy-two traps were installed in June 1989 and litter was gathered each month for most traps. Theft of traps has meant that the full array will never be possible (numbers were as low as 55 in September 1989), but selective replacement has enabled work to continue with at least one trap in all but a few sites. Some gaps will be inevitable in these data. In October 1989, there were 58 traps collected; none was lost. All litter returned to the laboratory has been sorted, dried and weighed between monthly collections.

D. Surface salinity and temperature of water fronting each site are monitored each month when litter is collected and shoots scored.

6.5.3 Seedling Growth

A. A total of seventy seedlings of *Rhizophora mangle* planted in six sites approximately two and three years ago were tagged and measured in August and September 1989. Sites were grouped according to their two main characteristics, namely, closed canopy/unoiled and open canopy/oiled.

B. Twenty-one seedlings of *Rhizophora mangle* in each of six sites were tagged with long-term markers of stainless steel wire stuck through the vertical stem. Monitoring started in September 1989, and another twelve sites will be set-up.

6.5.4 Primary Consumption

A. In a pilot study, excluders for crawling herbivores were installed on three separate branches of *Rhizophora mangle* in 25 sites in the vicinity of previously tagged shoots and litter traps. Excluders consisted of 20 cm plastic plates, cut, stapled and glued back together around branch stems up to 2 cm diameter. Branches were selected with eight to ten shoots, mostly, and other branches were trimmed to form "moats" around experimental branches. Data on leaf herbivory will be collected after complete canopy turnover has taken place.

B. T.J. Smith (visiting STRI short-term fellow) collaborated on a study of crab predation of mangrove propagules during November and December 1989. The study objectives include comparisons of crab predation in oiled and unoiled sites.

6.6 Preliminary Conclusions

6.6.1 Canopy Growth of Surviving Adult Trees in Oiled Areas is Similar to Growth in Nearby Unoiled Areas

The observation that canopy growth of adult trees in oiled areas is similar to growth in nearby unoiled areas suggests that a continuing presence of residual oil in the substratum around survivors no longer directly threatens these trees. However, survivors now find themselves more exposed and generally unsupported by their

missing neighbors. For this reason, healthy but exposed trees from time to time fall over and die during periods of strong winds.

Initial estimates of litter fall productivity for *Rhizophora mangle* in Bahía Las Minas (Table 6.6) are comparable to measurements made elsewhere for *R. mangle* and other *Rhizophora* species (Woodroffe 1982). Thus, for all sites, the average total litter fall production is conservatively $7.60 \text{ t}\cdot\text{ha}^{-1}\cdot\text{yr}^{-1}$, with around 66.6% leaves. There do not appear to be any consistent trends across treatments and habitats. However, the data will be more fully assessed after data for one complete year have been collected and compiled. This step is necessary because of the considerable seasonal variation expected (Duke *et al.* 1984).

Shoot studies (Figure 6.4) show some of this seasonal change in both the standing stock parameter of leaves per shoot and the rates of leaf production and fall. This figure also demonstrates that there are important differences between habitats, and that combined with these different locations, unoiled and oiled sites are not distinguishable. This conclusion also applies to net canopy production (difference between production and fall rates).

Table 6.6 Litter fall productivity of leaves, stipules, wood, reproductive parts and totals for *Rhizophora mangle* mature trees. Determination made in oiled and unoiled sites of three fringe habitats through July to September 1989. Component amounts expressed as mean weights (in g) per m^2 per day.

	Open		Channel		River	
	Unoiled mean(1 se)	Oiled mean(1 se)	Unoiled mean(1 se)	Oiled mean(1 se)	Unoiled mean(1 se)	Oiled mean(1 se)
July-September 1989						
Leaves	1.37 (0.06)	1.02 (0.09)	1.72 (0.14)	1.28 (0.16)	1.43 (0.17)	1.50 (0.15)
Stipules	0.24 (0.02)	0.22 (0.02)	0.28 (0.02)	0.20 (0.03)	0.20 (0.02)	0.20 (0.02)
Wood	0.08 (0.02)	0.08 (0.04)	0.09 (0.03)	0.19 (0.06)	0.06 (0.02)	0.10 (0.03)
Reprod.	0.24 (0.03)	0.23 (0.03)	0.65 (0.09)	0.18 (0.03)	0.46 (0.08)	0.48 (0.10)
Total	1.92 (0.07)	1.55 (0.15)	2.74 (0.21)	1.85 (0.23)	2.15 (0.25)	2.28 (0.23)

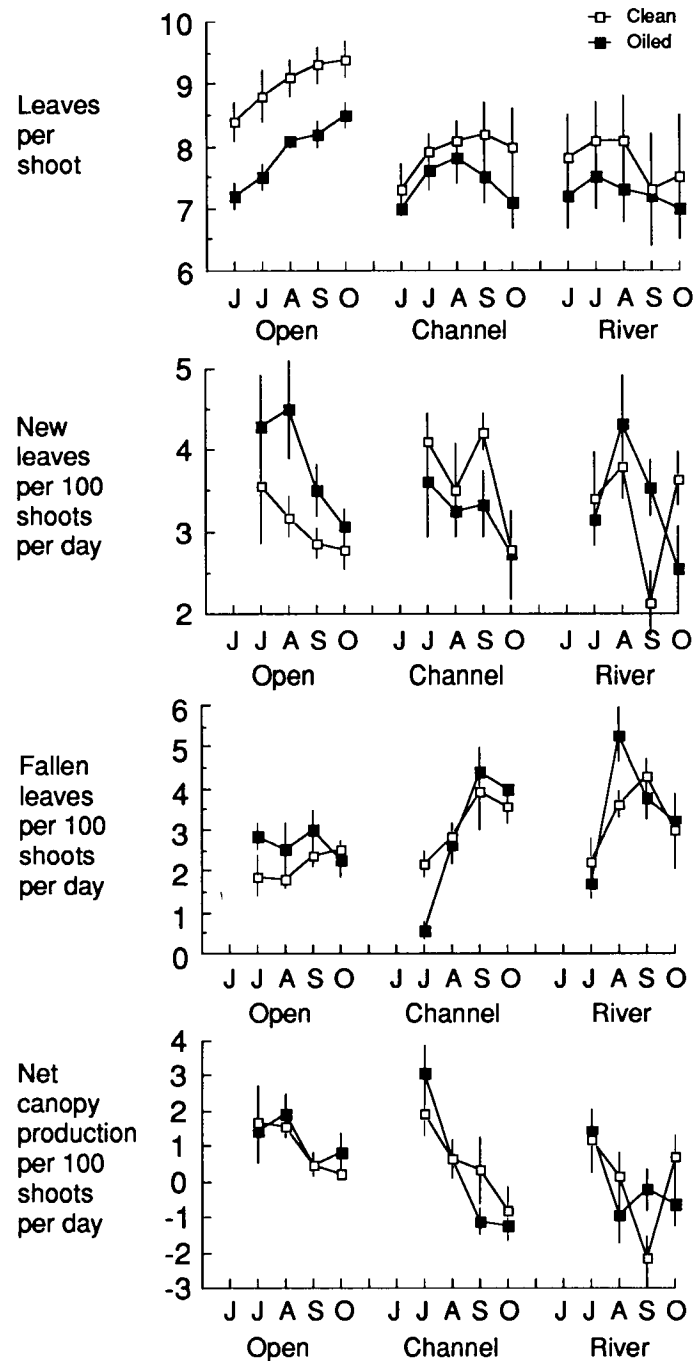


Figure 6.4 Standing stock leaves per shoot, new leaf appearance, fallen leaves and net leaf production (± 1 se) in shoots of *Rhizophora mangle* mature trees. Determination from oiled and unoled sites of three fringe habitats through June to October 1989. Rate units expressed as mean numbers of leaves per 100 shoots per day.

Results from litter and shoot studies may be combined to provide two other standing stock estimates. In Table 6.7, using results from July 1989 as an example, canopy density estimates of shoots and leaves per m² are presented. Again, there are no consistent trends across treatments and habitats; these estimates will be better evaluated using a complete year of data.

6.6.2 Seedlings (1-2 Years Old) Survived Oiling while Surrounding Adults Died

One finding, for which there are several comments, is that one- to two-year-old seedlings survived the oiling while surrounding adults died. Such seedling survival has implications concerning recovery of deforested mangroves after oil spills and the need for human intervention. First, it appears that somehow young seedling structure (perhaps the lack of prop roots) enabled individuals to tolerate periods of immersion in oil. Second, destructive preparation of the substratum prior to artificial planting would remove such survivors, setting back forest recovery. The value of artificial planting relies on the assumption that planted recruits will outperform natural ones, irrespective of when they arrive. Note that Teas *et al.* (1989) make little or no reference to natural recruits, either by recognizing their presence when planting, or by marking planted seedlings so that future natural recruits could be easily distinguished in the field. These omissions make it difficult to say whether planting seedlings has facilitated the recovery of these forests.

To measure recruitment and evaluate whether artificial planting enhances forest recovery, seedling growth will be monitored closely. The primary objective will be to establish age-related parameters that will enable a detailed demographic study for recruitment in natural- and oil-deforested sites. We recently determined that aging seedlings is possible (Duke and Pinzón, in prep.). Furthermore, age is relatively easily estimated once growth rates have been determined for a particular site. The best parameter is the number of leaf scar nodes on the seedling stem. This equates to leaf production rate, and curiously this rate appears to be about the same in shoots of small seedlings and of large mature trees.

In Figure 6.5, two graphs are presented comparing seedling parameters of vertical height and vertical node number versus time since planting. These results, although preliminary, show how the regressions for node numbers are better predictors of age than vertical height. The two categories demonstrate the importance of light, and the closed-canopy group represents perhaps the slowest sustainable rate. In addition, the correlation appears to be linear through all age classes, shown by the corresponding estimates for mature tree shoots. Age, in this case, represents the period of observation. The slope of the linear equations equates to node production per month, and may be used to calculate values of node production per year and leaf production rates (Table 6.8). These values for open-canopy seedlings are comparable with upper-canopy shoots (Table 6.9). Furthermore, shoot growth in Florida (Gill and Tomlinson 1971) was remarkably

Table 6.7 Estimates of canopy density during July 1989. Determined as shoots $\cdot\text{m}^{-2}$ and leaves $\cdot\text{m}^{-2}$, derived from rates of new leaf appearance, loss of leaves and net change (units) in shoots and litter fall of *Rhizophora mangle* mature trees in oiled and non-oiled sites of three fringe habitats. Values of leaf and stipule mean dry weights from litter fall are included for comparison.

	Open		Channel		River	
	Unooled mean(1 se)	Oiled mean(1 se)	Unooled mean(1 se)	Oiled mean(1 se)	Unooled mean(1 se)	Oiled mean(1 se)
Shoot Rates (units.100 shoots⁻¹.day⁻¹)						
New	3.56 (0.71)	4.29 (0.62)	4.11 (0.35)	3.63 (0.70)	3.41 (0.58)	3.14 (0.25)
Lost	1.88 (0.49)	2.84 (0.31)	2.18 (0.32)	0.56 (0.21)	2.23 (0.54)	1.69 (0.33)
Change	1.68 (1.07)	1.45 (0.92)	1.94 (0.57)	3.07 (0.82)	1.18 (0.90)	1.45 (0.39)
Litter Rates (units.m⁻².day⁻¹)						
New	3.81 (0.39)	4.80 (0.79)	5.72 (0.49)	3.67 (0.99)	5.46 (0.54)	5.45 (0.96)
Lost	2.90 (0.18)	2.27 (0.48)	3.00 (0.25)	1.58 (0.32)	3.25 (0.35)	0.32 (0.29)
Change	0.99 (0.33)	2.53 (0.35)	2.73 (0.36)	2.09 (0.86)	2.21 (0.79)	3.13 (0.97)
Shoot Density (shoots.m⁻²)						
New	107.0	111.9	139.2	101.1	160.1	173.6
Lost	54.3	79.9	137.6	282.1	145.7	137.3
Mean	130.7	95.9	138.4	191.6	152.9	155.5
Leaf Density (leaves.m⁻²)						
Mean	1124	705	1052	1399	1216	1143
Mean Weights (dry weight in g)						
Leaves	0.50 (0.03)	0.53 (0.04)	0.58 (0.04)	0.64 (0.02)	0.57 (0.04)	0.59 (0.02)
Stipules	0.08 (0.00)	0.07 (0.01)	0.07 (0.01)	0.08 (0.01)	0.06 (0.01)	0.07 (0.01)

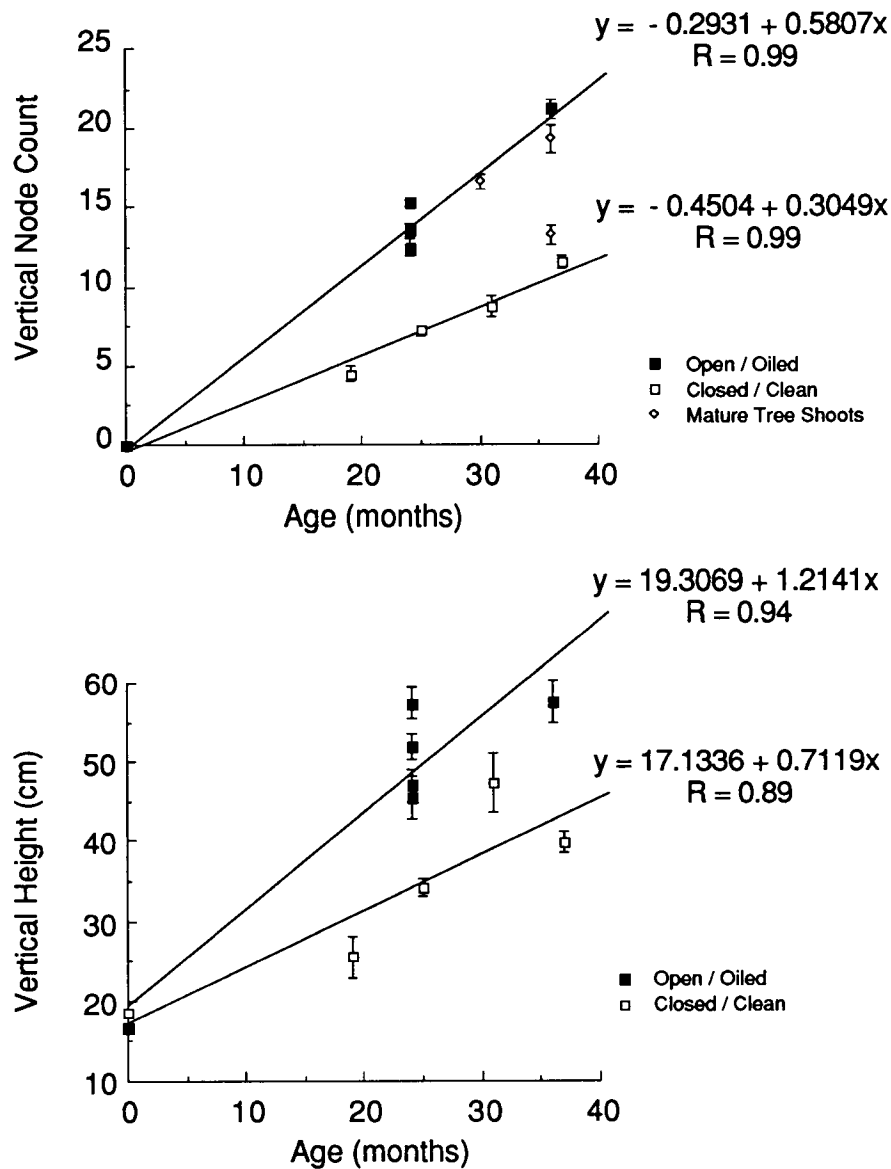


Figure 6.5 Age-related parameters in 95 planted seedlings of *Rhizophora mangle* in two major site groups. Groups are open canopy with oiled substratum and closed canopy with clean substratum. Generally, the six sites were assessed only once, but one was scored twice over a six month period. Furthermore, similar-age shoot node numbers for mature trees are plotted for comparison.

Table 6.8 Leaf production rates. Determined from node production rates for the two canopy conditions and oil treatments observed in *Rhizophora mangle* seedlings of known age (Figure 6.5). Compare with leaf production rates observed in mature trees (Table 6.9).

Canopy - Treatment	Node Production/ month	Node Production/ year	Leaf Production/ 100 Shoots/day
Closed - Unoiled	0.30	3.66	2.00
Open - Oiled	0.58	6.98	3.82

Table 6.9 New leaf appearance in shoots of *Rhizophora mangle* mature trees. Determination from oiled and clean sites of three fringe habitats through July to October 1989. Units expressed as mean numbers of leaves per 100 shoots per day.

	Open		Channel		River	
	Unoiled mean(1 se)	Oiled mean(1 se)	Unoiled mean(1 se)	Oiled mean(1 se)	Unoiled mean(1 se)	Oiled mean(1 se)
July-October 1989 - Mean						
New	3.10 (0.18)	3.85 (0.33)	3.66 (0.33)	3.24 (0.19)	3.24 (0.38)	3.39 (0.37)

similar, with the same range of mean rates from 1.97 to 3.92 leaves per 100 shoots per day.

These results have far-reaching implications for mangrove studies in general, but for this study they provide the means to quantify and compare longer term growth for trees and seedlings subjected to oiling.

Chapter 7

Effects of the April 1986 Oil Spill at Isla Payardi on the Epibiota of Mangrove (*Rhizophora mangle* L.) Roots

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7.1 Introduction

Mangrove-dominated shores are characteristic of tropical and subtropical coastlines worldwide (Chapman 1976). As development of tropical shores has increased, degradation of mangrove forests through clearing and other man-made changes has proceeded rapidly (Odum and Johannes 1975; Cintron and Schaeffer-Novelli 1983; Saenger *et al.* 1983). One growing threat to tropical coastlines is the risk of oil pollution caused by increases in tanker traffic. Knowledge of the effects of oil on mangroves and associated species is limited (Gundlach and Hayes 1978; Lewis 1983; National Research Council 1985).

The red mangrove *Rhizophora mangle* L. is widely distributed in the Caribbean and occurs extensively on both coasts of Panama (West 1977). It grows along much of the south coast of Florida (Odum *et al.* 1982; Gill and Tomlinson 1969) and is found patchily elsewhere along the southeastern coast of the United States. Prop roots of *Rhizophora* are breeding and nursery areas for many marine species and substrata for a diverse group of epibiotic organisms (Figure 7.1, e.g., Batista 1980; Perez and Victoria 1980; Sutherland 1980; Odum *et al.* 1982; Perry 1988). The surface of submerged roots is thus a living hard substratum available for the attachment of marine plants and animals.

In April 1986, at least 50,000 barrels of oil spilled into the sea from a ruptured storage tank at Isla Payardi on the central Caribbean coast of the Republic of Panama (Cubit *et al.* 1987, 1988a; Jackson *et al.* 1989). Oil washed into a large area of mangrove forest near the Smithsonian Tropical Research Institute's Galeta Laboratory. The epibiota of the roots of *R. mangle* had been sampled in 1981 and 1982, providing information before the spill. The epibiota of mangrove roots were resampled quarterly beginning in August 1986, including samples from areas that were oiled and that escaped oiling. We here report on results through May 1989.



Figure 7.1 Diagram of the structure of a red mangrove (*Rhizophora mangle*) tree. Cross sectional view, indicating muddy substratum (stippled), sea surface, and prop roots at various stages of growth. Inset: a small tree in isolation, showing prop roots entering the water.

7.2 Study Sites

Three intertidal zone habitats of the red mangrove, *Rhizophora mangle*, in the area around Isla Payardi include (1) trees fronting the open ocean, generally along the inner margins of fringing reef flats, (2) those along the banks of channels leading from the open sea, and of lagoons into which the channels lead, and (3) those in brackish water streams and man-made ditches that run from landward into lagoons or drain interior areas of mangrove forest.

The coverage of space on intertidal mangrove roots in the above three habitats (hereafter called open, channel and stream) was first surveyed during September - October 1981 and again in January and June 1982. In initial sampling, areas of shore (=sites) within each habitat were haphazardly chosen (Figure 7.2A). After the spill, additional sites were chosen to increase sample sizes and to include both oiled and unoiled areas.

Sampling in the first year after the spill (August 1986-May 1987) was concentrated in areas studied during 1981-1982 (Figure 7.2A). In August 1987, sites were relocated to sample representatively from the coastline affected by the spill and to increase distance between sites. Beginning in August 1987, at least four sites were sampled in each of the six treatment types (e.g., open, oiled). These changes are shown in Figure 7.2B. Site names are listed in Table 7.1 and details are discussed for each habitat type below. All oiled sites were haphazardly chosen within larger oiled areas. Initial definition of "oiled" vs. "unoiled" was necessarily subjective, based on extensive personal observations, percent cover and vertical extent of oil on roots and examination of aerial photographs of oil slicks. These definitions will be further refined by hydrocarbon analyses and examination of sentinel organisms (K. Burns, in preparation).

On the open coast, three points adjacent to the Galeta Laboratory were sampled as two sites (Punta Galeta Mangrove, Isla Mina Mangrove) in 1981-1982; these areas were subsequently oiled. In 1986-1987, oiled roots were also sampled from a fourth point adjacent to and south of the original sites (these data are included in Isla Mina Mangrove). No open, unoled areas could be found near Bahía Las Minas; unoled, control areas were located ~25 km east, at María Soto and Portobelo (Table 7.1, Figure 7.2A). After August 1987, three additional sites were followed on oiled open coastline and sampling was discontinued at Punta Galeta Mangrove. Two additional unoled sites were added east of Portobelo (Figure 7.2B).

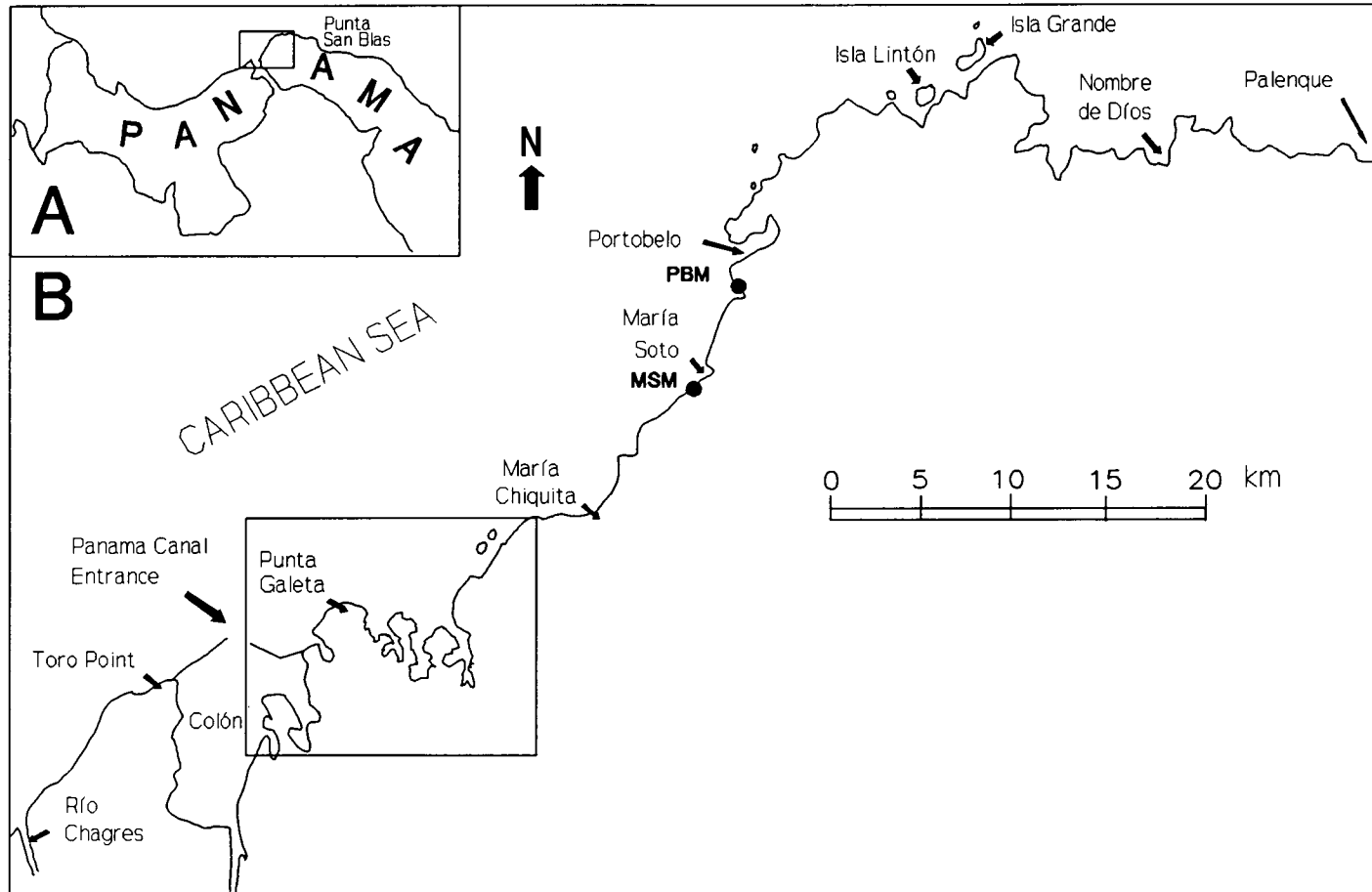


Figure 7.2 Study sites. A. Sites monitored during August 1986-May 1987. See Table 7.1 for explanations, habitat types, and monitoring history. See Figure 1.1 for further details.

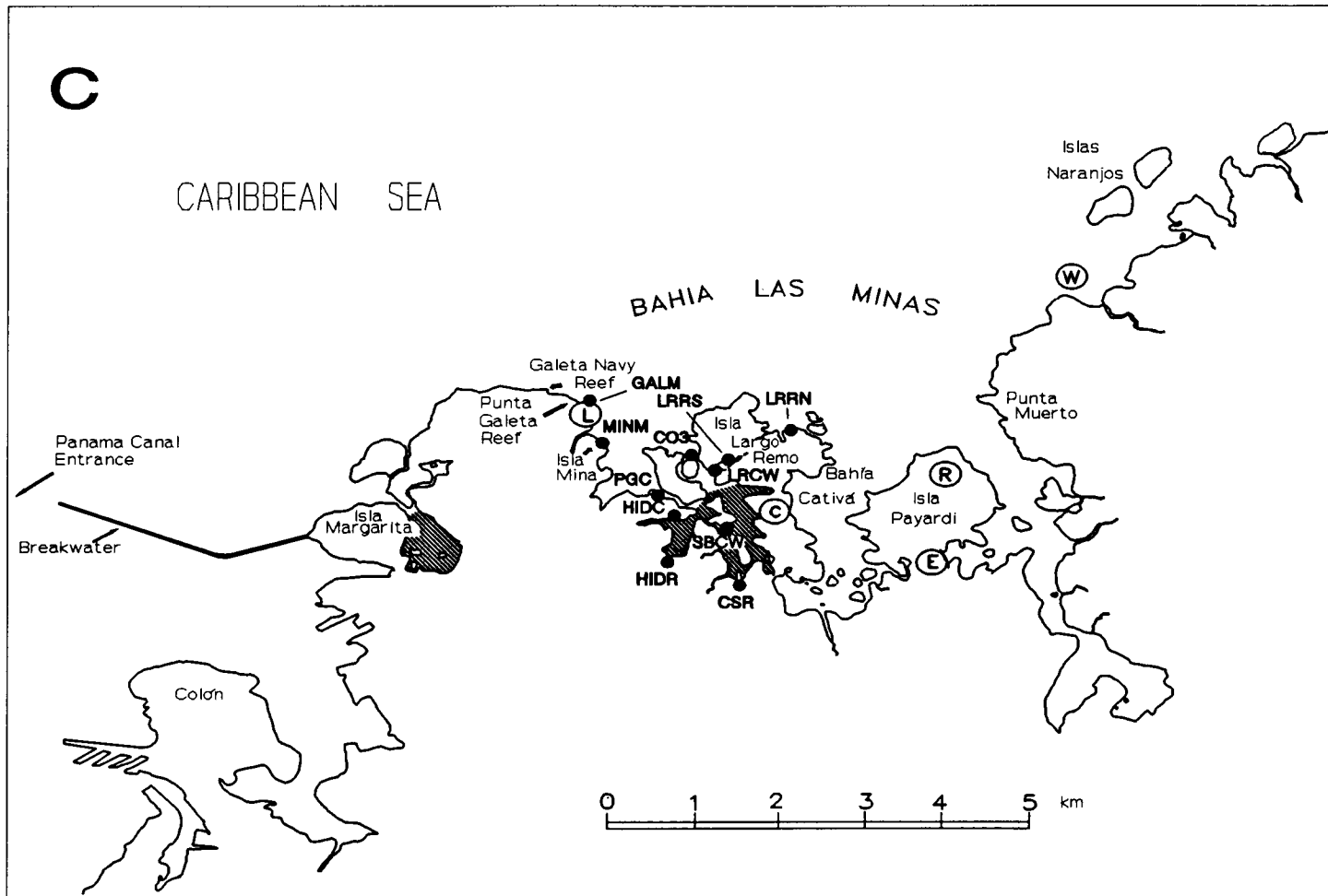


Figure 7.2 Study sites. A. Sites monitored during August 1986-May 1987 (continued).

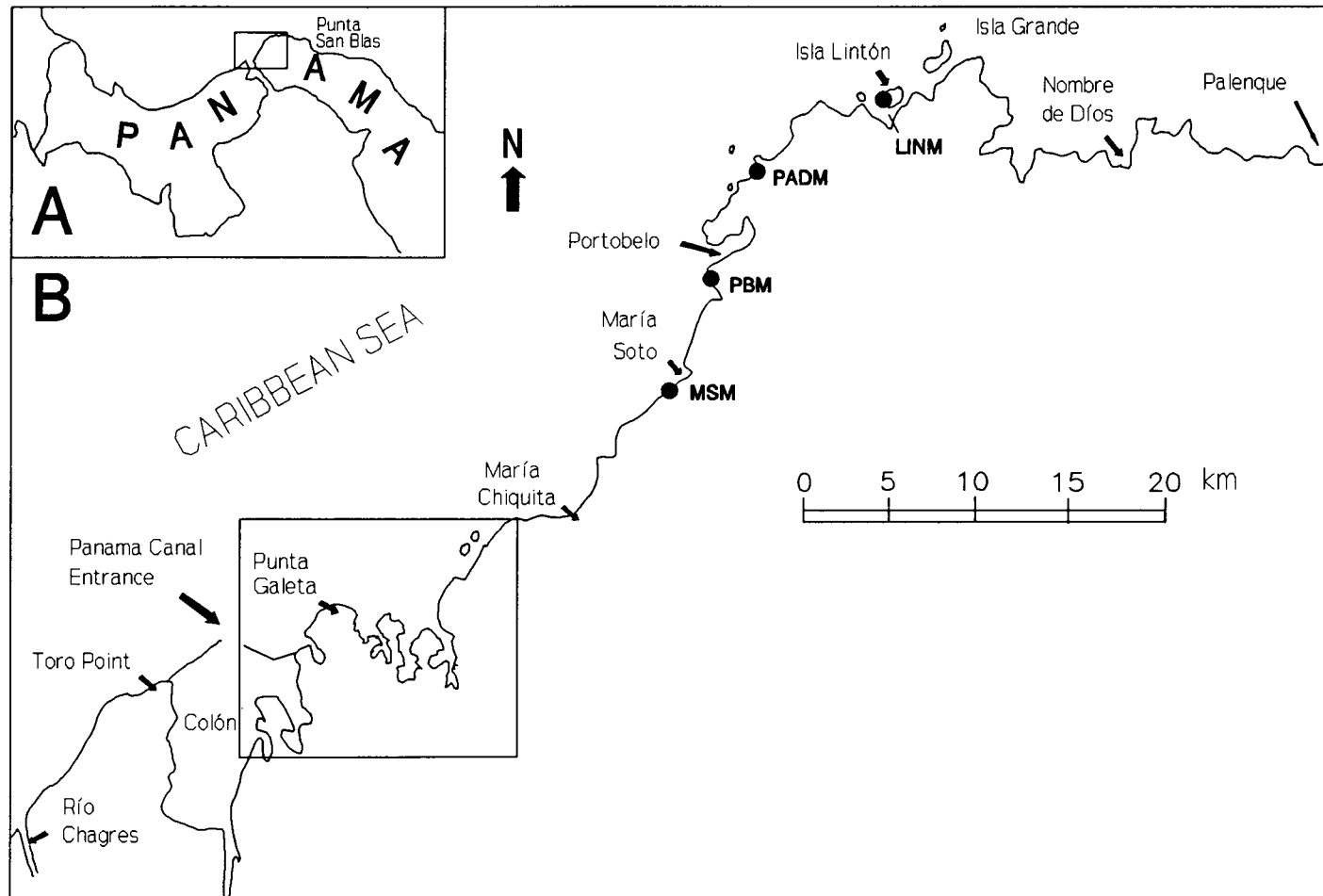


Figure 7.2 Study sites. B. Sites monitored from August 1987-present. See Table 7.1 for explanations of site abbreviations, habitat types, and monitoring history. See Figure 1.1 for further details.

Table 7.1 History of site monitoring and site names.

A. Unoiled Open Sites		
María Soto Mangrove (MSM)	8-86	-- present
Portobelo Mangrove (PBM)	8-86	-- present
Isla del Padre Mangrove (PADM)	8-87	-- present
Isla Lintón Mangrove (LINM)	8-87	-- present
B. Oiled Open Sites		
Punta Galeta Mangrove (GALM)	10-81	-- 8-87
Isla Mina Mangrove (MINM)	10-81	-- present
Peña Guapa Mangrove (PGM)	8-87	-- present
Isla Droque Mangrove (DROM)	8-87	-- present
Punta Muerto Mangrove (PMM)	8-87	-- present
C. Unoiled Channel Sites		
Hidden Channel (HIDC)	10-81	-- 5-88
Samba Bonita Channel West (SBCW)	8-86	-- present
Margarita Channel North (MACN)	8-87	-- present
Margarita Channel South (MACS)	8-87	-- present
Largo Remo Channel South (LRCS)	8-88	-- present
D. Oiled Channel Sites		
Peña Guapa Channel (PGC) *	8-86	-- 5-87
Largo Remo Channel West (LRCW)	8-86	-- present
Largo Remo Channel Site CO3 (CO3)	8-86	-- 5-87
Payardi Channel East (PCE)	8-87	-- present
Payardi Channel South (PCS)	8-87	-- present
Samba Bonita Channel East (SBCE)	8-87	-- present
Samba Bonita Channel South (SBCS)	8-87	-- present
E. Newly Oiled Channel Sites		
Hidden Channel (HIDC) **	8-88	-- present
F. Unoiled River Sites		
Hidden River (HIDR)	10-81	-- present
Río Coco Solo (CSR)	8-86	-- 5-87
Unnamed River (UNR)	8-87	-- present
Río Alejandro (ALER)	8-87	-- present
Quebrada Las Mercedes (MERR)	8-87	-- present
G. Oiled River Sites		
Largo Remo River South (LRRS)	6-82	-- present
Largo Remo River North (LRRN)	8-86	-- 5-87
Payardi River (PAYR)	8-87	-- present
Punta Muerto River East (PMRE)	8-87	-- present
Punta Muerto River West (PMRW)	8-87	-- present

Names are listed for each site; the abbreviation listed is that found in computer data files on deposit. Sites currently monitored will be followed for the remainder of the study.

* Sections of Peña Guapa Channel were monitored as Hidden Channel in 1981-1982

** Hidden Channel was oiled between May and August 1988. See text.

In channels and lagoons, two several-kilometer-long sections of shore were sampled in 1981-1982 (data combined as Hidden Channel). Oiling here was patchy; parts of the originally sampled area ranged from heavily oiled to unoiled (these unoiled areas are best viewed as being lightly oiled despite the absence of oil on roots, because of their proximity to oiled sites). Sections of shore in the lagoon system were added to both oiled (two sites) and unoiled (one site) sampling areas in July 1986 (Figure 7.2A, Table 7.1). These areas also should be considered moderately to heavily oiled vs. lightly oiled. In August 1987, two control sites were added where no oil had penetrated (Figure 7.2B) and four oiled sites were located in channels closer to the refinery (Figure 7.2B, Table 7.1). The additional unoiled sites, Margarita North and Margarita South, were in a separate lagoon that was protected from oiling by booms during the spill.

In August 1988, one control site, Hidden Channel, was secondarily oiled and is now considered newly (lightly) oiled. The source of the oil will be determined by hydrocarbon analysis. A replacement control site, Largo Remo Channel West, was added to the study at this time.

Two streams were sampled initially (Figure 7.2A, Table 7.1). One was subsequently heavily oiled (Largo Remo River South), while the other escaped oiling (Hidden River). After the spill, an additional unoiled stream (Río Coco Solo) was added and sampling extended throughout the length of the oiled stream. In August 1987, three oiled and three unoiled streams were added to the study (Figure 7.2B) and Río Coco Solo was removed because of cutting of its watershed by local fishermen.

7.3 Methods

7.3.1 Abundance of Epibiota

In 1981-1982, a starting point (tree) within each site was picked at random. From this point, 25 roots to the left and 25 roots to the right were chosen for sampling using a random numbers table. Roots that had not grown into the water and those firmly attached to the mud were rejected, and the nearest root that met the sampling criteria was sampled. Each selected root was lifted from the water and length from waterline to the longest root tip and diameter at the waterline were measured. Only roots ≥ 20 cm in length are included in this analysis, to make these samples comparable to those taken in 1986 and later. Percent cover of fouling and encrusting organisms was estimated visually.

Sampling was repeated in January 1982 and again in June 1982. For the latter survey, there are data for 25 roots each in channels and streams but for no roots on the open coast.

Mangrove roots were sampled again beginning in July-August 1986, roughly three months after the oil spill. Monitoring has continued quarterly at the sites described in the preceding section (Chapter 7.2). This sampling is designated "long-term census." Method of selection of roots is random, as in 1981-2. Roots attached

to the bottom or not extending 20 cm below the high tide line are rejected and the closest root sampled instead.

Several methods of determining percent cover were examined. Strings of varying lengths with 100 marked points in a stratified random array proved to be the most simple and accurate method and are now used exclusively. An appropriate-sized string is held against the root and what lies under each point is recorded. Organisms present but not found under a point are recorded as traces and assigned a percent cover of 0.01%.

As sampling progressed, defoliation of trees and deterioration of prop roots became evident. Beginning in February 1987, root condition was monitored. Random samples of roots in each habitat were selected and root condition recorded. In May and August 1987, data were collected only on roots sampled for percent cover, but were recorded separately. After November 1987, root condition was recorded along with percent cover for each root. A three letter code separated roots into two age categories: old (entered the water prior to the spill) and new (entered the water after the spill). The physical condition of roots was also recorded. This included the number of actively growing tips, if any, and whether the root appeared live (firm, covered with bark) or dead (spongy, with partial bark loss, and visible cover of fungus, bacteria and/or teredo tubes).

Beginning in August 1987, the focus of the study was widened and two additional types of data were collected quarterly. First, we began investigating the development of assemblages of organisms on mangrove roots from the time roots enter the water. A cohort of roots that were just about to enter the water was marked with flagging tape. Unlike the randomly chosen long-term census roots, all these roots entered the water well after (~16 months) the oil spill. Five randomly chosen roots were measured and sampled for percent cover and root condition at each site during each monitoring. Beginning in May 1988, sample size was increased to 10 roots per site.

Second, patterns of recruitment are being followed using artificial roots. Five one-half inch hardwood dowels ~90 cm in length are hung vertically in the water at each site and collected three months later. (In May, August and November 1987, sections of aerial roots were used in pilot experiments. Their use was then discontinued because of rotting and fungal growth). After collection, dowels are floated in seawater in the laboratory and examined under a magnifying light. Percent cover of organisms that have settled is estimated using nylon lines with 100 inked dots in a stratified random array. The number of sites used was variable until May 1988, when all 26 sites were included.

7.3.2 Species Identifications and Analyses

Species identifications are ongoing and current estimates of species richness are necessarily tentative, especially in groups such as sponges, tunicates, hydroids, and some foliose algae. When percent cover estimates of plants and animals are taken, unidentified taxa are assigned code names until they are identified. In addition,

considerable space may be occupied by categories: species that cannot be differentiated in the field (i.e., diatoms, mixed algal turfs, mixed arborescent bryozoans, hydroids, and crustose coralline algae).

Numbers of roots sampled in a given period varied among sites. Comparisons of algal species richness were made using rarefaction (Simberloff 1978). A sample size of 20 roots was chosen for comparisons.

7.3.3 Environmental Monitoring

The length and physical condition of each mangrove root monitored for percent cover is recorded quarterly (see above, 7.3.1). In addition, the following physical factors are being followed:

Salinity

Sites may differ in the salinity regime; salinity may also change over time. Two samples are collected, using scintillation counter vials, from the surface and from 1 m depth at each site every quarter. An optical refractometer is used in the laboratory to measure salinity. These measurements were initiated in November 1988.

Water Temperature

Sites may experience differences in water temperature; water temperature may also change over time. Temperatures are measured and recorded in the field using a thermometer attached to a meter-long stick. Two measurements are taken at each site quarterly while the stick floats on the surface, and two are taken while the stick is submerged to a depth of 1 m. These measurements were initiated in February 1989.

Depth from High Water Line to the Sediment

Sediment stability may differ among habitats and at oiled vs. unoiled sites. Fifty positions along the shore (anchored roots) were flagged at the high water line (HWL), and the vertical distance from each tag to the sediment was measured with a meter stick or a weighted meter tape. These measurements (a) give a depth profile for each site for comparative purposes and (b) will monitor accretion or erosion of sediment within sites. Depths are monitored yearly, starting in 1989.

Light Intensity

Light intensity is a measure of relative defoliation and may affect algal abundance on roots. At each site, on non-overcast days from 1000 - 1400, 20 pairs of light meter readings (in lux) are taken. Each pair consists of one reading out in the open, the other under the canopy where roots are monitored. The difference

between the two is an indicator of relative shading. Light intensity is monitored yearly (initiated in December 1988).

Water Movement

The amount of water flowing around roots may affect which organisms settle and survive on the roots; further, oiling has potential indirect effects on patterns of water flow. Clod cards (dental plaster half-spheres attached to galvanized metal plates) are set out subtidally, two per site, for several days, then collected (methods after H. M. Caffey, personal communication). Weight lost from the clod cards, measured in the laboratory, is related to the amount of water movement experienced (H. M. Caffey, personal communication). These measurements were initiated in 1989 and are conducted twice a year.

Sediment/core Analysis

As part of the Bermuda Biological Station for Research (BBSR) hydrocarbon monitoring, three 10 cm diameter by 30 cm depth core samples were collected from each site in May 1989. Detailed notes were taken in the field, and notes, drawings and photographs were made in the laboratory as cross sections at different depths in each core were taken for chemical analysis (J. MacPherson and K. Burns, personal communication). Data pertinent to the field study included presence of oil in each core (visible, by smell, or none) and substratum type (e.g., peat, mud, sand, rubble, coral fragments). Core sampling is to be repeated in July 1990.

7.4 Results

We here present data on (1) the physical characteristics of the open coast, channel and stream *Rhizophora* habitats, (2) the extent of oiling among habitats, including changes over time, (3) the direct and indirect effects of oiling on the health of *R. mangle* roots since the spill, and (4) changes in the abundances of the marine epibiota on prop roots in oiled and unoiled areas.

Most figures show results from each of the three parts of this study, and compare oiled and unoiled treatments. **Long-term census** results refer to the monitoring of abundances on randomly chosen roots that extend 20 cm or more downward from the high tide mark, but are not yet attached to bottom sediments. **Community development** results refer to monitoring of a cohort of roots marked in August 1987 as they entered the water and first randomly subsampled in November 1987. **Recruitment** results refer to artificial roots set out at each site for three months, then monitored for percent cover in the laboratory.

7.4.1 Physical Characteristics of the Habitat

Limited data are currently available on the physical characteristics of the 26 sites under study. Salinity and water temperature data are summarized in Table E.1. (Appendix). Site depths are shown in Table E.2 and sediment types in Table E.3. Monitoring of physical characteristics has been limited; supplemental information will be available from the Environmental Sciences Program at the Galeta Marine Laboratory (J. Cubit, personal communication; Chapter 10).

Mangroves on the open coast live in a habitat dominated by oceanic waters. Salinity has remained high at all sites, although it can drop occasionally during periods of heavy rains (Cubit *et al.* 1988b). Water temperatures show little evidence of depth stratification thus far and are similar at all sites. Site depths averaged 55-65 cm. Substrata at all open coast sites are dominated by sand and shell fragments, with lesser amounts of coral rubble, peat, inorganic mud, rock and calcareous algal fragments.

Channels and lagoons have had high salinities in the three samples available to date. In May 1989, surface salinity was lower than salinity at 1 m depth in seven of 10 samples, probably as a result of heavy rains. There has been little evidence to date of thermal stratification. Depths were highly variable and shallower than at open sites. The substratum was characterized by finer grained sediments: peat, coral rubble and organic and inorganic muds.

Physical characteristics of drainage streams were far more variable. Both surface and 1 m salinity readings in oiled streams have been high. In contrast, unoiled streams have had variable surface salinity and relatively high salinity at 1 m. Only Unnamed River has had surface salinity <15 o/oo in the three monitorings. Deeper waters have been more saline, only once dropping below 20 o/oo. We attribute this difference to the fact that streams with more freshwater drainage, deeper in the lagoons, were less likely to be oiled than those with less current flow and higher saltwater input. This is especially important given the time of the oil spill. In the early rainy season, stream flow increases when the rains begin and saltwater intrusions decrease. Streams that drain larger interior areas probably pick up freshwater inflow faster than those with more limited drainage. Thus, oiling was more likely in streams closer to the mouth of Bahía Las Minas and on Isla Largo Remo. There was also high variation in depth among unoiled streams. Streams were deeper than either channels or the open coast. Sediments were very fine grained, with inorganic muds and peat.

7.4.2 Extent of Oiling

Mangrove roots were first examined three to three-and-one-half months after the spill began. By then, oil had come ashore throughout the entire area around Isla Payardi and past Punta Galeta toward Colón. All areas of mangrove on the open coast between Isla Margarita and the Islas Naranjos were oiled to some degree. Mangrove channels and lagoons at Isla Margarita were protected by booms and

remained unoiled, and most of the back lagoons and channels between Isla Payardi and Punta Galeta were either unoiled or lightly oiled (Figure 7.2). Streams and rivers draining into this lagoon were unoiled, except for a stream draining Isla Largo Remo. The lagoon between Isla Payardi and Punta Muerto was heavily oiled, as was Bahía Cativá. In these bays, most small streams draining interior mangroves were heavily oiled, but larger streams escaped oiling (Figure 7.2B).

At the first monitoring after the spill, oil covered much of the area on prop roots at all oiled sites. A thick coat of diatoms covered much of this oil at channel and especially open sites. To estimate oiling, we measured the distance oil or oil and diatoms covered on a haphazardly selected sample of roots in streams, channels and on the open coast where oil had come ashore. At the oiled, open site, oil coated an average of 55.9 cm (range 36-70 cm, n=98) of the root surface. This was longer (146%) than the average length of roots sampled simultaneously for percent cover (mean = 38.4 cm, range 20-85 cm, n=80). In oiled channels, 37.6 cm (range 15-94 cm, n=50) was covered while average root length was 40.1 cm (range 20-111 cm, n=80, 94% of the surface covered). Along the oiled stream (Isla Largo Remo), oil averaged 29.0 cm (range 11-48 cm, n=49) on the roots; root length was 38.5 cm (range 20-92 cm, n=76, 75% of the root surface oiled). Thus, four months after the spill, wherever oiling had occurred, nearly the entire wetted surface of the prop roots of *R. mangle* had been oiled. As water levels rose and fell after the spill, layers of oil were successively deposited on root surfaces. Wave action on the open shore probably resulted in the increased coverage there relative to the other habitats.

Oil appeared as a category of percent cover on roots through May 1989, three years after the spill. We examined the proportion of roots (or dowels) with at least a trace of oil on their surfaces for the long-term census, community development, and recruitment studies (Figure 7.3). From August 1986 through May 1987, more than 90% of long-term census roots sampled from all three oiled habitats had at least a trace of oil on their surface (Figure 7.3, no data for open coast roots for August 1986, when diatoms obscured oil). The fraction of oiled roots has remained high, declining slowly to ~70% on the open coast and in channels and remaining >95% in streams through May 1989.

The actual amount of oil on these same roots showed a different pattern over time (Figure 7.4). Percent cover on long-term census roots at open sites peaked at 80% in February 1987, then declined rapidly to ~10-15%. In channels, oil percent cover was lower than on open coast roots, but more has persisted, averaging ~20-40% through 1988 and ~20% since. In streams, oil cover was almost 90% for the first year and has declined more slowly than in the other two habitats. In May 1989, oil covered an average of ~40% of root surfaces in streams.

Community development roots entered the water after August 1987, so any oil on their surfaces has come from reoiling since then (Figure 7.3). On the open coast, 40% of these roots were oiled within six months, and ~80% within nine months; since then the fraction of oiled roots has declined to ~40%. In channels, ~40% of all new roots were oiled within three months of entering the water, and oil has persisted (~55% oiled in May 1989). In contrast, all roots entering the water in

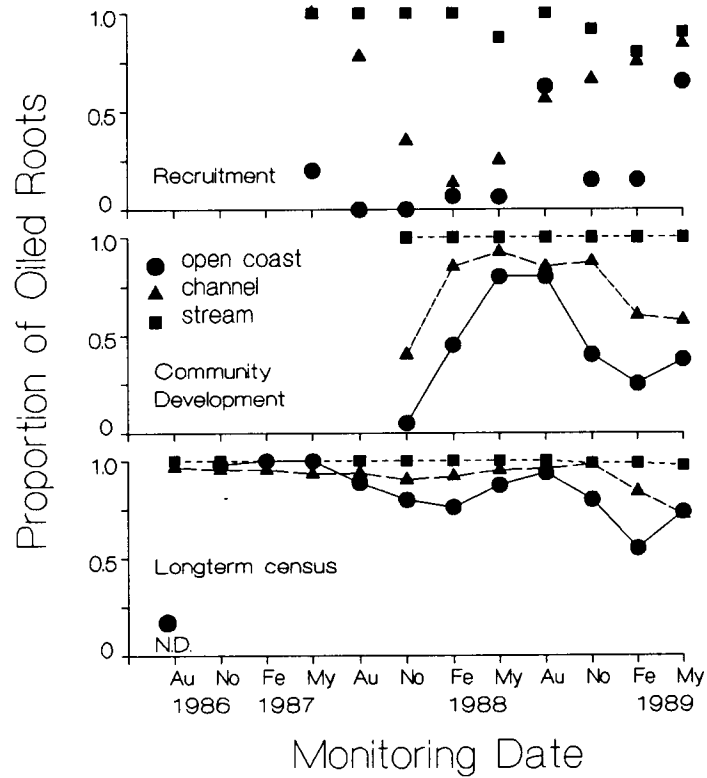


Figure 7.3 Proportion of sampled roots or dowels with oil visible. ND indicates "no data" for the open coast long-term census roots in August 1986, because oil was hidden by a heavy diatom cover. Numbers of sites and roots sampled within sites are variable.

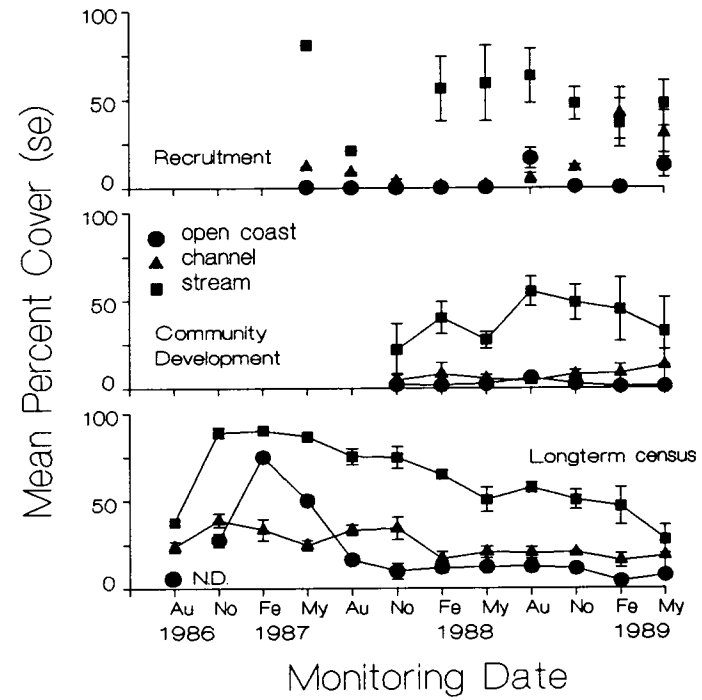


Figure 7.4 Percent cover of oil on sampled roots or dowels. Each symbol is an among site mean, with standard error bars. N=1-5 sites/date. See text and Figure 7.3 for details.

streams were oiled within three months and have remained oiled. This suggests highest levels of residual oiling in streams, followed by channels, then the open coast. Percent cover data for community development roots showed the same pattern (Figure 7.4); oil cover was low on the open coast (<10%) and in channels (<15%) relative to streams (~20-55%).

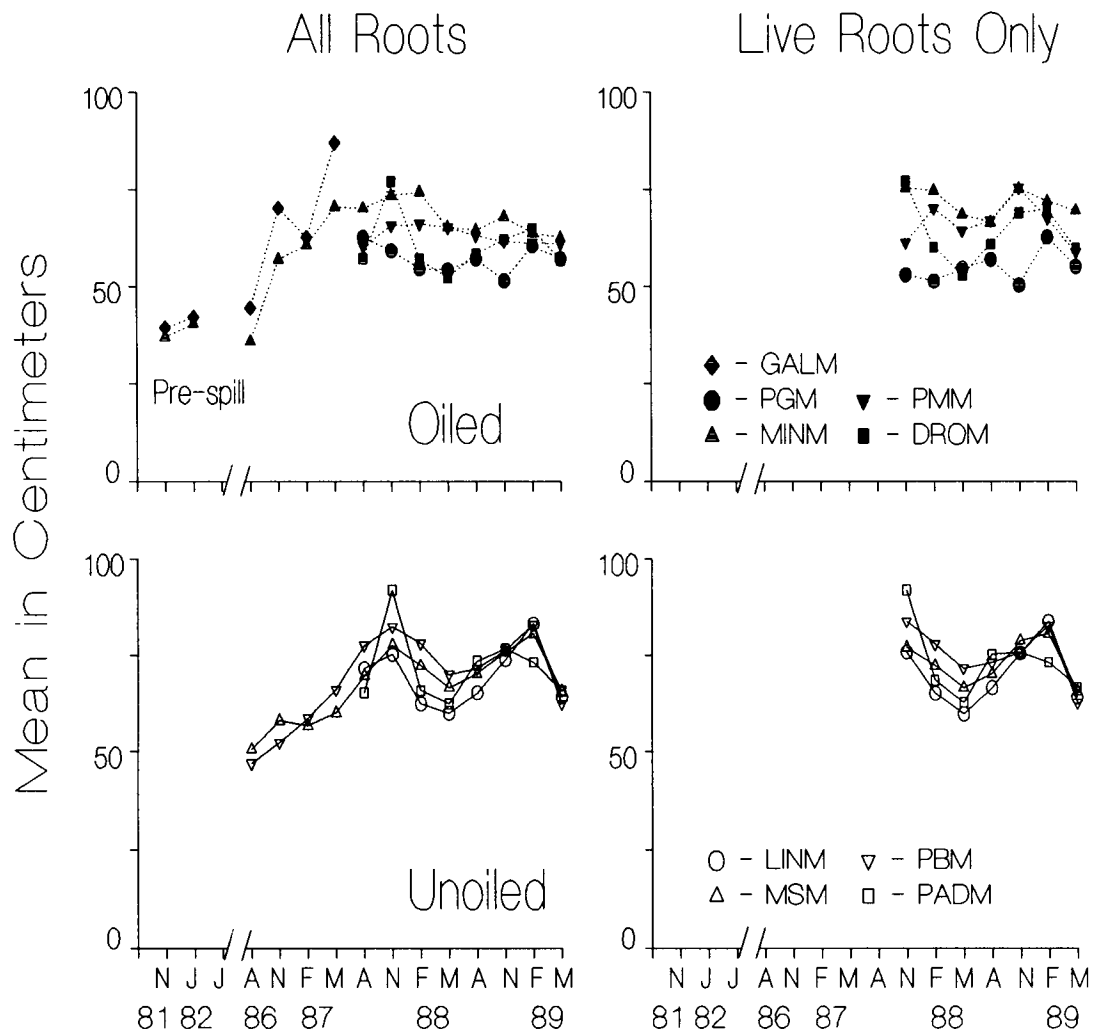
Recruitment data were taken from artificial roots (dowels) set out for three-month periods (beginning in August 1987). The occurrence and percent cover of oil on dowels give a relative measure of the amount of reoiling occurring over short, discrete periods (Figures 7.3, 7.4). At this scale, oiling on the open coast was highly episodic. Only in August 1988 and May 1989 were more than 20% of artificial roots oiled, and mean percent cover of oil was <20% both times. In channels, the fraction of roots oiled declined from 100% in August 1987 to 16% in February 1988, but increased steadily since that time. Percent cover of oil was also low, except in May 1989 when it reached 34%. In streams, 80-100% of the dowels were oiled to some degree in each monitoring period, and oil always averaged more than 40% cover.

Oil may flush from fill under the refinery when rains are heavy, thereby reoiling nearby areas; in addition, sediments in protected waters may be saturated with oil that leaks out when the surface is disturbed or in heavy rain. As a rough measure of the current oil burden in sediments, we tabulated the visual or olfactory evidence of oiling from sediment cores collected by J. MacPherson and S. Garrity for hydrocarbon analysis in May 1989 (Chapter 9, Table 9.13). No evidence of oil was found in cores from any unoiled site. At open, oiled sites, seven of 12 cores smelled of oil, and oil was visible in three of these. However, at one site (Punta Muerto), all three cores appeared clean. In oiled channels, all fifteen cores smelled oily and nine contained visible oil. In oiled streams, 10 of 12 cores had an oily smell and six of 12 also had visible oil. One of three cores at each of two sites appeared clean, but the two other cores from the same sites had visible oil. Thus, even at this crude level, oil was still present in sediments at all but one of thirteen oiled sites three years after the spill. Hydrocarbon results from these cores should verify these observations quantitatively.

In summary, (1) all three mangrove habitats were oiled, (2) few long-term census roots (i.e., randomly-sampled) at all sites had not been in contact with oil through 1989, and (3) oiling was heaviest initially in the open coast, but most repetitive and persistent in drainage streams, with channels and lagoons intermediate in amounts of oiling and reoiling. Finally, (4) even on the open coast, where percent cover of oil dropped to relatively low levels within a year after the spill, there has been some reoiling, with occurrence of oil on roots increasing in two rainy season monitorings (August 1988, May 1989).

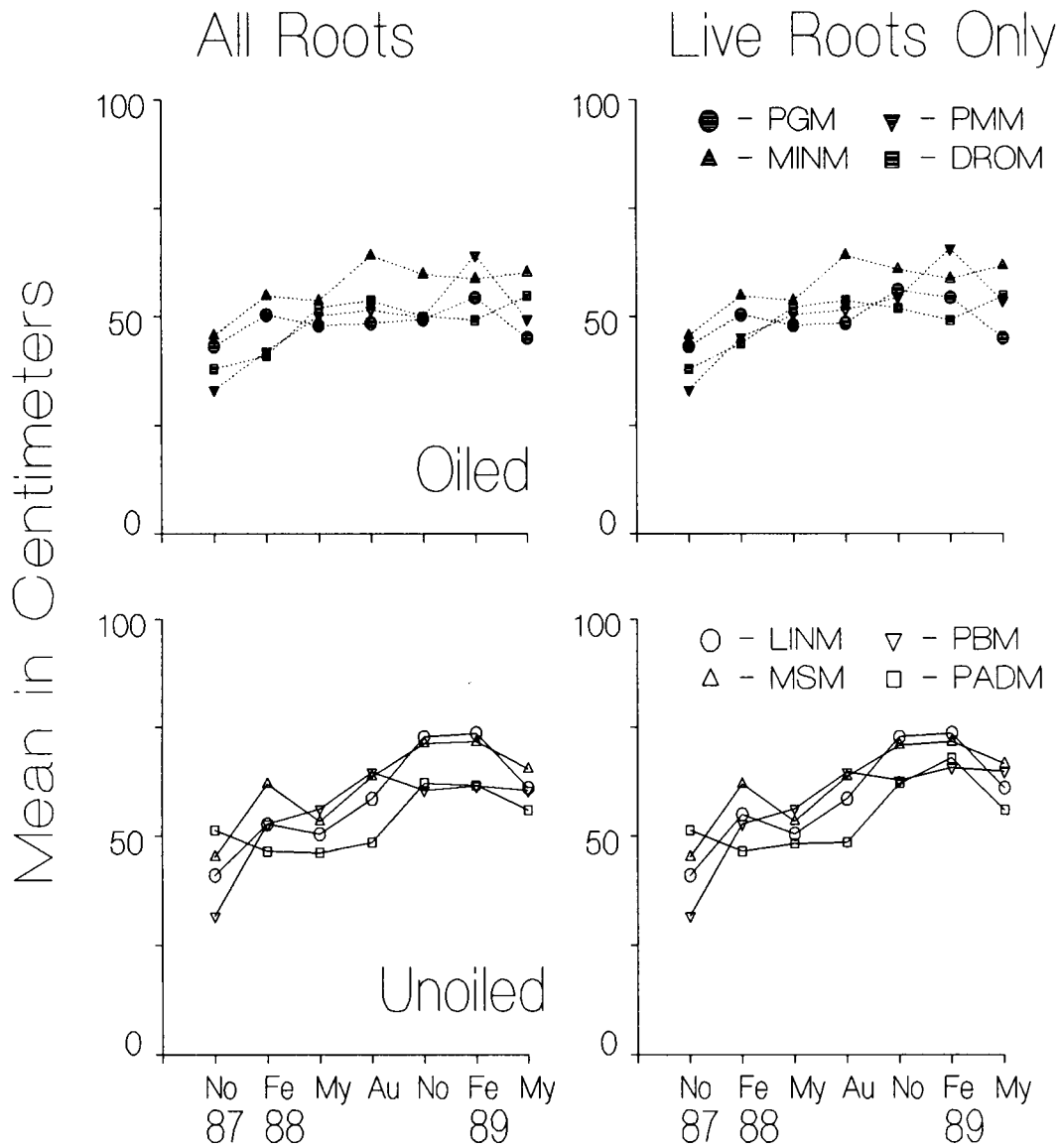
7.4.3 Root Size

Root length, measured in centimeters from the high water line (HWL) to tip, was taken for all roots (Figures 7.5-7.10). For long-term census roots, lengths increased over time (Figures 7.5, 7.7, 7.9). Mean lengths in all habitats and in oiled



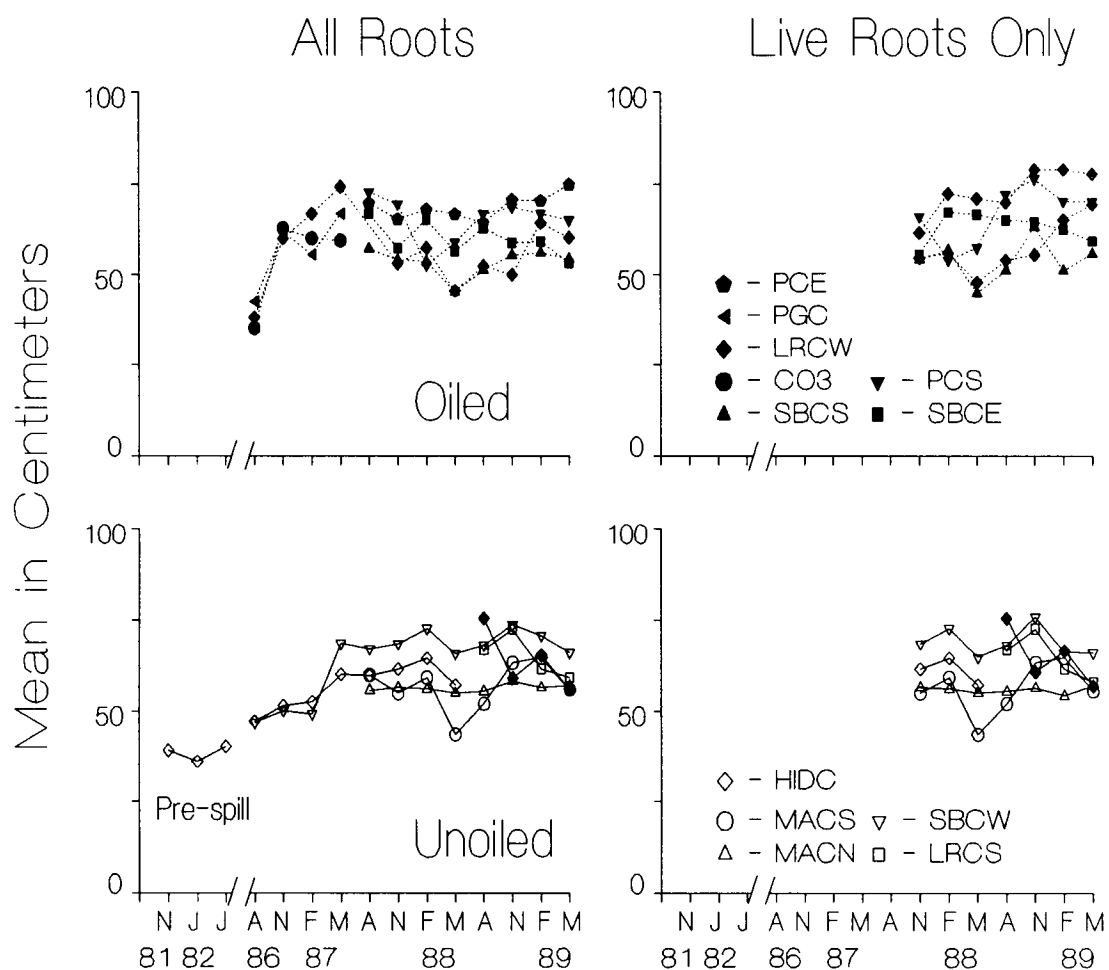
Root Length - Open Coast, Longterm census

Figure 7.5 Lengths of all sampled roots and of live roots only in long-term censuses, from the open coast. Measurements were made from HWL to end of longest tip. Data are means by site. For data from all roots, $n=15-75$ (before November 1987), then 20-25. For data from live roots, $n=9-25$. A=August, M=May, first J in 1982=January, second J in 1982=June.



Root Length - Open Coast, Comm. Dev.

Figure 7.6 Lengths of all sampled roots and of live roots only in community development substudy, from the open coast. Measurements were made from HWL to end of longest tip. Data are means by site. Sample size is 5 roots per site for November 1987 and February 1988, 10 roots per site from May 1988 on. For live roots, n=4-10. See text for details.



Root Length - Channels, Longterm census

Figure 7.7 Lengths of all sampled roots and of live roots only in long-term censuses, from channels and lagoons. Measurements were made from HWL to end of longest tip. Data are means by site. For data from all roots, $n=18-50$ before November 1987, then 20-25. For data from live roots, $n=9-25$. H1DC, which was secondarily oiled between May and August 1988, is indicated by filled symbols in the lower panel. See text for details and Figure 7.5 caption for month abbreviations.

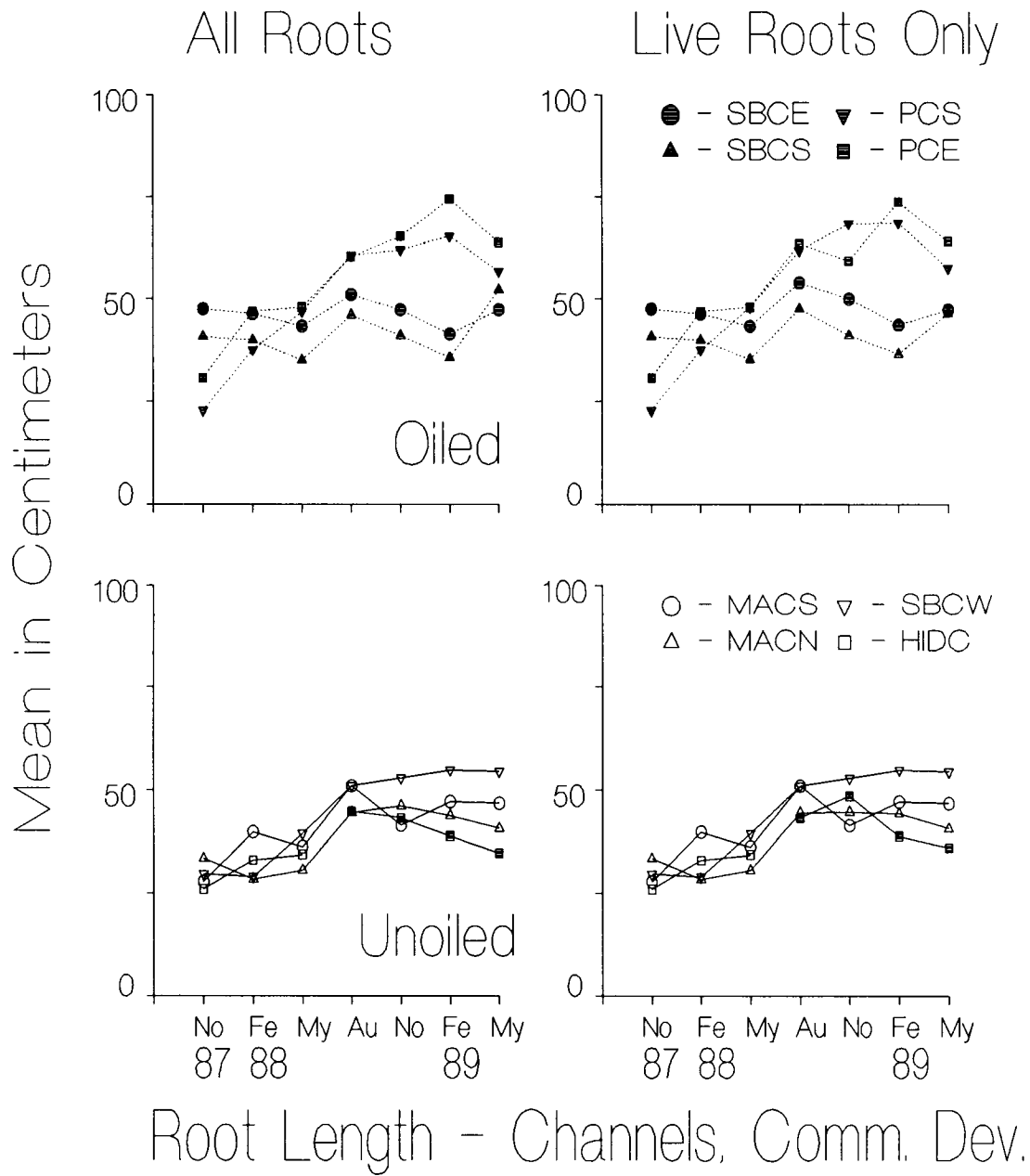
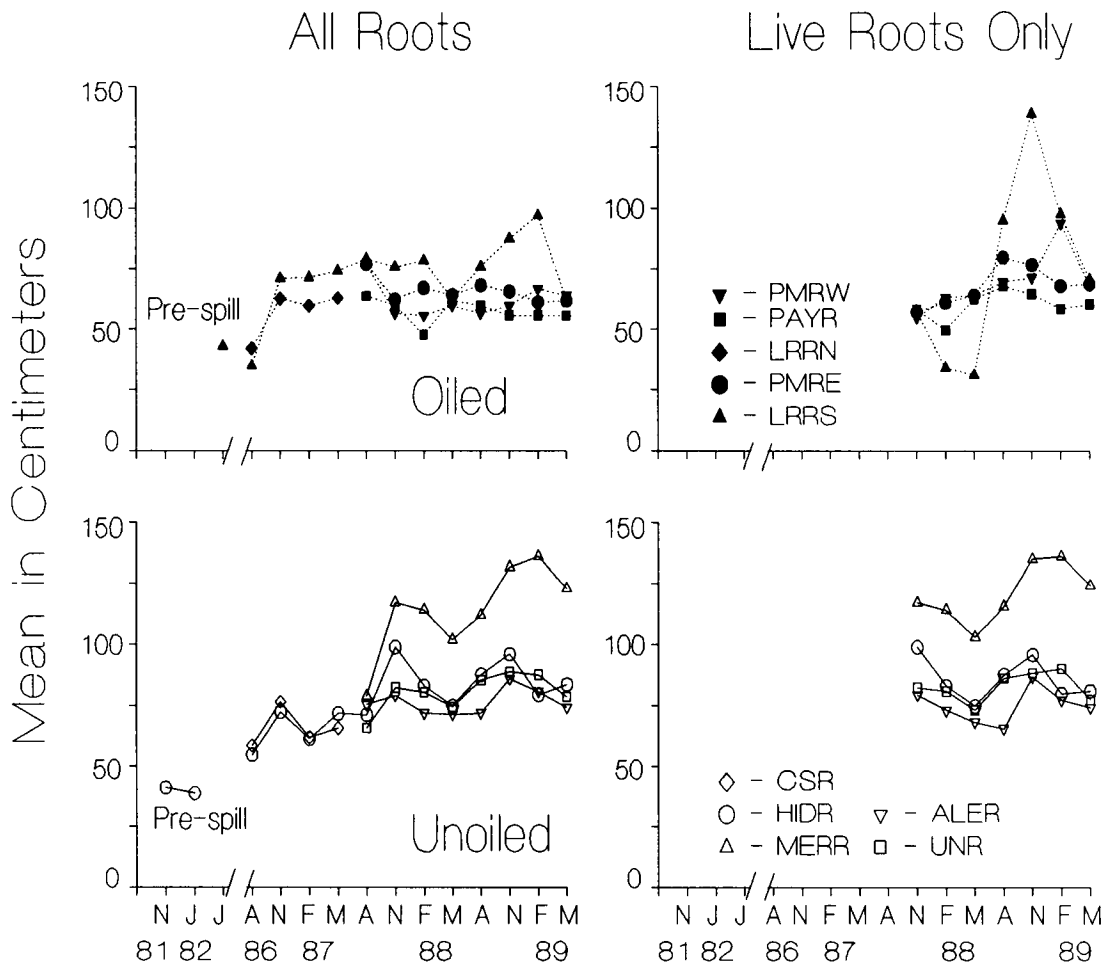
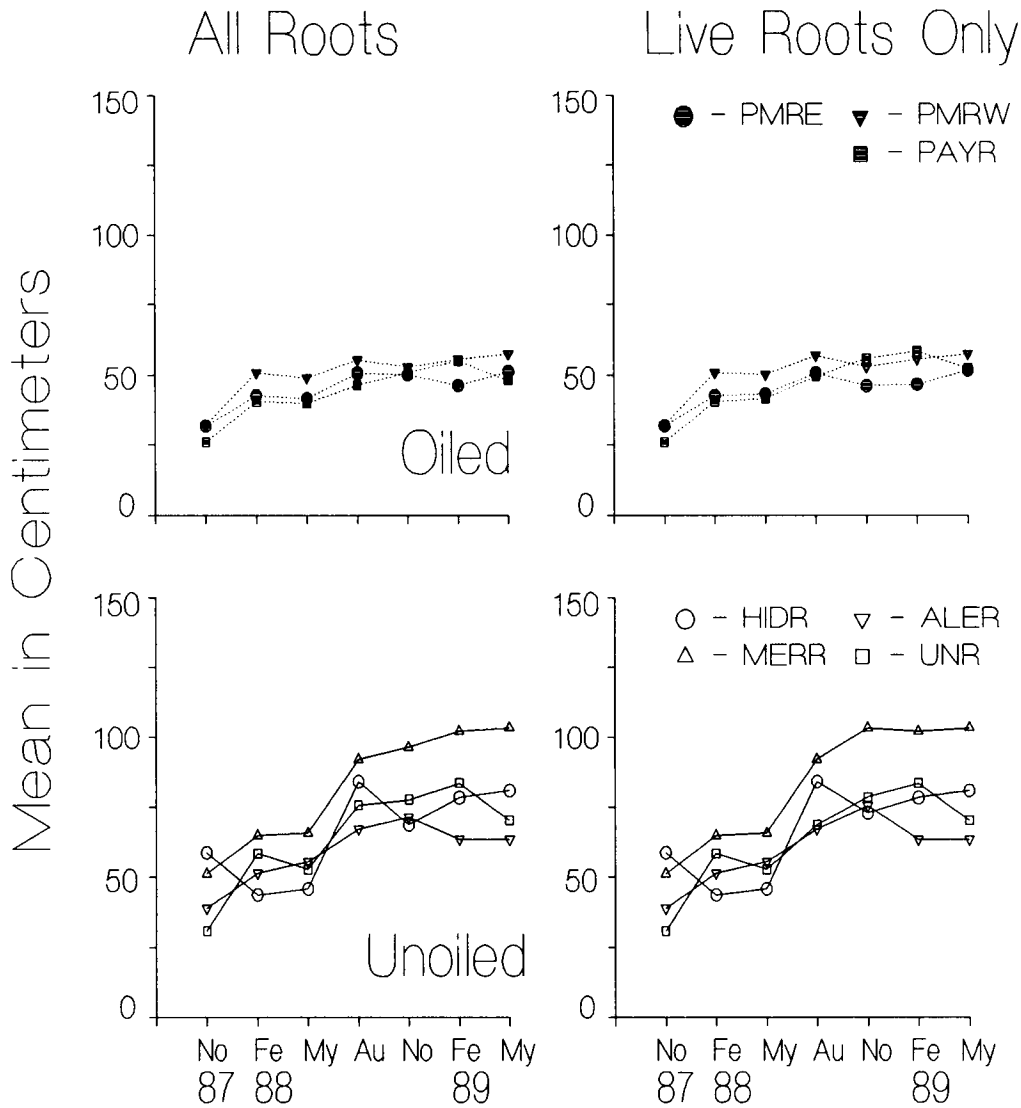


Figure 7.8 Lengths of all sampled roots and of live roots only in community development substudy, from channels and lagoons. Measurements were made from HWL to end of longest tip. Data are means by site. Sample size is 5 roots per site for November 1987 and February 1988, 10 roots per site for May 1988 on. For live roots, n=5-10. HIDC, which was secondarily oiled between May and August 1988, is indicated by filled symbols in the lower panel. See text for details.



Root Length - Streams, Longterm census

Figure 7.9 Lengths of all sampled roots and of live roots only in long-term censuses, from drainage streams and rivers. Measurements were made from HWL to end of longest tip. Data are means by site. Sample sizes for data from all roots is 16-50 before November 1987, then 20-25. Data from live roots only is 1-25. See text for details and Figure 7.5 caption for month abbreviations.



Root Length - Streams, Comm. Dev.

Figure 7.10 Lengths of all sampled roots and of live roots only in community development substudy, from drainage streams and rivers. No roots were marked at LRRS, due to the poor condition of trees and roots. Measurements were made from HWL to end of longest tip. Data are means by site. Sample size is 5 roots per site for November 1987 and February 1988, 10 roots per site for May 1988 on. For live roots, n=5-10. See text for details.

and unoiled treatments were greater in May 1989 than they were either pre-spill (1981-2) or just after the spill (August 1986). Root lengths in unoiled treatments may have a seasonal component: open coast roots, and to a lesser extent channel and stream roots, showed peaks in late wet or early dry seasons and decreases in average length in early rainy seasons, for both 1988 and 1989.

In May 1989, root lengths were longer at unoiled than at oiled open coast sites (mean \pm standard error, N=4 sites, 20 roots/site: unoiled, 64.5 ± 0.8 cm vs. oiled, 59.5 ± 1.5 cm) and in drainage streams (unoiled, 89.9 ± 11.2 cm vs. oiled, 60.9 ± 1.9 cm). Roots from unoiled and oiled channels and lagoons were very similar in length (unoiled, 4 sites: 58.9 ± 1.9 cm; oiled, 5 sites: 61.5 ± 4.0 cm).

To determine whether differences in root length resulted from greater root mortality at oiled sites (see below), we looked at root lengths of live long-term census roots only (Figures 7.5, 7.7, 7.9, beginning in November 1987, when sites and root condition codes were fully standardized). Roots that were spongy, with bark peeling off and no evidence of tip growth, were considered dead; we recalculated mean length without dead roots. Again, looking at overall means (sites combined), in November 1987 stream and open coast roots were longer at unoiled sites than at oiled sites (unoiled streams: 94.5 ± 8.7 cm, oiled streams: 56.8 ± 0.8 cm; unoiled open coast: 82.3 ± 3.7 cm, oiled open coast: 66.7 ± 5.8 cm); channels did not differ. Similarly, in May 1989 roots in unoiled streams and on the open coast were longer than those at oiled sites (unoiled streams: 89.2 ± 11.7 cm, oiled streams: 67.1 ± 2.3 cm; unoiled open coast: 64.8 ± 0.9 cm, oiled open coast: 60.1 ± 3.1 cm). Roots from oiled channels were longer than unoiled ones at this date (unoiled channels: 58.7 ± 1.9 cm, oiled channels: 66.4 ± 4.0 cm). These data suggest length differences were due to differences in growth of roots from oiled and unoiled areas, rather than an artifact resulting from differences in the numbers of dead (ungrowing) roots.

Figures 7.6, 7.8 and 7.10 show mean root lengths, by site, of the community development cohort, marked in August 1987. There was considerable variability among sites, but roots in all treatments and habitats increased in length over time, indicating root growth. Community development roots from unoiled streams and open shores had grown more by May 1989 than those in oiled sites (unoiled streams: 79.5 ± 8.8 cm, oiled streams: 52.2 ± 2.8 cm; unoiled open coast: 60.7 ± 2.0 cm, oiled open coast: 52.4 ± 3.3 cm). In channels and lagoons, oiled roots were longer than those at unoiled sites (unoiled: 44.1 ± 4.2 cm, oiled: 54.9 ± 3.5 cm).

Figures 7.6, 7.8, and 7.10 show community development root length data using live roots only. These did not differ from results using all roots, perhaps because relatively few roots in the community development cohort died, even at oiled sites, compared to those in the long-term censuses (see below, 7.4.4).

7.4.4 Changes in Root Condition

Root condition, sampled starting in March 1987, is shown in Table E.4 (Appendix) and Figures 7.11-7.12. Differences in sampling methods for root

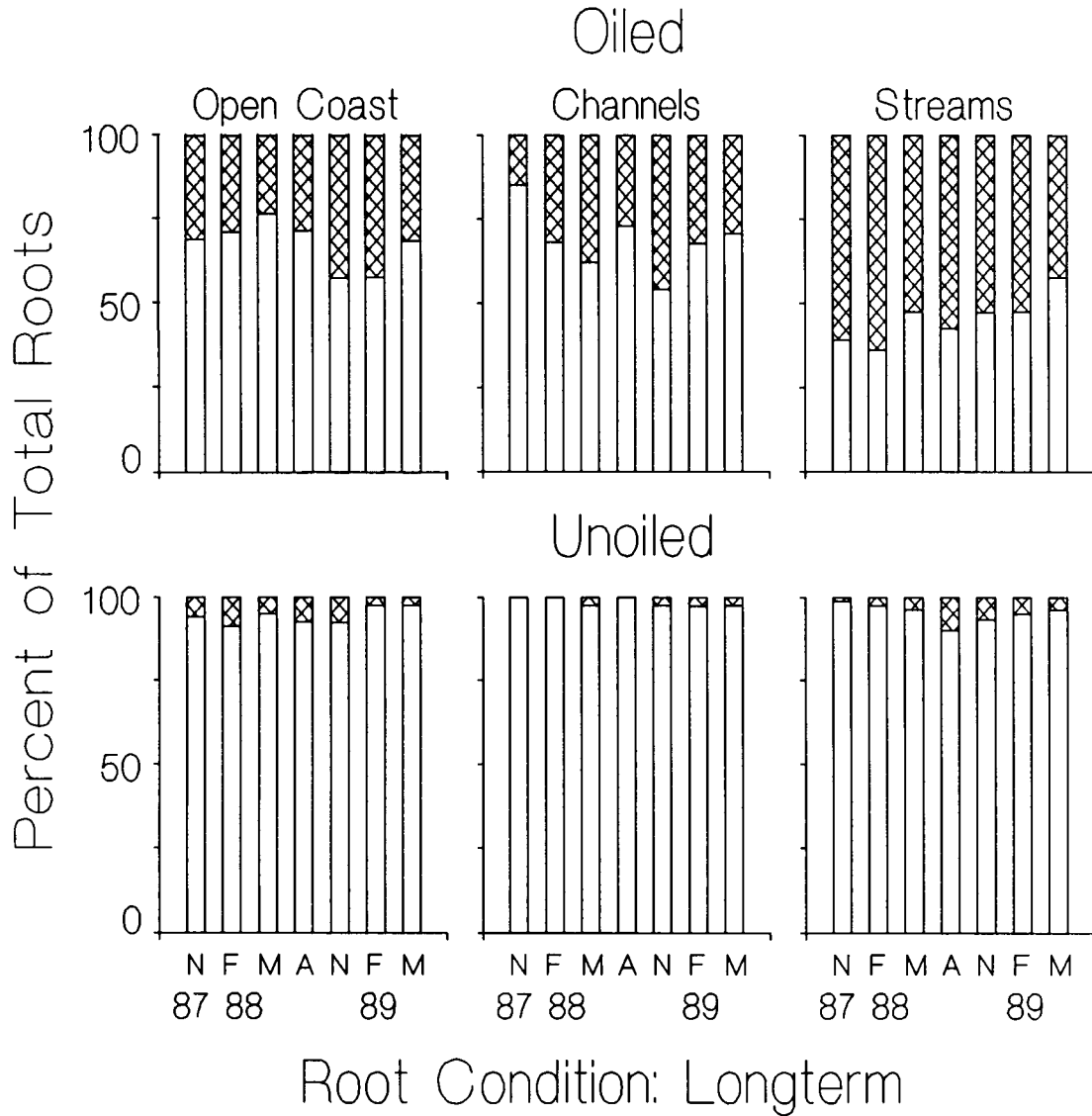


Figure 7.11 Percent of live and dead roots monitored for long-term censuses, in each habitat. Open part of each bar represents the proportion of live roots, hatched portion represents dead roots. The top graph shows oiled sites over time, bottom shows unoiled sites. Each bar is a sample of 80 roots from four sites, except for channels which had 80-100 roots from 4-5 sites. Monitoring dates are quarterly, beginning in November 1987. M=May, A=August.

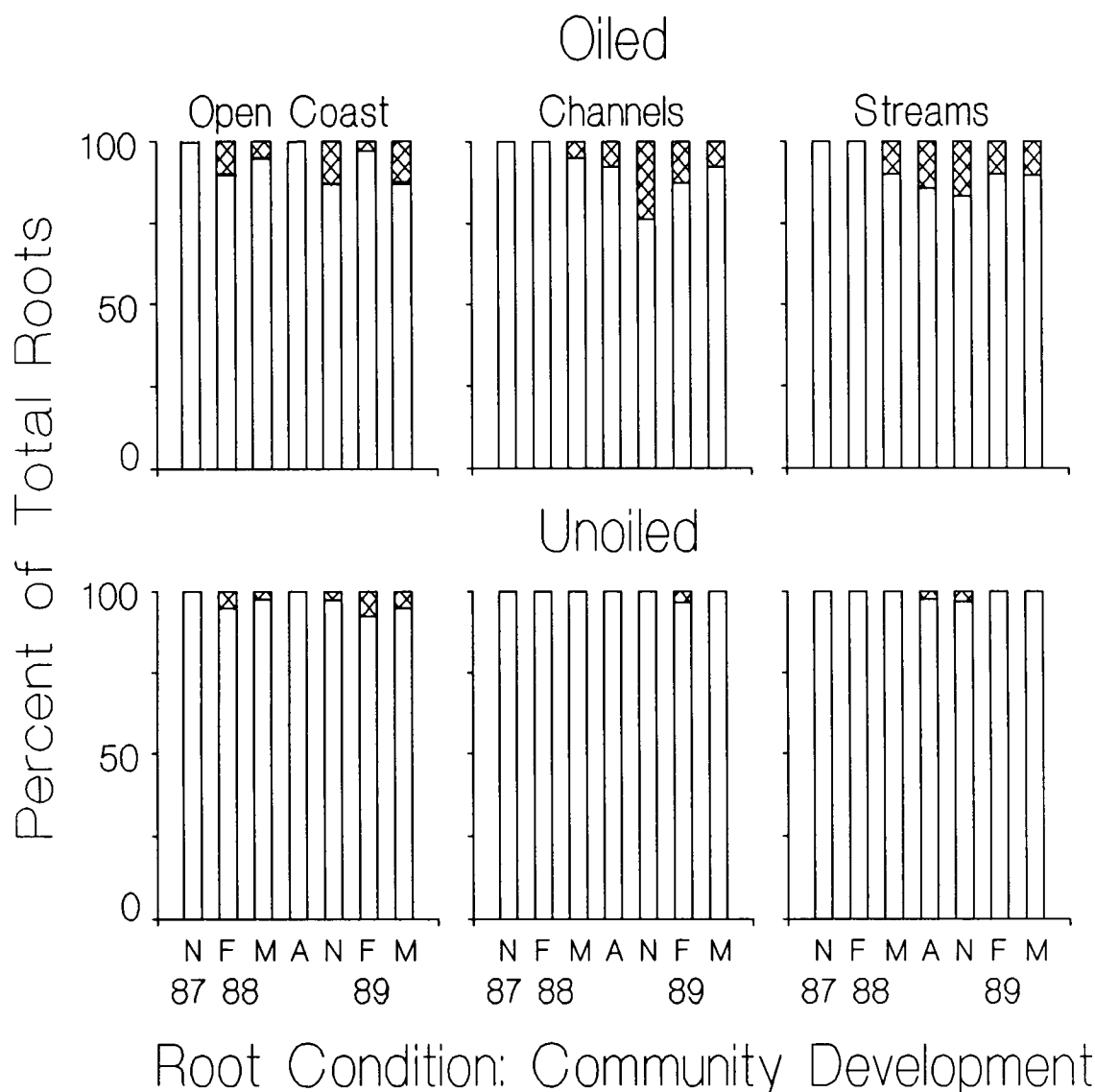


Figure 7.12 Proportion of live and dead roots monitored for community development substudy, in each habitat. Open part of each bar represents the proportion of live roots, hatched portion represents dead roots. The top graph shows oiled sites over time, bottom shows unoiled sites. Each bar is a sample of 20-40 roots from four sites. Monitoring dates are quarterly, beginning in November 1987.

conditions were not completely standardized until November 1987; these details are described in the methods section (see above, Chapter 7.3.1).

Root condition differed significantly between oiled and unoiled treatments in long-term census data (Figure 7.11). At unoiled sites, 10% or fewer of roots were dead and rotting in 82 of 84 monitorings from November 1987 to May 1989. The maximum fraction damaged was 20% at one site. In contrast, up to 92% of the roots at oiled sites were classified as dead and rotted. In May 1989, the percentage of dead and rotted roots was 31.3%, 29.3% and 42.5% of the total at oiled open, channel and stream sites, respectively. These values actually underestimated the number of decaying roots; those with any signs of regrowth (e.g., production of new tips) were counted as roots in good condition.

Root condition also varied among oiled habitats (Figure 7.11). The fraction of dead roots in the three habitat types was consistently highest in drainage streams and 10-20% lower on the open coast or in channels. This pattern may be related to heavy, sustained oiling in streams (Figures 7.3-7.4).

Roots sampled for community development were marked in August 1987 and subsequently entered the water. Survival of roots at unoiled sites has been high through May 1989, with at most one or two dead roots sampled at each sampling date (Figure 7.12). In contrast, roots from the same-aged cohort at oiled sites have since been more likely to die in all three habitat types.

The number of live and dead roots visible on the vertical surface of cores collected in May 1989 for hydrocarbon analysis was tallied (see Chapter 9, Table 9.12 for details). There were slightly more live than dead roots in cores from unoiled open coast and channels, while the opposite occurred in streams. However, there were large differences in the number of dead roots between treatments in all three habitats: there were at least one-and-a-half times more dead roots at oiled than unoiled channel and stream sites and more than six times as many dead roots at oiled, open sites.

Summaries of root condition point out some of the long-term damage that oiled trees have received. In May 1989, three years after the spill, live roots represented only 68.8%, 70.7% and 57.5% of roots sampled on the oiled open coast, in channels and in streams, respectively. The comparable percentages from unoiled sites were 97.5%, 97.5% and 96.3%. Clearly, the mangrove root habitat itself has been damaged by oiling and has not recovered.

7.4.5 Secondary Effects of Oil Damage

Damage to mangrove trees from oiling changed the physical characteristics of the habitat. At least two types of damage may have had secondary effects on the epibiota of mangrove roots. First, defoliation removed the weight of leaves from branches and, in some cases, the branches flexed upwards and lifted roots out of the water. Many organisms that had survived oiling were then killed by desiccation or heat stress or both. This process was especially common in oiled channel sites (see Figure 2 in Jackson *et al.* 1989).

Defoliation should decrease shading and increase light available for algal growth. Such changes in light intensity may contribute to later differences in algal cover. Data are shown in Table 7.2 as means and ranges (in lux) for readings taken outside the canopy and inside, where roots are monitored (n=20 paired readings for each site). "Percent of outside" is the ratio of the mean of illumination inside the canopy and the mean of illumination outside the canopy multiplied by 100.

Shading increased from open coast to channels to drainage streams (Table 7.2). At unoiled, open coast sites, 30-37% of light recorded outside the canopy was available within it. This was reduced to 12-24% in channels and lagoons and to less than 21% in drainage streams. Illumination levels were always patchy, indicating irregular breaks in the canopy.

More light was on average available at oiled than unoiled sites in all three habitat types. The difference was small on open coasts: percent of outside light penetration through the canopy was 34-64% on oiled open coasts and 30-37% at unoiled sites. In oiled channels and lagoons, 34-62% percent of outside light was recorded, 3-4 times as much as at unoiled sites. Light levels in oiled channels were thus as high as those on oiled and unoiled open coast. Oiled drainage streams showed even greater differences: 6-8 times as much light was found in oiled as compared to unoiled streams, and illumination was greater than either on the open coast or in channels.

Increases in light levels occurred in oiled sites in all three habitats. Differences were largest in channels and drainage streams.

7.4.6 Pre-spill Epibiotic Assemblages on *Rhizophora* Roots

Table 7.3 shows the rank abundance of various taxa on the open coast, in channels, and in streams based on roots sampled in October 1981 and January 1982. These pre-oiling samples were chosen because all habitats were sampled with approximately the same effort at the same time. Data are the percent of roots on which a given category was found at least once.

Overall, there was a decrease in the number of categories of percent cover (including bare space and mixed species groups) from open coast (36) to channels (10) and streams (11). This difference appeared although slightly fewer roots (n=70) were sampled on the open coast than in channels or streams (n=75 each habitat). Individual taxa tended to be rarer in occurrence on the open coast than in channels or streams. The rarest species in channels and streams occurred on almost a third of all roots, while only 25% of the species found on the open coast were that common.

Table 7.2 Illumination (lux) measured next to mangrove canopies and just above the water where prop roots are measured. Paired measurements, n=20 per site; 5-12 December 1988; 1000-1300; clear or partly cloudy conditions.

Site	Outside of Canopy			Inside of Canopy			Percent of Outside
	Mean	Minimum	Maximum	Mean	Minimum	Maximum	
A. Oiled, Open Coast							
Isla Droque Mangrove	2458	2200	2750	1048	250	2475	43
Isla Mina Mangrove	2935	1800	3600	996	175	3200	34
Peña Guapa Mangrove	2424	1900	3100	1550	325	3000	64
B. Unoiled, Open Coast							
Isla Lintón Mangrove	3619	3400	3825	1091	80	3600	30
María Soto Mangrove	3327	3200	3425	1226	90	3300	37
Isla del Padre Mangrove	3058	2875	3200	1119	45	3100	37
Portobelo Mangrove	3440	3000	3700	1051	95	3400	31
C. Newly Oiled Channels and Lagoons							
Hidden Channel	2074	1900	2200	629	100	1625	30
D. Oiled Channels and Lagoons							
Largo Remo Channel West	2145	1875	2400	1321	460	2200	62
Payardi Channel East	2801	2000	3150	938	210	2500	35
Payardi Channel South	2764	2100	3200	946	140	2800	34
Samba Bonita Channel East	2748	2200	3175	1316	500	2800	48
Samba Bonita Channel South	3164	2750	3375	1393	450	2900	44
E. Unoiled Channels and Lagoons							
Largo Remo Channel South	2014	1675	2425	480	60	1900	24
Margarita Channel North	1598	1300	2050	212	70	550	13
Margarita Channel South	2894	2500	3100	346	80	2250	12
Samba Bonita Channel West	3179	2850	3500	465	150	1200	15
F. Oiled Drainage Streams							
Largo Remo River South	2630	2450	2800	1860	500	2675	71
Punta Muerto River East	3203	3150	3250	2210	450	3225	69
Punta Muerto River West	2988	2850	3050	1830	90	3050	61
Payardi River	2505	2200	2900	777	55	2500	31
G. Unoiled Drainage Streams							
Hidden River	3121	2900	3275	294	50	1925	9
Quebrada Las Mercedes	2273	2175	2350	121	45	790	5
Río Alejandro	2238	1900	2525	449	70	2100	20
Unnamed River	3220	3175	3275	670	45	2550	21

There was limited overlap in species occurrence among habitats in these samples taken before the oil spill. The exceptions were mixed species groups (e.g., diatoms, mixed algal turfs) and bare space. Where taxa did occur in more than one habitat, their abundance on roots tended to be quite different (e.g., *Balanus improvisus* at all sites, *Crassostrea rhizophorae* in streams and channels). Some entire groups were absent from a given habitat (e.g., sponges, hydroids and blue-green algae from streams, bivalve molluscs from the open coast). The open coast was dominated by foliose algae (42-62% cover, mean of mean cover for each sample date and site) and sessile filter-feeding invertebrates (13-22%). The edible oyster *Crassostrea rhizophorae* was the most common species in channels (50-54%) and the false mussel *Mytilopsis sallei* covered the most space in streams (59-64%). Thus, each habitat supported a distinct epibiotic community.

7.4.7 Post-spill Epibiota on Mangrove Roots

Figures 7.13-7.39 summarize percent cover data for major species, groups of species or other categories occupying space on open, channel and stream roots, for each quarterly monitoring through May 1989. Data from all sites within a given habitat (open, channel and stream) and treatment (oiled, unoiled) are combined in the figures; the means are calculated as the mean of all site means. Beginning in August 1987, each symbol thus represents a mean value (its associated standard error) from quarterly samples of 4-5 sites (80-100 roots) in the **long-term census** study, from 3-4 sites (40-50 roots) in the **community development** study, and 4-5 sites (from ~20 dowels, roughly, due to variable losses, set in groups of 5 at each site) in the **recruitment** study. Prior to this, 1-4 sites per habitat type were sampled (see Table 7.1, Methods section above, Chapter 7.3.1).

Open Coast Habitat -- Unoiled Sites

Long-term Census

The mangrove roots of unoiled, open coasts were best characterized by a diverse group of foliose algae (see also Batista 1980). Overall mean algal abundance ranged from 17 to 33% from August 1986 through May 1989 (Figure 7.13). Diatoms were similar to foliose algae in their pattern of abundance and ranged from 12 to 34% cover (Figure 7.14). Crustose algae were less abundant than foliose species, but increased by nearly a factor of ten since August 1986 (Figure 7.15). Barnacles, sponges, arborescent hydroids, bryozoans and other sessile invertebrates regularly occurred, but no one group occupied more than 5% cover at any monitoring (Figures 7.16-7.19). Bare space on roots varied from 16 to 34% cover (Figure 7.20). The remaining space was occupied by rare and intermittently present organisms such as anemones, amphipod tubes, tunicates, corals, vermetids and blue-green algae.

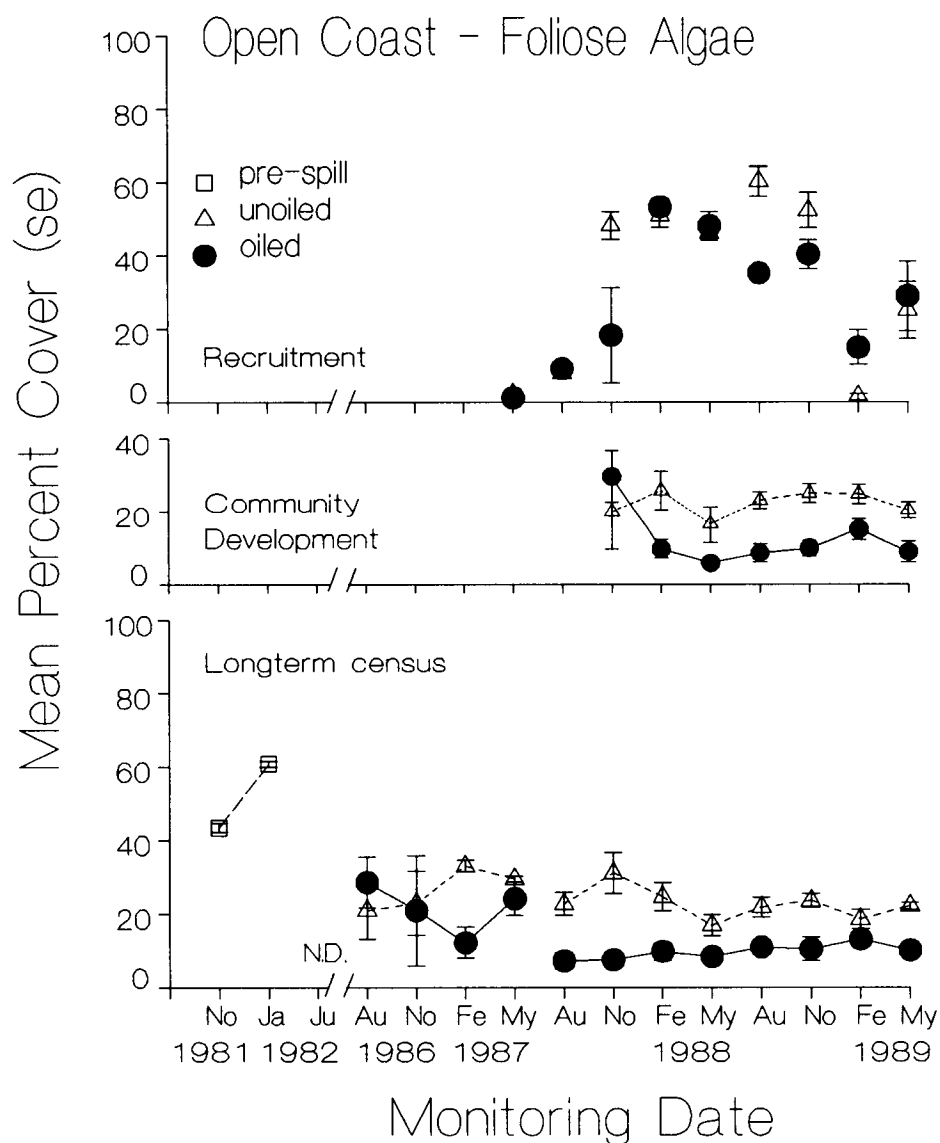


Figure 7.13 Abundance of foliose algae on roots or dowels sampled from the open coast. Data are stacked graphs of percent cover for recruitment, community development and long-term census substudies. Symbols represent among site means with standard error bars. N=1-4 sites/date. See text for details.

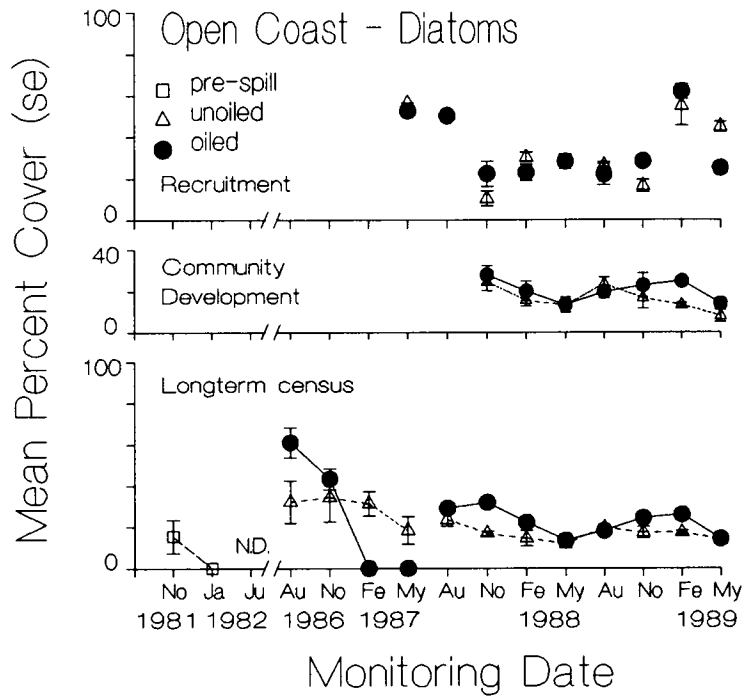


Figure 7.14 Abundance of diatoms on roots or dowels sampled from the open coast. Symbols represent among site means with standard error bars. N=1-4 sites/date. See text for details.

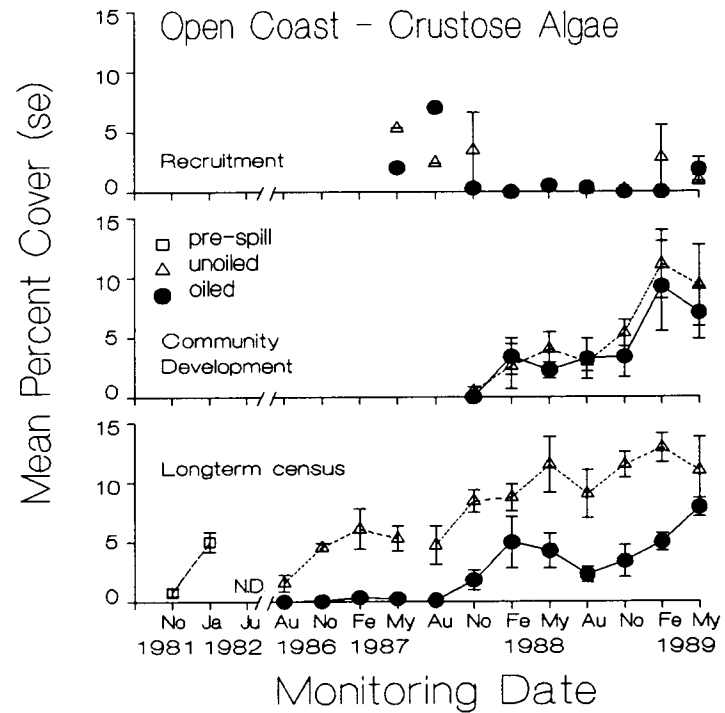


Figure 7.15 Abundance of crustose algae on roots or dowels sampled from the open coast. Symbols represent among site means with standard error bars. N=1-4 sites/date. See text for details.

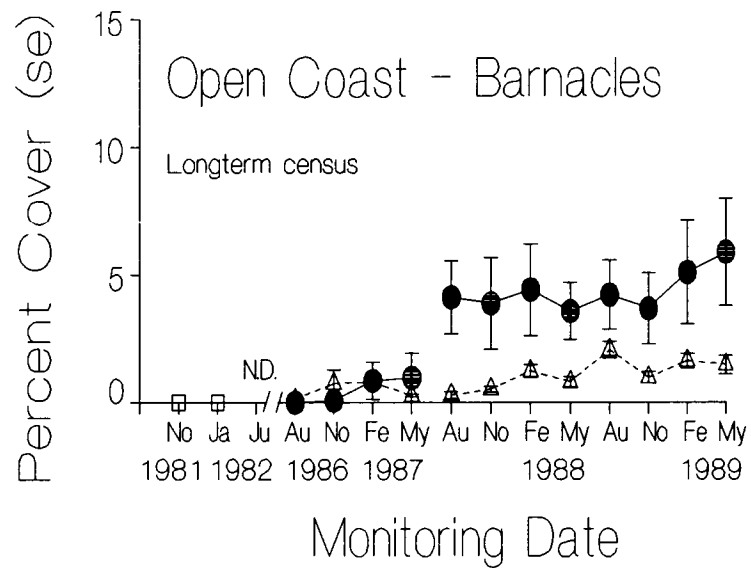


Figure 7.16 Abundance of barnacles on roots in long-term censuses from the open coast. Symbols represent among site means with standard error bars. N=2-4 sites/date. Closed circles=oiled sites, open triangles=unoiled sites, open squares=pre-spill data. See text for details.

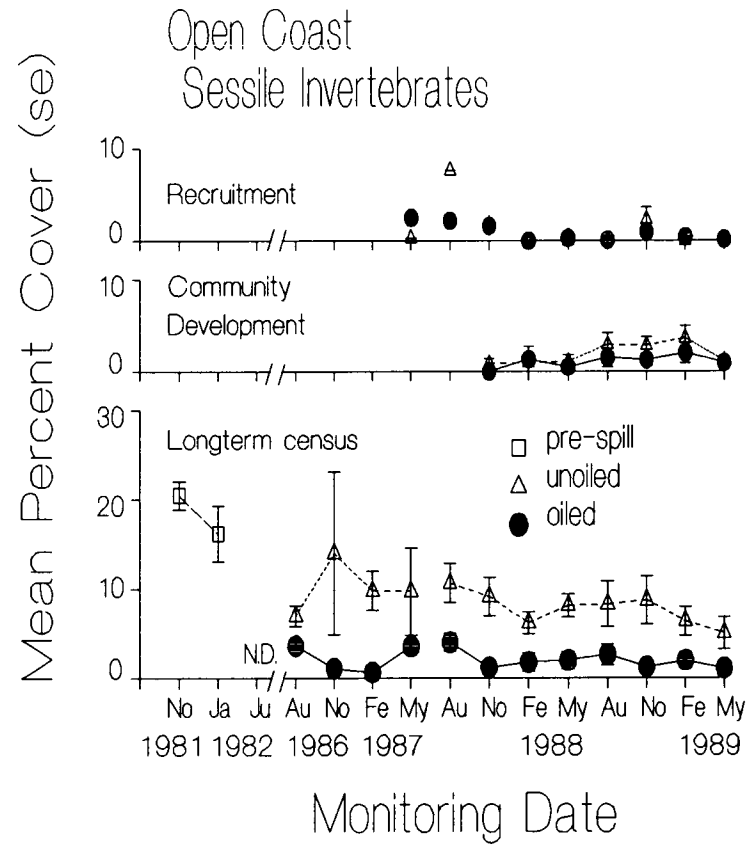


Figure 7.17 Abundance of sessile invertebrates excluding barnacles and bivalves on roots or dowels from the open coast. Symbols represent among site means with standard error bars. N=1-4 sites/date. See text for details.

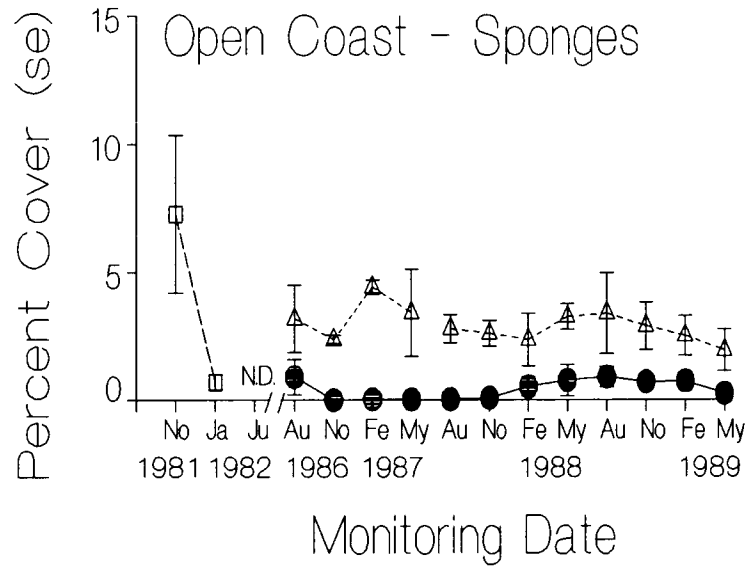


Figure 7.18 Abundance of sponges on roots in long-term censuses from the open coast. Symbols represent among site means with standard error bars. N=2-4 sites/date. Closed circles=oiled sites, open triangles=unoiled sites, open squares=pre-spill data. See text for details.

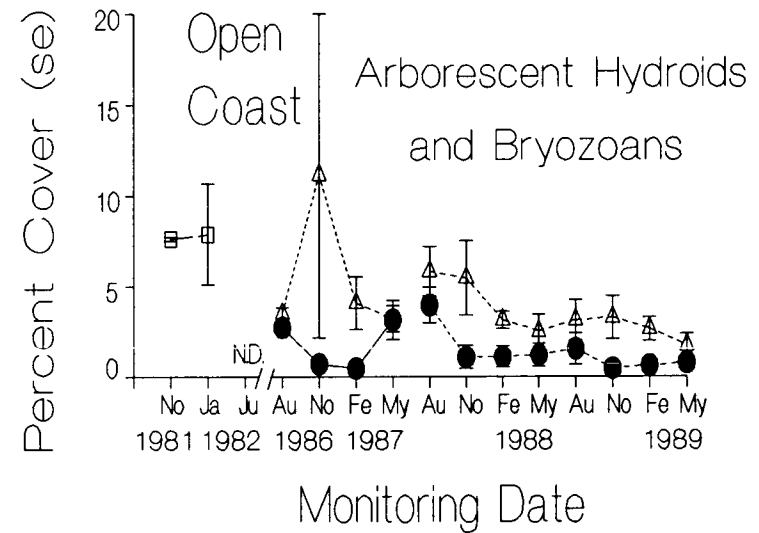


Figure 7.19 Abundance of arborescent hydroids and bryozoans on roots in long-term censuses from the open coast. Symbols represent among site means with standard error bars. N=2-4 sites/date. Closed circles=oiled sites, open triangles=unoiled sites, open squares=pre-spill data. See text for details.

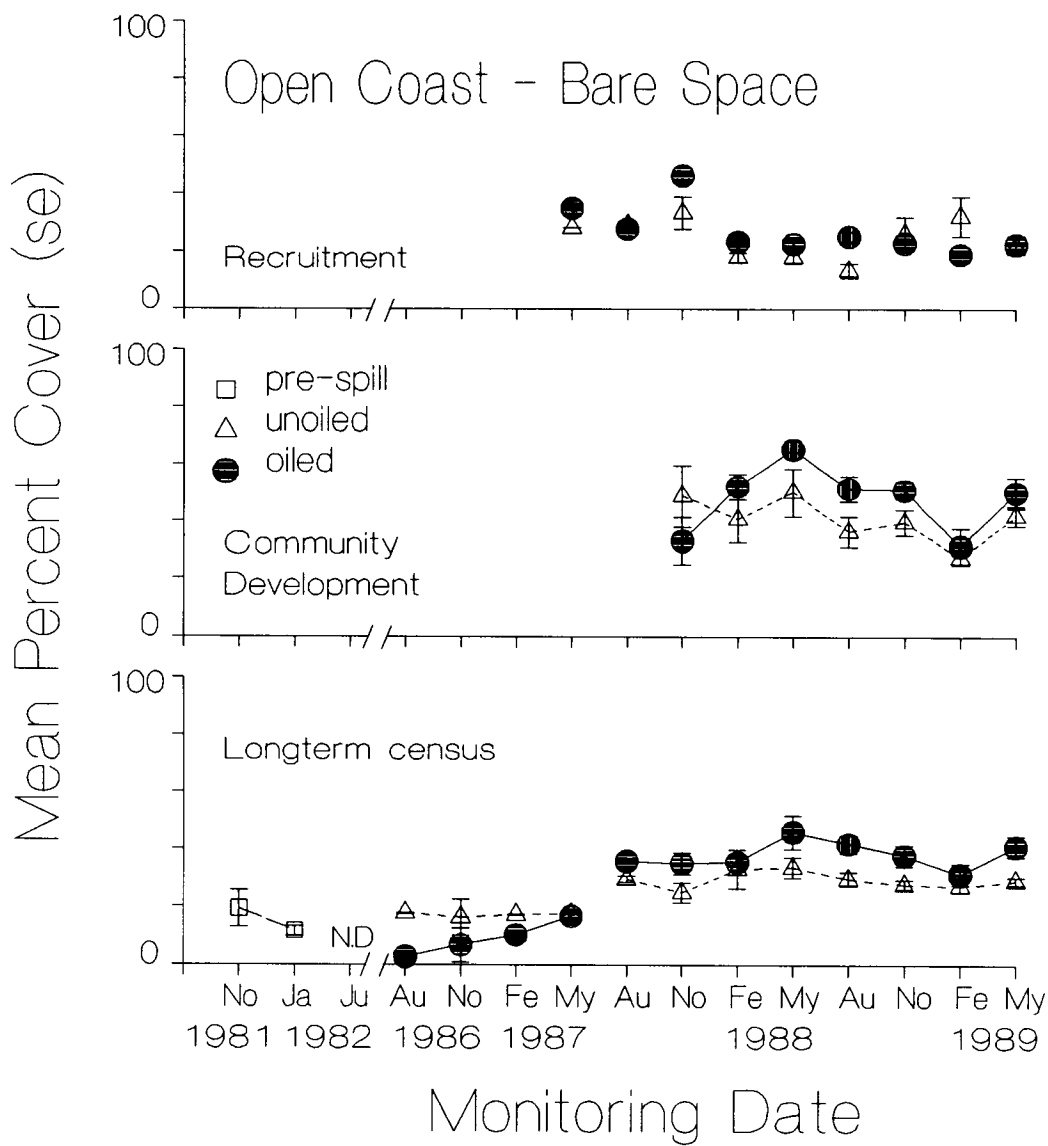


Figure 7.20 Abundance of bare space on roots or dowels sampled from the open coast. Symbols represent among site means with standard error bars. N=2-4 sites/date. See text for details.

Differences in species composition among sites are best discussed with reference to foliose algae (Tables 7.4-7.6). Species composition among all unoiled sites (1986-1989) was broadly similar. When rarefaction was used to estimate the expected number of species found in a sample of 20 roots, 8.2-14.6 species of foliose algae per site were expected (Figure 7.21) during this three-year sampling period.

Mixed algal turfs were most abundant, present on 875 of 1060 roots sampled in unoiled open sites. This category consisted of a <3 mm high turf composed of fine filaments of red, brown and green algal species (J. Connor, personal communication). Aside from mixed algal turfs, genera of algae that were regularly found included *Dictyota*, *Galaxaura*, *Acanthophora*, *Ceramium*, *Derbesia*, *Jania* and *Polysiphonia*.

Direct comparisons between 1986-1987 and later samples were difficult because of changes in site locations (Table 7.1). However, about the same number of species were collected at unoiled sites in the first and second years after the oil spill (Tables 7.4-7.5). Only three species collected in 1986-1987 were not found in 1987-1988 (4/665 occurrences in 1986-1987; each root an alga appeared on was counted as an occurrence, so occurrences could outnumber species) and six found in 1987-1988 were not found earlier (9/634 records). In 1987-1988, on 85 mangrove roots sampled at each of four distinct sites, each location had 17-27 species and 0-6 species not found at any other open unoiled site. In 1988-1989, on 80 roots sampled at the same four sites, each site had a total of 16-21 species and 2-5 species not found at any other open unoiled site in 1988-1989 (Table 7.6).

Community Development

Rates of development in the assemblage of species found on the open coast differed, depending upon the group. Sessile invertebrates were slow to appear (Figure 7.17), with almost no sponges recorded a year-and-a-half after monitoring began. Crustose algae were initially rare, but now equal abundances on long-term census roots (Figure 7.15). Diatoms were steadily abundant, covering approximately 20% of the root surface, as on long-term census roots (Figure 7.14). Bare space was slightly more abundant on community development roots than long-term census roots (Figure 7.20).

Overall algal abundance was similar to that on long-term census roots from the first monitoring (when the assemblage was only a maximum of three months old) through May 1989 (Figure 7.13). We examined algal species composition for 1988-1989, after the assemblage had had a year to develop (Table 7.7). The species recorded were a subset of those found on long-term census roots, with only one occurrence of a species not also found on randomly sampled roots (one root, unidentified brown alga). The expected number of species in a sample of 20 roots ranged from 3.0-13.0 (Table 7.8). For three of four sites, the expected number of species was close to that found on randomly-sampled long-term census roots (Table 7.8, Figure 7.21). At Isla del Padre, where far fewer species were recorded, mixed algal turfs were present in high percent cover and few other species were found.

Table 7.4 Number of roots on which an algal species or category occurred during the first year after the spill. Data are for August 1986-May 1987, summed over all sites, for oiled and unoled habitats.

A. Open Coasts			B. Channels and Lagoons		
Category	Oiled	Unoled	Category	Oiled	Unoled
<i>Acetabularia crenulata</i>	7		<i>Acanthophora spicifera</i>	7	
<i>Acanthophora spicifera</i>	73	48	<i>Bostrychia tenella</i>	6	22
<i>Bostrychia tenella</i>		11	<i>Bostrychia</i> sp.2	11	2
<i>Caloglossa letrieurii</i>		1	<i>Caloglossa letrieurii</i>		17
<i>Caulerpa fastigata</i>	13	14	<i>Caulerpa fastigata</i>	1	21
<i>Caulerpa mexicana</i>	2		<i>Caulerpa sertularioides</i>	5	
<i>Caulerpa racemosa</i>	1		<i>Caulerpa verticillata</i>	44	21
<i>Caulerpa sertularioides</i>	11	4	<i>Ceramium</i> sp.	17	5
<i>Caulerpa verticillata</i>	87	8	<i>Chaetomorpha</i> sp.	1	
<i>Ceramium</i> sp.	14	29	? <i>Gelidium pusillum</i>	3	18
<i>Chaetomorpha</i> sp.	1	12	<i>Enteromorpha</i> sp.	3	
<i>Cladophora</i> sp.	2	19	<i>Gelidiella acerosa</i>	1	
<i>Codium repens</i>	2	2	<i>Herposiphonia secunda</i>	1	
? <i>Gelidium pusillum</i>		4	<i>Jania ?adherens</i>	1	3
<i>Derbesia</i> sp.	3	15	Mixed algal turf	11	21
<i>Dictyota divaricata</i>	18	41	<i>Polysiphonia</i> sp. 1	68	41
<i>Ectocarpus breviararticulatus</i>	5	3	<i>Polysiphonia</i> sp. 2	110	84
<i>Enteromorpha</i> sp.	3	11	<i>Polysiphonia</i> sp. 3	47	34
<i>Galaxaura comans</i>	2	25	<i>Rhizoclonium</i> sp.		1
<i>Gelidiella acerosa</i>	1	2	Total taxa:	17	13
<i>Gracilaria mammillaris</i>	1		Total occurrences:	337	290
<i>Halimeda opuntia</i>		1	Number of roots:	372	360
<i>Heterosiphonia</i> sp.	8		C: Drainage streams		
<i>Hypnea spinella</i>	4		Category	Oiled	Unoled
<i>Jania ?adherens</i>	4	30	<i>Bostrychia tenella</i>		33
<i>Laurencia obtusa</i>		2	<i>Caloglossa letrieurii</i>		64
<i>Laurencia papillosa</i>	2	3	<i>Caulerpa fastigata</i>		1
Mixed algal turf	86	332	<i>Caulerpa verticillata</i>	18	
<i>Padina gymnospora</i>	6		<i>Chaetomorpha</i> sp.		13
<i>Penicillus capitatus</i>	1	1	<i>Cladophora</i> sp.	1	
<i>Polysiphonia</i> sp. 1	2		Mixed algal turf		20
<i>Polysiphonia</i> sp. 2	1	5	<i>Polysiphonia</i> sp. 1		108
<i>Polysiphonia</i> sp. 3	37	48	<i>Polysiphonia</i> sp. 2		32
<i>Rhizoclonium</i> sp.	10	3	<i>Polysiphonia</i> sp. 3		33
<i>Spyridia filamentosa</i>	1		Total taxa:	2	8
<i>Sinuvea anastomosans</i>		1	Total occurrences:	19	304
Total taxa:	30	27	Number of roots:	371	391
Total occurrences:	408	665			
Number of roots:	378	398			

Table 7.5 Distribution of algal species or categories on open coasts, long-term census, second year after the spill. Data are for August 1987 - May 1988. N=85 roots/site, 340/habitat type. Site codes as in Table 7.1. Sum = total occurrences/category at oiled or unoiled open sites.

Category	Oiled					Unoiled				
	DROM	MINM	PGM	PMM	Sum	LINM	MSM	PADM	PBM	Sum
<i>Amphiroa ?fragilissima</i>	2	1		3	6		2		1	3
<i>Acanthophora spicifera</i>	7	2	2	1	12	7	16	1	5	29
<i>Bostrychia tenella</i>	1		3	1	5		5	1	2	8
<i>Bostrychia</i> sp.2							1			1
<i>Caulerpa fastigata</i>	1		2	2	5	3	7	7	5	22
<i>Caulerpa mexicana</i>				1	1	1	3			4
<i>Caulerpa racemosa</i>		1			1		1			1
<i>Caulerpa sertularioides</i>				1	1					
<i>Caulerpa verticillata</i>	3	5	2		10		1	6	4	11
<i>Ceramium</i> sp.			1		1	18	2	15	6	50
<i>Chaetomorpha</i> sp.	1	1			2	7	1		2	10
<i>Cladophora</i> sp.	1	2	1	1	5	5	10	4	6	25
<i>Corallina</i> sp.						2			1	3
<i>?Gelidium pusillum</i>							2			2
<i>Derbesia</i> sp.	2	3		1	6	10	9	4	7	30
<i>Dictyota divaricata</i>	1	4			5	11	15	6	5	37
<i>Dictyota</i> sp. 2						1				1
<i>Ectocarpus breviarticulatus</i>						1		1	3	5
<i>Enteromorpha</i> sp.									1	1
<i>Galaxaura comans</i>		1			1	12	1	3	2	18
<i>Gracilaria mammillaris</i>							2			2
<i>Halimeda monile</i>							1			1
<i>Heterosiphonia</i> sp.						1		1	1	3
<i>Hypnea spinella</i>		2			2				1	1
<i>Jania ?adherens</i>		1			1	16	10	11	12	49
<i>Laurencia obtusa</i>							1			1
<i>Laurencia papillosa</i>	2				2	2	1	1		4
Mixed algal turf	40	44	52	31	167	65	73	54	80	272
<i>Murrayella pericladus</i>		1		1						
<i>Penicillus capitatus</i>						1				1
<i>Polysiphonia</i> sp. 1						2	3	3	5	13
<i>Polysiphonia</i> sp. 2				2	2	1	1			2
<i>Polysiphonia</i> sp. 3							1			1
Red algal blade							1			1
<i>Rhizoclonium</i> sp.									2	2
<i>Struvea anastomosans</i>				3	3	2	7	4	2	15
<i>Ventricaria ventricosa</i>							2	2	1	5
Total taxa:	11	13	7	11	21	20	27	17	22	35
Total occurrences:	61	68	63	47	239	168	198	114	154	634

Table 7.6 Distribution of algal species or categories on open coasts, long-term census, third year after the spill. Data are for August 1988 - May 1989. N=80 roots/site, 320/habitat type. Site codes as in Table 7.1. Sum = total occurrences/category at oiled or unoiled sites. ".": taxon did not occur.

Category	Oiled					Unoiled				
	DROM	MINM	PGM	PMM	Sum	LINM	MSM	PADM	PBM	Sum
<i>Amphiroa ?fragilissima</i>	1	.	.	1
<i>Acanthophora spicifera</i>	9	3	3	3	18	5	13	2	2	22
<i>Bostrychia tenella</i>	4	1	9	9	23	.	14	.	.	14
<i>Bostrychia</i> sp.2	.	.	1	.	1
<i>Caloglossa letrieurii</i>	1	.	.	1
<i>Caulerpa fastigata</i>	2	5	1	3	11	8	6	25	2	41
<i>Caulerpa mexicana</i>	.	1	.	.	1	3	.	.	.	3
<i>Caulerpa racemosa</i>	3	.	.	3
<i>Caulerpa verticillata</i>	12	15	4	1	32	2	2	1	4	9
<i>Ceramium</i> sp.	3	3	2	3	11	11	14	2	9	36
<i>Chaetomorpha</i> sp.	2	1	1	.	4	4	3	4	5	16
<i>Cladophora</i> sp.	.	2	2	1	5	4	5	3	2	14
<i>Derbesia</i> sp.	.	.	.	1	1	.	4	1	2	7
<i>Dictyota divaricata</i>	4	3	1	3	11	11	9	6	4	30
<i>Ectocarpus breviararticulatus</i>	.	.	2	2	4	1	.	.	.	1
<i>Enteromorpha</i> sp.	1	.	.	.	1
<i>Galaxaura comans</i>	1	1	1	.	3	6	2	1	4	13
<i>Gelidiella acerosa</i>	1	3	.	.	4
<i>Halimeda opuntia</i>	1	.	.	.	1
<i>Herposiphonia secunda</i>	.	.	.	1	1	1	.	.	.	1
<i>Hypnea spinella</i>	.	2	.	.	2	1	2	1	.	4
<i>Jania ?adherens</i>	.	3	.	.	3	16	11	5	8	40
<i>Laurencia obtusa</i>	1	1
<i>Laurencia papillosa</i>	3	.	.	1	4
Mixed algal turf	48	49	39	36	172	68	76	66	71	281
<i>Murrayella pericladus</i>	1	1
<i>Neomeris annulata</i>	1	.	1	2
<i>Padina gymnospora</i>	1	.	.	.	1	.	1	1	.	2
<i>Polysiphonia</i> sp. 1	1	.	1
<i>Polysiphonia</i> sp. 2	1	.	1
<i>Polysiphonia</i> sp. 3	1	.	1	.	2
<i>Struvea anastomosans</i>	.	1	.	2	3	16	3	4	10	33
<i>Ventricaria ventricosa</i>	2	.	.	.	2
Total taxa:	10	14	12	12	19	21	20	17	16	32
Total occurrences:	86	90	66	65	307	166	174	125	127	592

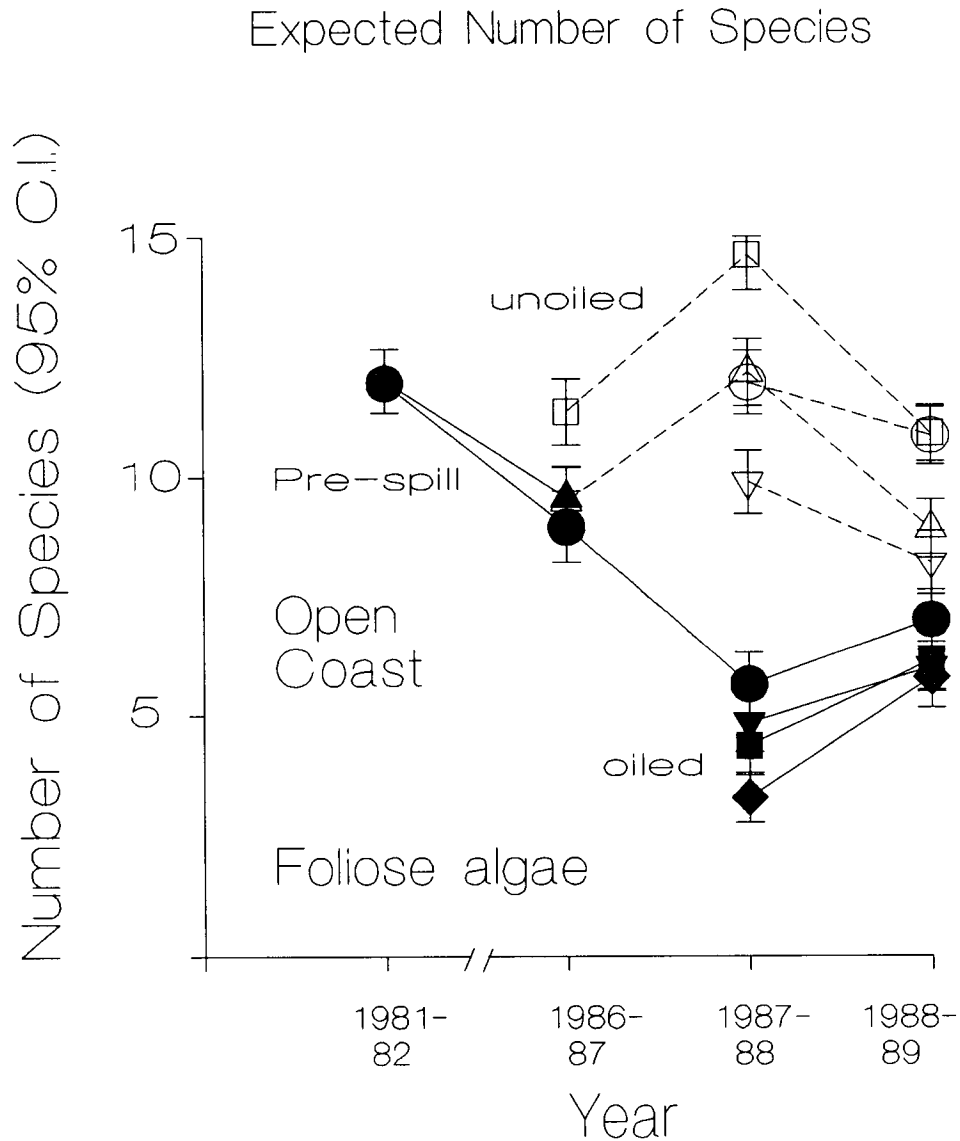


Figure 7.21 The number of categories of foliose algae (\pm 95% confidence intervals) expected to be found on a sample of 20 roots, calculated using rarefaction analysis. Each point represents a separate site. Values calculated using all roots sampled in a given year (8/86-5/87, 8/87-5/88, 8/88-5/89, $n=32-281$ roots/site). Oiled=filled symbols, unoiled=open symbols. Data from 1981-1982 sites (GALM and MINM) were unoiled; both were oiled in 1986. See text for further explanation.

Table 7.7 Distribution of algal species or categories on open coast community development roots for the third year after the spill. Data are for August 1988-May 1989. N=40 roots/site, 160/habitat type. Site codes as in Table 7.1. Sum=total occurrences/category at oiled or unoiled sites. ".": taxon did not occur.

Category	Oiled					Unoiled				
	DROM	MINM	PGM	PMM	Sum	LINM	MSM	PADM	PBM	Sum
<i>Acetabularia crenulata</i>	.	1	.	.	1	1	.	.	.	1
<i>Acanthophora spicifera</i>	3	4	3	2	12	.	9	.	2	11
<i>Bostrychia tenella</i>	1	.	.	1
Brown (tubular,branched)	1	1
<i>Caulerpa fastigata</i>	.	7	3	1	11	2	4	12	6	24
<i>Caulerpa mexicana</i>	1	.	.	.	1
<i>Caulerpa verticillata</i>	1	1	.	1	3	2	1	.	.	3
<i>Ceramium</i> sp.	2	5	2	.	9	2	4	.	3	9
<i>Chaetomorpha</i> sp.	.	1	.	.	1	3	4	.	4	11
<i>Cladophora</i> sp.	.	2	.	.	2	1	1	.	1	3
<i>Derbesia</i> sp.	1	.	1	2
<i>Dictyota divaricata</i>	3	3	.	2	8	1	14	1	3	19
<i>Dictyota</i> sp. 2	1	.	.	1
<i>Ectocarpus breviarticulatus</i>	1	.	.	1
<i>Enteromorpha</i> sp.	.	1	2	.	3
<i>Galaxaura comans</i>	.	4	1	.	5	2	1	.	.	3
<i>Gelidiella acerosa</i>	1	.	.	.	1	.	2	.	1	3
<i>Gracilaria mammillaris</i>	.	2	.	.	2
<i>Heterosiphonia</i> sp.	1	.	2	3
<i>Hypnea spinella</i>	.	3	.	.	3	.	1	.	.	1
<i>Jania ?adherens</i>	1	.	.	.	1	3	1	.	1	5
<i>Laurencia papillosa</i>	3	.	.	.	3
Mixed algal turf	26	28	24	20	98	40	37	33	32	142
<i>Padina gymnospora</i>	3	.	.	.	3
<i>Penicillus capitatus</i>	.	1	.	.	1	.	1	.	.	1
<i>Polysiphonia</i> sp. 2	.	.	1	.	1
<i>Spyridia filamentosa</i>	1	.	.	1
<i>Struvea anastomosans</i>	.	1	.	1	2	4	4	1	1	10
Total taxa:	8	15	7	6	19	13	20	4	12	24
Total occurrences:	40	64	36	27	167	65	90	47	58	260

Table 7.8 Rarefaction estimates of the number of taxa of foliose algae expected in a sample of 20 roots, open coast, community development, 1988-1989.

	Mean \pm 95% Confidence Interval
A. Oiled	
Isla Droque Mangrove	5.9 \pm 0.5
Peña Guapa Mangrove	5.3 \pm 0.5
Punta Muerto Mangrove	4.0 \pm 0.5
Isla Mina Mangrove	10.8 \pm 0.6
B. Unoiled	
María Soto Mangrove	13.0 \pm 0.7
Portobelo Mangrove	9.2 \pm 0.6
Isla Lintón Mangrove	9.3 \pm 0.5
Isla del Padre Mangrove	3.0 \pm 0.3

Recruitment

Percent cover on dowels measures settlement and subsequent survival over the preceding three months. Foliose algae and diatoms were the only common groups on the open coast (Figures 7.13, 7.14). Both were present in blooms, together covering almost 100% of the dowel surface. Essentially no crustose algae or sessile invertebrates were recorded (Figures 7.15, 7.17).

Patterns of species occurrence of foliose algae from August 1988 to May 1989 are shown in Table 7.9. Mixed algal turfs were the most abundant category on dowels, with rare occurrences of other species. The expected number of species of foliose algae on a sample of 20 dowels was 5.3 \pm 0.6 species (calculated from the sum of occurrences at four sites).

Open Coast Habitat -- Oiled Sites

Long-term Census

The effects of the oil spill on open coast roots have been both measurable and persistent (Figures 7.13-7.21). Oil was recorded on most roots three years after the spill (Figure 7.3); percent cover of oil averaged 10-15% in 1989 (Figure 7.4).

Table 7.9 Distribution of foliose algal species or categories on recruitment dowels, open coast, third year after the spill. Data for August 1988-May 1989. Site codes as in Table 7.1. Sum=total occurrences/category at oiled or unoiled sites. ".": taxon did not occur.

Category	Oiled					Unoiled				
	DROM	MINM	PGM	PMM	Sum	LINM	MSM	PADM	PBM	Sum
<i>Acanthophora spicifera</i>	2	.	.	.	2
<i>Bostrychia tenella</i>	.	.	2	.	2	.	2	.	.	2
<i>Bostrychia</i> sp.2	1	.	.	1
<i>Bryopsis pennata</i>	.	1	.	.	1
<i>Caulerpa fastigata</i>	.	.	.	3	3	1	.	1	.	2
<i>Caulerpa verticillata</i>	.	.	1	.	1
<i>Ceramium</i> sp.	1	.	.	3	4	.	.	.	1	1
<i>Dictyota divaricata</i>	3	.	.	2	5	.	1	.	1	2
<i>Gracilaria mammillaris</i>	.	1	.	.	1
<i>Hypnea spinella</i>	.	.	2	.	2
Mixed algal turf	15	14	15	13	57	14	12	14	13	53
<i>Padina gymnospora</i>	1	.	.	.	1
<i>Polysiphonia</i> sp. 1	1	2	2	.	5	1	1	1	5	8
<i>Polysiphonia</i> sp. 2	.	1	.	.	1	1	.	2	2	5
Total taxa:	5	5	5	4	12	5	5	4	5	9
Total occurrences:	21	19	22	21	83	19	17	18	22	76
Sample size:	18	17	19	18	72	19	18	19	15	71

The abundance of foliose algae was similar to that in unoiled areas for the first two monitorings after the spill (Figure 7.13). Since then, it has been lower in eight of 10 monitorings, with no indication of an increasing trend. Crustose algae essentially disappeared at oiled sites for 1.5 years following the spill; since then they have fluctuated similarly to crusts in unoiled areas, but at lower abundance (Figure 7.15). There was a bloom of diatoms immediately after the spill, growing on top of a layer of hardened oil (Figure 7.14). Diatom cover has since declined to about that found at unoiled sites. Sessile invertebrates (excluding barnacles and bivalves) dropped to <5% cover (Figures 7.17-7.19) and show little recovery to date.

The species composition of foliose and crustose algae differed strongly between oiled and unoiled roots. In 1986-1987, crustose coralline algae occurred on 43% of unoiled roots and 6% of oiled roots. In the second year after the spill, crustose coralline algae were present on 64% of unoiled roots compared to 24% of oiled roots, increasing to 82% and 41%, respectively, during 1988-1989. The same scale of differences persisted in other classes of crustose algae. Overall, crustose algae showed incomplete recovery from the effects of the spill after three years.

Species of foliose algae showed (1) changes in the average number of times a species was found and (2) differences in species composition between oiled and unoiled sites (Tables 7.4-7.6, Figure 7.21). The estimated number of species on a sample of 20 roots was consistently lower at oiled than unoiled sites through May 1989. Individual species in 1986-1987 were rarer at oiled than unoiled sites (408 vs. 665 records, Table 7.4). Only a few species, most commonly *Caulerpa verticillata* and *Acanthophora spicifera*, were found more often at oiled than unoiled sites. When pre- and post-spill patterns at Punta Galeta and Isla Mina were compared, differences in species present were strong (Table 7.10). *Ceramium*, *Chaetomorpha*, *Bostrychia tenella*, *Dictyota divaricata*, *Caloglossa letrieurii*, *Polysiphonia* and mixed algal turfs were all common in 1981-1982; these plants were rare or absent after the spill. For Isla Mina, mixed algal turfs were present on 46% of roots in 1981-1982, but only 24% in 1986-1987. The percent then increased to 52% in 1987-1988 and 62% in 1988-1989. Despite this evidence of partial recovery, three years after the spill foliose algae clearly had not regained either the abundance or number of species found in 1981-1982.

Sessile animals (shown excluding barnacles and bivalves) were similarly strongly affected by oiling (Figure 7.17-7.19, Tables 7.11-7.13). All groups dropped strongly in abundance compared to unoiled sites; only a relatively few arborescent hydroids and bryozoans were found at oiled sites in the first year after the spill. In 1987-1988, there was patchy, but unsustainable, cover of arborescent hydroids and bryozoans. However, through May 1989, few or no sponges, anemones, corals or tunicates were recorded at oiled sites.

Only one group has done better on oiled than unoiled open coast mangrove roots. Beginning in August 1987, oiled sites had a higher percent cover of barnacles, especially *Chthamalus* sp. (Figure 7.16). At unoiled sites, a few barnacles were usually present, but rarely was mean cover >1.0%. A possible explanation for this effect is the much reduced density of grazers, such as *Littorina angulifera*, in oiled areas. Grazers are known to affect settlement success in barnacles (e.g., Levings and Garrity 1984) and might be a cause of this pattern; experimental evidence is needed to separate the direct effects of oiling from possible secondary effects like those of grazers.

Community Development

Roots that entered the water after August 1987 showed a mixed pattern of percent cover when oiled and unoiled sites were compared. Oiled roots tended to have more bare space than unoiled ones (Figure 7.20) and about the same cover of diatoms (Figure 7.14). Sessile invertebrates were rare on both oiled and unoiled roots (Figure 7.17). Algal crusts increased in cover at about the same rate at both oiled and unoiled sites (Figure 7.15).

Table 7.10 Number of roots an alga appeared on before the oil spill (1981-1982) and in the first, second and third years after the oil spill.

Category	1981 -1982	Year 1	Category	1981 -1982	Year 1	Year 2	Year 3
Punta Galeta Mangrove			Isla Mina Mangrove				
<i>Acetabularia crenulata</i>		4	<i>Acetabularia crenulata</i>		3		
<i>Acanthophora spicifera</i>		21	<i>Amphiroa ?fragilissima</i>			1	
<i>Bostrychia tenella</i>	23		<i>Acanthophora spicifera</i>		52	2	3
<i>Caloglossa letrieurii</i>	10		<i>Bostrychia tenella</i>	30			1
<i>Caulerpa fastigata</i>		1	<i>Caloglossa letrieurii</i>	14			
<i>Caulerpa sertularioides</i>	6	3	<i>Caulerpa fastigata</i>		12		5
<i>Caulerpa verticillata</i>	19	45	<i>Caulerpa mexicana</i>		2		1
<i>Ceramium</i> sp.	12	6	<i>Caulerpa racemosa</i>		1	1	
<i>Chaetomorpha</i> sp.	6		<i>Caulerpa sertularioides</i>	4	8		
<i>Cladophora</i> sp.		1	<i>Caulerpa verticillata</i>	23	42	5	15
<i>Derbesia</i> sp.		1	<i>Ceramium</i> sp.	9	8		3
<i>Dictyota divaricata</i>	17	4	<i>Chaetomorpha</i> sp.	9	1	1	1
<i>Ectocarpus breviarticulatus</i>	7	3	<i>Cladophora</i> sp.		1	2	2
<i>Enteromorpha</i> sp.		1	<i>Codium repens</i>		2		
<i>Herposiphonia secunda</i>	10		<i>Derbesia</i> sp.		2	3	
<i>Heterosiphonia</i> sp.		5	<i>Dictyota divaricata</i>	15	14	4	3
<i>Jania ?adherens</i>		3	<i>Ectocarpus breviarticulatus</i>	5	2		
Mixed algal turf	14	19	<i>Enteromorpha</i> sp.		2		
<i>Padina gymnospora</i>		2	<i>Galaxaura comans</i>		2	1	1
<i>Polysiphonia</i> sp.1		1	<i>Gelidiella acerosa</i>		1		
<i>Polysiphonia</i> sp.3	14	19	<i>Gracilaria mammillaris</i>		1		
Red algal blade	10		<i>Herposiphonia secunda</i>	8			
Number of roots:	33	97	<i>Heterosiphonia</i> sp.		3		
			<i>Hypnea spinella</i>		4	2	2
			<i>Jania ?adherens</i>		1	1	3
			<i>Laurencia papillosa</i>		2		
			Mixed algal turf	17	67	44	49
			<i>Murrayella pericladus</i>			1	
			<i>Padina gymnospora</i>		4		
			<i>Penicillus capitatus</i>		1		
			<i>Polysiphonia</i> sp. 1		1		
			<i>Polysiphonia</i> sp. 2		1		
			<i>Polysiphonia</i> sp. 3	18	18		
			Red algal blade	12			
			<i>Rhizoclonium</i> sp.		10		
			<i>Spyridia filamentosa</i>		1		
			<i>Struvea anastomosans</i>				1
			Number of roots:	37	281	85	80

Table 7.11 Number of roots on which a group of sessile organisms occurred in oiled and unoiled habitats, long-term census, first year after the spill. Data for August 1986-May 1987. Sample sizes as in Table 7.4.

Group	Oiled	Unoiled
Open coast		
Anemones	12	4
Corals	0	7
Encrusting bryozoans	1	0
Arborescent hydroids & bryozoans	114	232
Sponges	12	91
Colonial tunicates	1	23
Solitary tunicates	3	3
Channels and lagoons		
Anemones	9	53
Encrusting bryozoans	33	173
Arborescent hydroids & bryozoans	75	81
Sponges	196	209
Colonial tunicates	51	42
Solitary tunicates	111	154
Drainage streams		
Anemones	0	2
Arborescent hydroids & bryozoans	0	42
Sponges	0	3
Solitary tunicates	0	16

Table 7.12 Distribution of groups of sessile organisms, long-term census, open coasts, second year after the spill. Data for August 1987 - May 1988. N=85 roots/site, 340/habitat type. Site codes as in Table 7.1. Sum=total occurrences/group in oiled or unoiled open sites. "Arb.": arborescent. "bryoz.": bryozoans. ".": taxon did not occur.

Category	Oiled					Unoiled				
	DROM	MINM	PGM	PMM	Sum	LINM	MSM	PADM	PBM	Sum
Anemones	.	1	.	1	2	2	3	8	1	14
Corals	4	6	2	2	14
Arb.hydroids, bryoz.	21	43	18	30	112	61	28	65	73	227
Sponges	2	10	2	11	25	22	16	29	35	102
Colonial tunicates	.	.	.	1	1	5	2	7	17	31
Solitary tunicates	.	1	.	1	2	.	.	1	3	4

Table 7.13 Distribution of groups of sessile organisms, long-term census, open coasts, third year after the spill. Data for August 1988 - May 1989. N=80 roots/site, 320/habitat type. Site codes as in Table 7.1. Sum=total occurrences/group in oiled or unoiled open sites. "Arb.": arborescent. "bryoz.": bryozoans. ".": taxon did not occur.

Category	Oiled					Unoiled				
	DROM	MINM	PGM	PMM	Sum	LINM	MSM	PADM	PBM	Sum
Corals	3	4	1	.	8
Encrusting bryozoans	1	1
Anemones	.	5	1	1	7	.	1	8	1	10
Arb. hydroids, bryoz.	12	38	17	23	90	41	12	44	54	151
Sponges	5	19	7	12	43	17	27	38	36	118
Colonial tunicates	1	2	2	1	6	4	.	9	24	37
Solitary tunicates	2	.	1	.	3	.	2	7	3	12

The strongest differences were in the cover and species composition of foliose algae (Figure 7.13, Tables 7.7, 7.8); these differences reflected those found on long-term census roots (see above). Percent cover at unoiled sites averaged ~20% since this substudy began in August 1987. At oiled sites, mean percent cover was approximately half that. Fewer species were represented overall, with two notable exceptions. At Isla Mina (oiled), the number of species of algae expected in a sample of 20 roots was 10.8 ± 0.6 , as high as that for the unoiled sites; at Isla del Padre (unoiled), the expected number of species was as low (3.0 ± 0.3) or lower than that estimated for oiled sites. The Isla Mina site thus appeared to be recovering more rapidly than the other three oiled sites. Total percent cover of foliose algae has been above 15% since August 1988, higher than other oiled sites, but lower than unoiled sites. Roots at Isla del Padre were dominated by mixed algal turfs. Percent cover of foliose algae was as high as that at other unoiled sites, but there were fewer species. The reason for this difference in unoiled sites is unknown.

Recruitment

The pattern of percent cover at oiled and unoiled sites has been very similar through May 1989 (Figures 7.13-7.15, 7.17, 7.20). Occasionally, oiled sites showed lower cover of crustose or foliose algae, but there was no trend for increased recruitment at unoiled sites. Recruitment dowels at oiled sites recorded similar species and numbers of occurrences of foliose algae (Table 7.9) as unoiled sites. The number of species of foliose algae expected to be found on a sample of 20 dowels was 6.4 ± 0.6 , approximately that calculated for unoiled areas.

Open Coast: Summary

Open coast mangroves are continually washed by wave action. Flushing re-sorts sediments and helps remove oil from the area. Thus open coast mangroves might be expected to show relatively quick signs of recovery. Although some plants and animals are returning to abundances similar to those found before the spill, the open coast showed persistent effects of the 1986 spill three years later. Differences in the development of the epibiotic assemblage on roots that entered the water in August 1987 still occur. Early differences in recruitment onto artificial roots have not persisted. There was no evidence of an algal bloom following the spill. Full recovery appears to be several years in the future.

Channels or Mangrove Lagoons -- Unoiled Sites

Long-term Census

Mangrove roots in unoiled channels and lagoons were characterized by bivalve molluscs, which covered up to 75% of the space on roots. The most abundant of these was the edible oyster *Crassostrea rhizophorae* (14-54% cover, Figure 7.22); four other bivalves were also relatively common (Figure 7.23, Table E.5). Because oyster valves detach well after the animal's death, dead *Crassostrea* covered 1-7% of the roots (Figure 7.24). The barnacle, *Balanus improvisus*, rare on the open coast, averaged between 4-18% cover on channel roots (Figure 7.25). Foliose algae were less abundant than on the open coast (5-12% cover, Figure 7.26) and diatoms covered $\leq 15\%$ of the root surface (Figure 7.27). There was relatively little unoccupied space (5-37% cover, Figure 7.28) on channel roots. The remainder was covered by various rare categories: sponges, anemones, blue-green algae, hydroids, vermetids, tunicates and bryozoans. Sessile invertebrates, excluding barnacles and bivalves, fluctuated in abundance but rarely exceeded a total cover of 10% (Figure 7.29).

One difference in percent cover taken in 1981-1982 compared with later samples is the almost total absence of foliose algae and sessile filter feeders other than molluscs and barnacles in 1981-1982. No foliose algae were recorded; no sponges or arborescent hydroids and bryozoans were present in three samples, and other sessile species were rare (total cover 2-4%). In 1986-1988 data, cover of sessile filter feeders at the same site (Hidden Channel) increased to 4-18% while foliose algae covered 1-13%. This difference may be due to increasing development (and subsequent nutrient enrichment) of the watershed of this lagoon.

Unoiled channels differed in the pattern of percent cover on roots depending upon location. The two sites in the lagoon between Punta Galeta and Isla Margarita had (1) lower cover of *Crassostrea* (Figure 7.30) and (2) more bare space

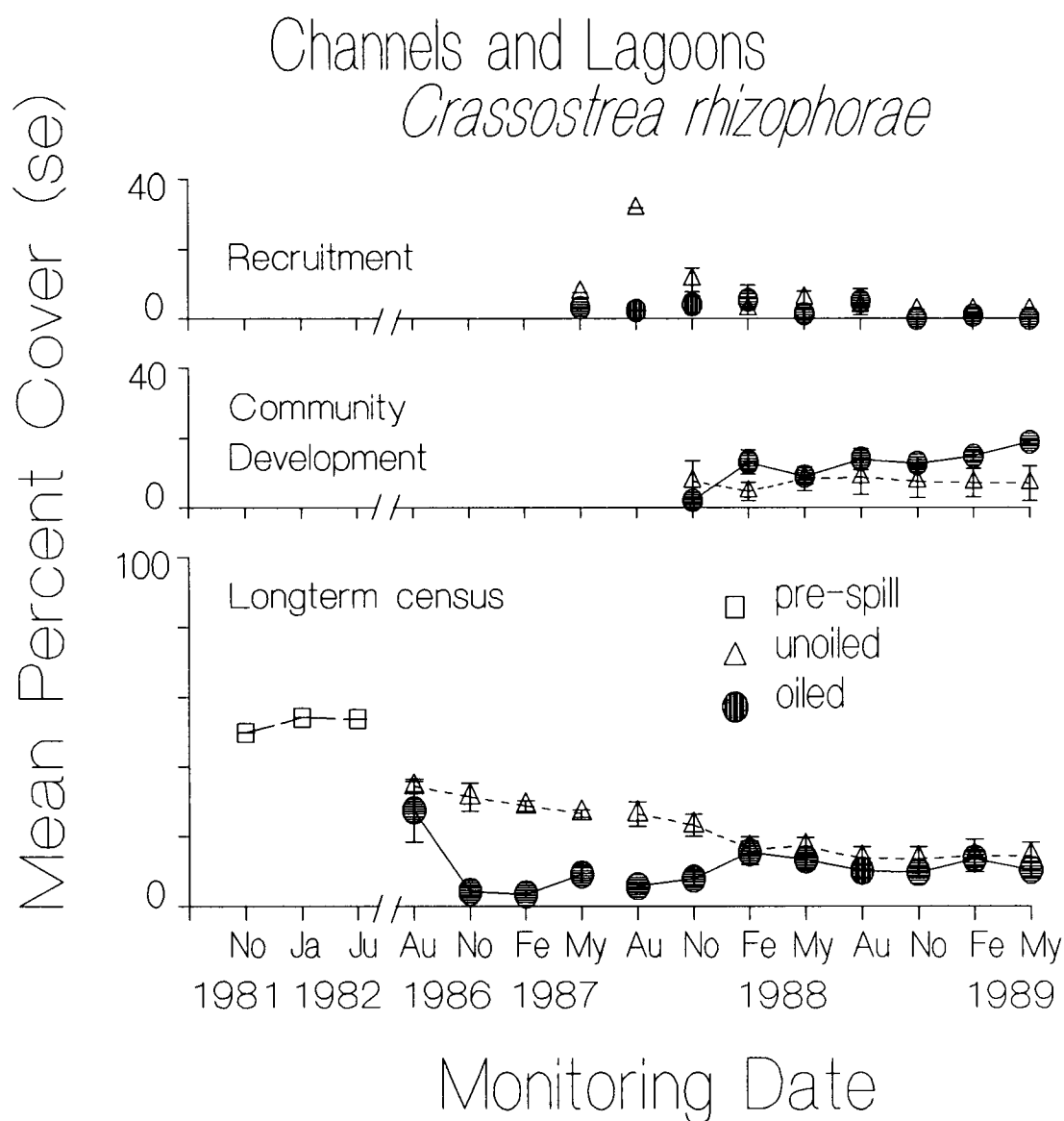


Figure 7.22 Abundance of *Crassostrea rhizophorae* on roots or dowels sampled from channels and lagoons. Data are stacked graphs for recruitment, community development and long-term census substudies. Symbols represent among site means of percent cover with standard error bars. N=1-5 sites/date. See text for details.

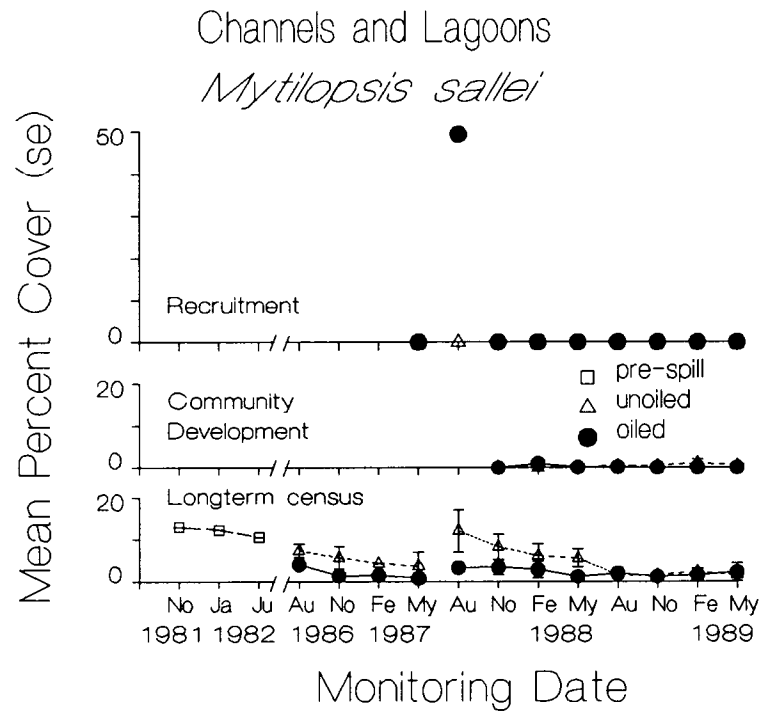


Figure 7.23 Abundance of *Mytilopsis sallei* on roots or dowels sampled from channels and lagoons. Symbols represent among site means with standard error bars. N=1-5 sites/date. See text for details.

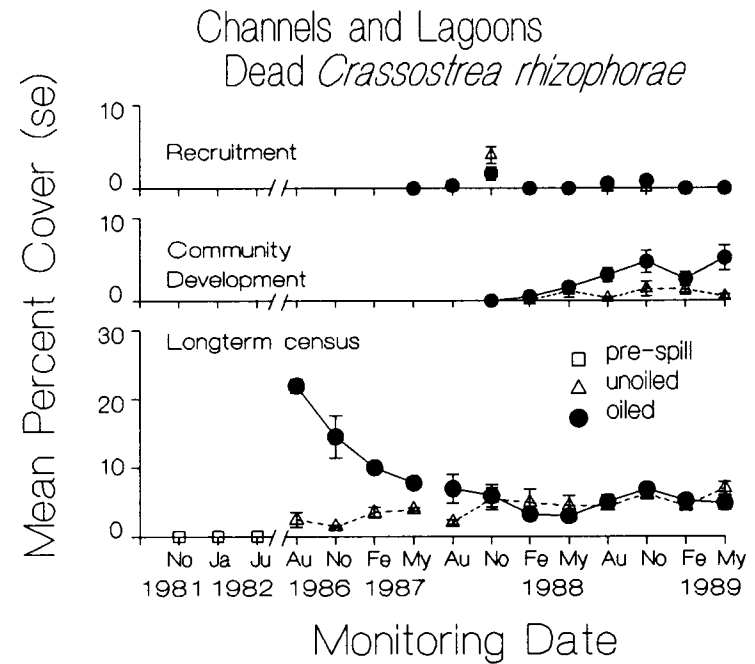


Figure 7.24 Abundance of dead *Crassostrea* on roots or dowels sampled from channels and lagoons. Symbols represent among site means with standard error bars. N=1-5 sites/date. See text for details.

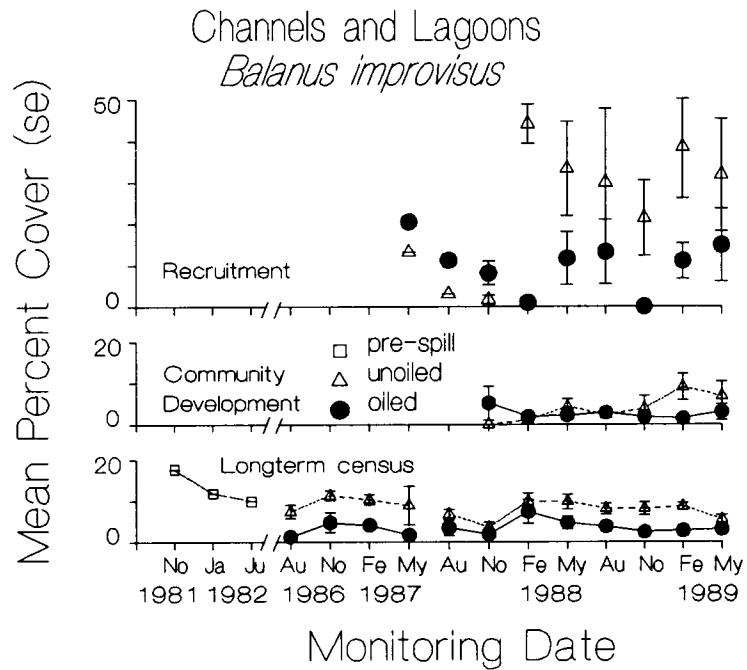


Figure 7.25 Abundance of *Balanus improvisus* on roots or dowels sampled from channels and lagoons. Symbols represent among site means with standard error bars. N=1-5 sites/date. See text for details.

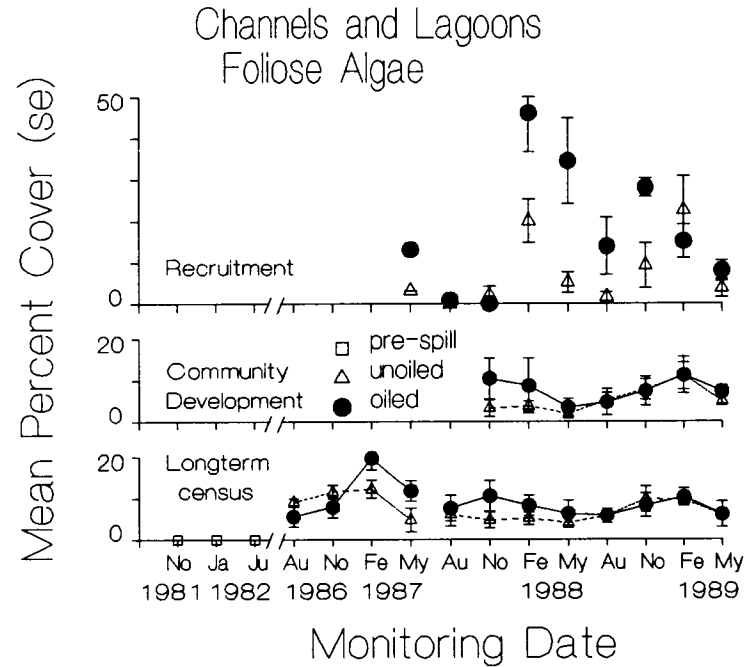


Figure 7.26 Abundance of foliose algae on roots or dowels sampled from channels and lagoons. Symbols represent among site means with standard error bars. N=1-5 sites/date. See text for details.

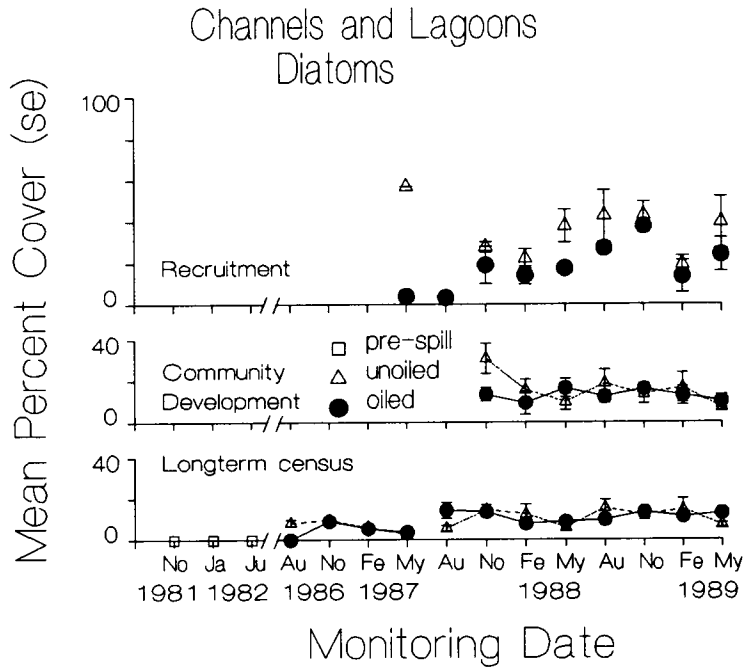


Figure 7.27 Abundance of diatoms on roots or dowels sampled from channels and lagoons. Symbols represent among site means with standard error bars. N=1-5 sites/date. See text for details.

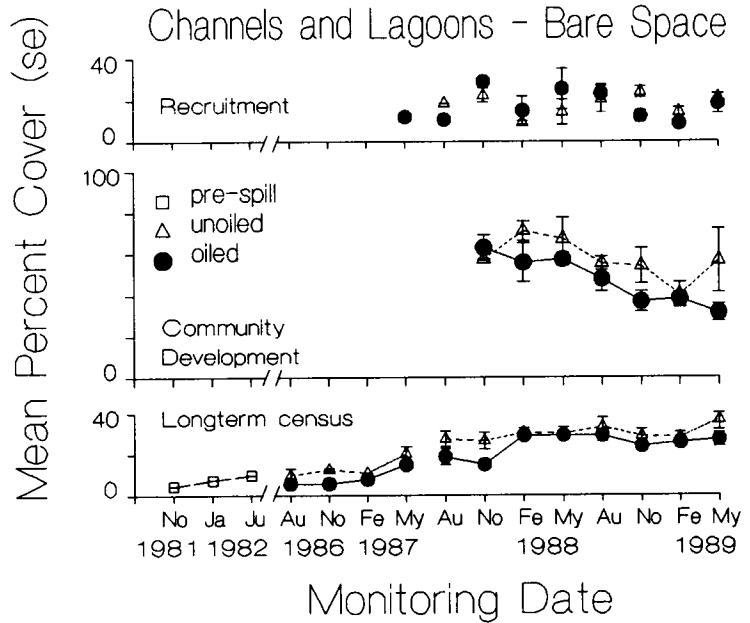


Figure 7.28 Abundance of bare space on roots or dowels sampled from channels and lagoons. Symbols represent among site means with standard error bars. N=1-5 sites/date. See text for details.

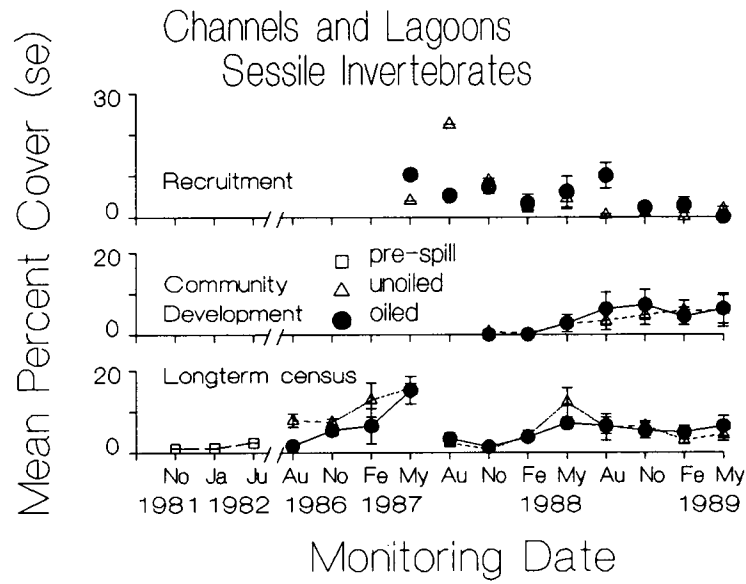


Figure 7.29 Abundance of sessile invertebrates excluding barnacles and bivalves on roots or dowels from channels and lagoons. Symbols represent among site means with standard error bars. N=1-5 sites/date. See text for details.

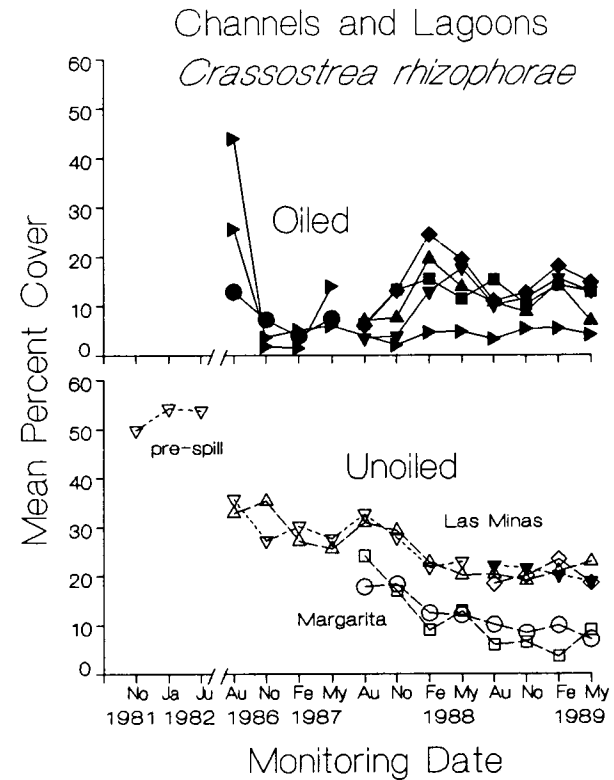


Figure 7.30 Abundance of *Crassostrea rhizophorae* by site on roots in long-term censuses from channels and lagoons. Symbols represent site means. N=15-50 roots/site. HIDC, which was secondarily oiled between May and August 1988, is indicated by filled symbols in the lower panel. See text for details.

(Figure 7.31) on roots than at the unoled sites in Bahía Las Minas (Figure 7.2). In both areas, there was a decreasing trend in cover of *Crassostrea* since 1986, but because oysters were initially rarer, percent cover in Margarita is now <10%. The cause of these differences is unknown, but we suggest that a limited water exchange in the more confined Margarita lagoon is a contributing factor.

Foliose algae rarely covered >10% of the root surface (Figure 7.26). However, more than 20 species have been found to date. Mixed algal turfs, *Polysiphonia* spp., *Caulerpa* spp., and *Bostrychia* spp. were all common (Tables 7.4, 7.14-7.15). The number of species of algae expected on a sample of 20 roots was approximately half that on the open coast (Figure 7.21, Table 7.16).

Community Development

Some parts of the assemblage found on long-term census roots were slow to develop on roots that entered the water in August 1987. Bare space decreased over time, but over 50% of the root surface was still empty in May 1989 (Figure 7.28). Coverage of *Crassostrea* was <10% through May 1989 (Figure 7.22) and few dead valves were recorded (Figure 7.24). As with long-term census roots, fewer *Crassostrea* were recorded in Margarita Lagoon (MACS, MACN) than in Bahía Las Minas (HIDC, SBCW, Figure 7.32). Cover of *Balanus* was almost as high as that on long-term census roots by February and May 1989 (Figure 7.25). Sessile invertebrates, excluding barnacles and bivalves, reached covers similar to those on long-term census roots in August 1988 (Figure 7.29). Diatom and algal cover have also been similar to that on long-term census roots since that time (Figures 7.26, 7.27).

When the species of foliose algae present on community development roots were examined (Table 7.17), they were a subset of those found on long-term census roots (Tables 7.14-7.15). *Polysiphonia* was common, but *Bostrychia* was relatively rare. The number of species expected on a sample of 20 roots was slightly lower than that calculated for long-term census roots (Tables 7.16, 7.18).

Recruitment

Estimates using artificial roots show either (a) low recruitment with occasional peaks or (b) high variability in recruitment each monitoring period. Both *Crassostrea* and *Mytilopsis* were abundant at only one monitoring (August 1987, Figures 7.22, 7.23); sessile invertebrates were also abundant on dowels during this period (Figure 7.29). Cover of *Balanus* was low in 1987, but has been patchy and high throughout 1988-1989 (Figure 7.25). Settlement of *Balanus* on artificial roots has not been followed by a correlated increase in barnacle cover on either long-term census or community development roots, probably because of predation (S. D. Garrity, personal observations).

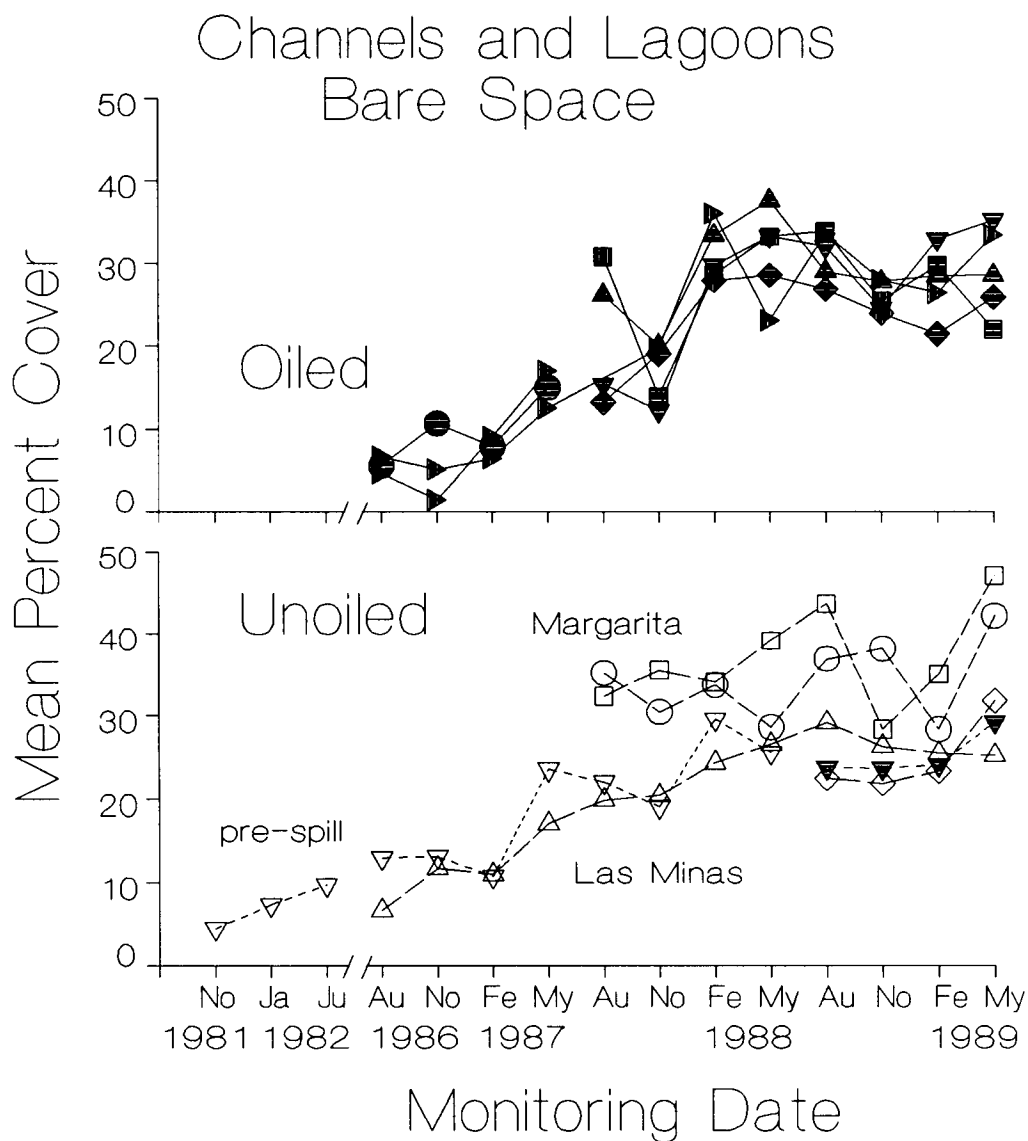


Figure 7.31 Abundance of bare space on roots sampled in long-term censuses by site in channels and lagoons. Symbols are site means of percent cover, $n=15-50$ roots each monitoring. HIDC, which was secondarily oiled between May and August 1988, is indicated by filled symbols in the lower panel. See text for details of sample sizes.

Table 7.14 Distribution of algal species or categories in channels and lagoons, long-term census, second year after the spill. Data for August 1987-May 1988. Site codes as in Table 7.1. Sum=total occurrences/category in oiled or unoiled channels. ".": taxon did not occur.

Category	Oiled					Unoiled					Sum
	LRCW	PCE	PCS	SBCE	SBCS	HIDC	MACN	MACS	SBCW		
<i>Amphiroa ?fragilissima.</i>	2	.	2	1	.	5	2	4	.	4	10
<i>Acanthophora spicifera</i>	.	.	2	4	.	6
<i>Bostrychia tenella</i>	.	2	.	.	.	2	2	.	8	2	12
<i>Bostrychia</i> sp.2	.	6	.	.	.	6	1	.	2	1	4
<i>Caloglossa letrieurii</i>	1	1	2	.	14	.	16
<i>Caulerpa fastigata</i>	4	1	1	3	2	11	8	3	.	8	19
<i>Caulerpa verticillata</i>	4	12	.	12	1	29	.	2	.	.	2
<i>Ceramium</i> sp.	.	4	.	.	.	4	.	.	1	.	1
? <i>Gelidium pusillum</i>	3	.	.	1	.	4	9	.	4	.	13
<i>Dictyota divaricata</i>	.	2	.	.	.	2
<i>Enteromorpha</i> sp.	.	.	.	2	1	3
<i>Gracilaria mammillaris</i>	1	.	1
<i>Herposiphonia secunda</i>	1	3	.	.	4
<i>Heterosiphonia</i> sp.	7	1	8	.	.	16	1	1	.	1	3
Mixed algal turf	1	1
<i>Polysiphonia</i> sp. 1	7	12	8	4	7	38	14	9	17	10	50
<i>Polysiphonia</i> sp. 2	11	32	22	9	2	76	6	5	13	8	32
<i>Polysiphonia</i> sp. 3	1	10	9	.	2	22	1	6	6	1	14
Total taxa:	9	10	7	8	6	15	11	8	9	9	15
Total occurrences:	40	82	52	36	15	225	47	33	66	36	182
Sample size:	60	85	85	85	85	380	85	85	85	85	320

Foliose algae and diatoms were patchily abundant on dowels (Figures 7.26, 7.27). Mixed algal turfs were the predominant algal group (Table 7.19). Using rarefaction, an estimated 4.6 ± 0.6 species of foliose algae were expected to be found on a sample of 20 dowels.

Channel and Lagoons -- Oiled Sites

Long-term Census

Oiling within channels was patchy, with less average percent cover immediately after the oil spill than was found on the open coast (Figure 7.4). However, in May 1989 oil still covered 18% of root surfaces. Decreases in oil cover have been slow.

Table 7.15 Distribution of algal species or categories in channels and lagoons, long-term census, third year after the spill. Data for August 1988-May 1989. N=80 roots/site. Site codes as in Table 7.1. Sum=total occurrences/category at oiled or unoiled sites. ".": taxon did not occur.

Category	Newly Oiled			Oiled			Unoiled					
	HIDC	LRCW	PCE	PCS	SBCE	SBCS	Sum	LRCS	MACN	MACS	SBCW	Sum
<i>Acetabularia crenulata</i>	.	.	1	.	.	.	1
<i>Acanthophora spicifera</i>	.	.	7	4	.	.	11	9	1	.	.	10
<i>Bostrychia tenella</i>	2	1	2	2	.	.	5	2	.	.	2	4
<i>Bostrychia</i> sp.2	4	.	8	1	.	.	9	3	.	4	4	11
<i>Caloglossa letrieurii</i>	6	3	3	1	.	16	.	17
<i>Caulerpa fastigata</i>	3	2	5	10	5	8	30	9	8	10	3	30
<i>Caulerpa mexicana</i>	.	.	3	.	1	.	4
<i>Caulerpa verticillata</i>	.	10	29	3	21	3	66	4	.	.	6	10
<i>Ceramium</i> sp.	.	.	2	.	.	.	2	.	1	1	1	3
<i>Chaetomorpha</i> sp.	1	.	1
<i>Cladophora</i> sp.	.	3	1	.	1	.	5
<i>Ectocarpus breviarticulatus</i>	.	.	1	.	.	.	1	.	.	.	1	1
? <i>Gelidium pusillum</i>	1	1	2
<i>Herposiphonia secunda</i>	.	.	.	1	.	.	1
Mixed algal turf	.	.	6	3	1	.	10	1	3	14	2	20
<i>Neomeris annulata</i>	.	.	1	.	.	.	1
<i>Polysiphonia</i> sp. 1	6	12	21	11	10	15	69	21	9	13	19	62
<i>Polysiphonia</i> sp. 2	1	6	14	8	1	9	38	14	5	1	8	28
<i>Polysiphonia</i> sp. 3	3	1	8	4	.	.	13	1	12	5	1	19
Red (fine, branched)	3	.	3
Red algal blade	1
Total taxa:	8	8	15	10	8	4	18	10	7	10	11	14
Total occurrences:	27	38	109	47	41	35	269	65	39	68	48	220

At unoiled sites, *Crassostrea* and *Balanus* together covered ~30-45% of root surfaces in 1986-1989 (Figures 7.22, 7.25). At oiled sites, the abundance of these species fell after oiling and continued to drop for the next year. Dead *Crassostrea* covered 20% of the oiled channel roots sampled in August 1986 (vs. 1-2% in unoiled areas; Figure 7.24); these valves gradually detached from the roots, or the roots themselves died and broke after oiling. Cover of dead *Crassostrea* subsequently declined. The cover of dead *Crassostrea* has been approximately the same at oiled and unoiled sites since November 1987.

Table 7.16 Rarefaction estimates of the number of taxa of foliose algae expected in a sample of 20 roots: channels and lagoons, long-term census.

	Mean \pm 95% Confidence interval		
	8-86 - 5-87	8-87 - 5-88	8-88 - 5-89
A. Oiled			
Peña Guapa Channel	6.5 \pm 0.6	-	-
Largo Remo Channel Site CO3	7.5 \pm 0.6	-	-
Largo Remo Channel West	6.0 \pm 0.6	6.5 \pm 0.5	4.9 \pm 0.5
Samba Bonita Channel East	-	4.7 \pm 0.5	3.7 \pm 0.4
Samba Bonita Channel South	-	2.6 \pm 0.5	3.4 \pm 0.3
Payardi Channel East	-	6.7 \pm 0.5	8.7 \pm 0.5
Payardi Channel South	-	4.8 \pm 0.4	6.3 \pm 0.6
B. Newly oiled			
Hidden Channel	-	-	4.2 \pm 0.5
C. Unoiled			
Hidden Channel	7.0 \pm 0.5	5.8 \pm 0.6	-
Samba Bonita Channel West	7.8 \pm 0.5	4.8 \pm 0.5	5.9 \pm 0.6
Margarita Channel South	-	6.2 \pm 0.5	6.7 \pm 0.5
Margarita Channel North	-	4.9 \pm 0.5	4.7 \pm 0.4
Largo Remo Channel South	-	-	6.3 \pm 0.5

By May 1989, there were some signs of recovery. At three of four oiled sites, cover of *Crassostrea* began increasing in November 1987 or February 1988. Oyster cover averaged ~10% in May 1989 (Figure 7.30). This was less than half the level found at two unoiled sites and one "newly oiled" site, also in Bahía Las Minas. Similarly, cover of *Balanus* in oiled channels was about half (2-4%) that of unoiled channels (6-8%) the entire third year after the oil spill.

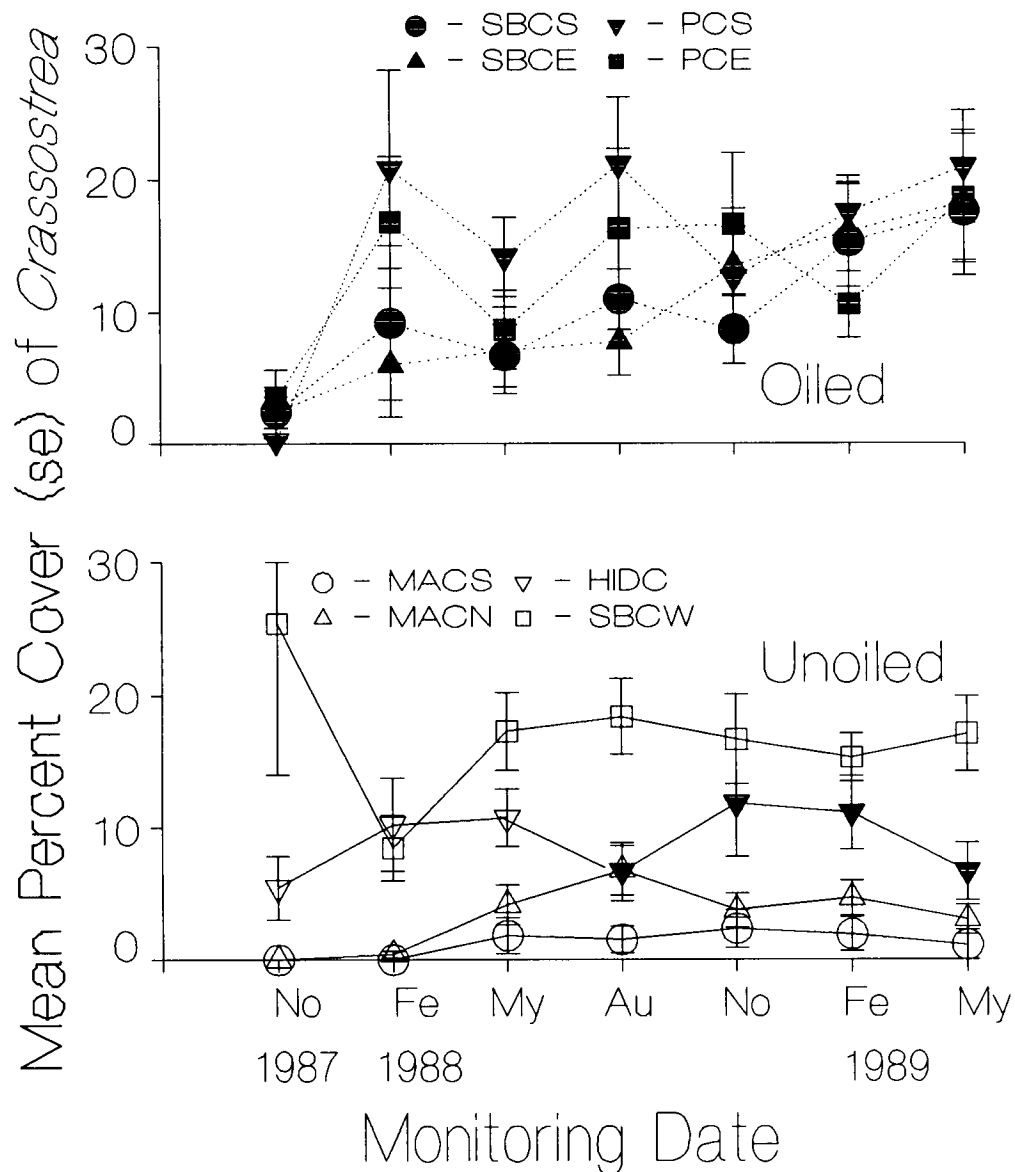


Figure 7.32 Abundance of *Crassostrea rhizophorae* on roots sampled in the community development substudy by site in channels and lagoons. Symbols are site means of percent cover, $n=5-10$ roots each monitoring. HIDC, which was secondarily oiled between May and August 1988, is indicated by filled symbols in the lower panel. See text for details of sample sizes.

Table 7.17 Distribution of algal species or categories in channels and lagoons, community development substudy, third year after the spill. Data for August 1988-May 1989. N=40 roots/site. Site codes as in Table 7.1. Sum=total occurrences/category at oiled or unoiled sites. ".": taxon did not occur.

Category	Newly oiled	Oiled					Unoiled			
	HIDC	PCE	PCS	SBCE	SBCS	Sum	MACN	MACS	SBCW	Sum
<i>Acanthophora spicifera</i>	.	2	1	1	.	4
<i>Bostrychia tenella</i>	.	2	.	.	.	2
<i>Bostrychia</i> sp.2	.	3	.	.	.	3	.	1	1	2
<i>Caloglossa letieri</i>	9	1	10
<i>Caulerpa fastigata</i>	.	6	1	1	1	9	2	.	2	4
<i>Caulerpa verticillata</i>	.	10	.	4	.	14	.	.	1	1
<i>Ceramium</i> sp.	1	.	1
<i>Cladophora</i> sp.	.	.	.	2	1	3
<i>Ectocarpus breviarticulatus</i>	.	1	.	.	.	1
<i>Enteromorpha</i> sp.	.	.	.	1	.	1
? <i>Gelidium pusillum</i>	.	2	.	.	.	2
Mixed algal turf	1	4	2	.	.	6	.	4	.	4
<i>Neomeris annulata</i>	.	3	.	.	.	3
<i>Polysiphonia</i> sp. 1	2	9	8	4	10	31	5	5	19	29
<i>Polysiphonia</i> sp. 2	.	6	9	.	2	17	1	2	7	10
<i>Polysiphonia</i> sp. 3	1	4	1	.	.	5	5	5	1	11
Total taxa:	3	12	6	6	4	14	4	7	7	9
Total occurrences:	4	52	22	13	14	101	13	27	32	72

Table 7.18 Rarefaction estimates of the number of taxa of foliose algae expected in a sample of 20 roots: channels and lagoons, community development substudy, 1988-1989.

	Mean \pm 95% Confidence interval
A. Oiled	
Samba Bonita Channel East	4.2 \pm 0.5
Samba Bonita Channel South	2.8 \pm 0.4
Payardi Channel East	10.4 \pm 0.4
Payardi Channel South	4.3 \pm 0.4
B. Newly oiled	
Hidden Channel	1.8 \pm 0.4
C. Unoiled	
Margarita Channel South	5.7 \pm 0.4
Margarita Channel North	3.2 \pm 0.3
Samba Bonita Channel West	4.8 \pm 0.5

Cover of foliose algae showed a different pattern (Figure 7.26). In the first two monitorings after the spill, slightly more foliose algae were found at unoiled sites. In February 1987, cover increased at oiled sites and remained at or slightly above cover at unoiled sites through May 1988. Algal species composition shifted with oiling, but results were different from those found on the open coast (Tables 7.4, 7.14-7.15). The number of species recorded and the number of roots individual species were found on were both higher in oiled than unoiled sites in the first year after the spill, returning to approximate equality in the second and third years after the spill. Rarefaction estimates suggest no overall change in species number at oiled sites (Table 7.16). However, there were differences in species composition when oiled and unoiled habitats were compared (Tables 7.4, 7.14-7.15). The same species that disappeared on the open coast were not recorded or were rare in oiled channels (e.g., *Caloglossa letrieurii* and *Bostrychia tenella*); *Caulerpa verticillata* also increased in oiled channels. These differences in species composition were still present, but less obvious, in 1988 and 1989.

When foliose algae were considered, Payardi Channel East (PCE) stood out from the other oiled sites. Percent cover was high (12-18%) and more species and occurrences of foliose algae were recorded there (Tables 7.14-7.15). Rarefaction estimates support this conclusion (Table 7.16). We do not know why this site has had consistently high populations of foliose algae compared to other oiled and unoiled channels.

Table 7.19 Distribution of algal species or categories in channels and lagoons, recruitment substudy, third year after the spill. Data for August 1988-May 1989. Site codes as in Table 7.1. Sum=total occurrences/category at oiled or unoiled sites. ".": taxon did not occur.

Category	Newly oiled			Oiled			Unoiled				
	HIDC	PCE	PCS	SBCE	SBCS	Sum	LRCS	MACN	MACS	SBCW	Sum
<i>Acetabularia crenulata</i>	.	1	.	2	.	3
<i>Acanthophora spicifera</i>	.	.	2	.	.	2	3	.	.	.	3
<i>Bostrychia</i> sp.2	1	.	1
<i>Caloglossa letrieurii</i>	1	.	1
<i>Caulerpa fastigata</i>	.	3	.	.	.	3	.	.	.	2	2
<i>Caulerpa verticillata</i>	.	.	.	1	.	1
<i>Ceramium</i> sp.	.	.	5	.	.	5
<i>Cladophora</i> sp.	1	.	.	1
Mixed algal turf	5	9	8	8	6	31	8	12	4	5	31
<i>Polysiphonia</i> sp. 1	.	5	7	2	2	16	.	.	.	2	2
<i>Polysiphonia</i> sp. 2	1	1	.	.	.	2	2
Red (fil., branched)	1	.	1
Total taxa:	1	4	4	4	3	8	2	2	4	4	9
Total occurrences:	5	18	22	13	9	62	11	13	7	11	42
Sample size:	17	16	18	17	18	69	11	16	19	16	62

It is possible there was a small algal bloom following oiling, with increases in oiled areas of a subset of algal species. Trees along channels were entirely or partially defoliated; measurement of the percent of surface illumination suggests light levels increased by a factor of 2-4 (Table 7.2, but this factor did not account for differences between Payardi Channel East and other sites). In addition, no or very few grazing snails were present and other changes (i.e., lack of *Crassostrea* as a major component on roots) occurred in oiled channels. The sum of these differences may have had a net positive effect on algae, despite any negative effects of oiling.

Sessile animals were less dramatically reduced in abundance in oiled channels than at open sites (Figure 7.29, Tables 7.11, 7.20-7.21). Encrusting bryozoans were found 41 times before the spill; they were much rarer at oiled than unoiled sites three years after oiling. Tunicates were still uncommon in oiled areas in May 1988, but had increased by May 1989. Sponges were more abundant at oiled than at unoiled sites in both 1988 and 1989.

Table 7.20 Distribution of groups of sessile organisms in channels and lagoons, long-term census, second year after the spill. Data for August 1987-May 1988. N=85 roots/site, 340/habitat type. Site codes as in Table 7.1. Sum = total occurrences/group in oiled or unoiled channels. "Arb.": arborescent. ".": taxon did not occur.

Group	Oiled					Unoiled				Sum
	PCE	PCS	SBCE	SBCS	Sum	HIDC	MACN	MACS	SBCW	
Anemones	.	1	8	2	11	8	2	.	7	17
Encrusting bryozoans	1	9	6	12	28	33	29	16	35	113
Arb. hydroids, bryozoans	6	20	17	14	57	11	13	4	23	51
Sponges	40	35	16	10	101	19	12	3	35	69
Colonial tunicates	1	10	1	.	12	20	9	1	5	35
Solitary tunicates	1	13	5	2	21	25	16	4	28	73

Table 7.21 Distribution of groups of sessile organisms in channels and lagoons, long-term census, third year after the spill. Data for August 1988-May 1989. N=80 roots/site. Site codes as in Table 7.1. Sum = total occurrences/group in oiled or unoiled channels. "Arb.": arborescent. ".": taxon did not occur.

Group	Newly Oiled	Oiled					Unoiled					Sum
	HIDC	LRCWP	PCE	PCS	SBCE	SBCS	Sum	LRCS	MACN	MACS	SBCW	
Anemones	.	4	1	2	1	3	11	10	.	.	15	25
Encrusting bryozoans	27	13	2	11	3	3	32	31	22	17	46	116
Arb. hydroids, bryozoans	5	14	11	15	2	8	50	18	9	14	19	60
Sponges	8	6	52	64	22	12	156	37	9	2	49	97
Colonial tunicates	9	.	14	7	4	5	30	6	4	.	2	12
Solitary tunicates	25	2	11	23	1	3	40	19	8	.	15	42

Community Development

Percent covers of major groups on roots that entered the water in August 1987 were similar between oiled and unoiled sites for most species and groups (Figures 7.22-7.30). Dead *Crassostrea* were more abundant at oiled than unoiled sites (Figure 7.24); *Balanus* was slightly more abundant at unoiled sites in 1989 (Figure 7.25). *Crassostrea* cover increased more slowly at unoiled than oiled sites (Figure 7.22). At unoiled sites, development of *Crassostrea* has essentially failed in Margarita Lagoon (sites MACN, MACS in Figure 7.32, see also Figure 7.30). Hidden Channel (HIDC) was secondarily oiled between May and August 1988, perhaps causing the small observed decrease in cover of oysters. Only at one unoiled site, Samba Bonita Channel West (SBCW), has cover of *Crassostrea* reached ~20%. This is similar to the oiled channel sites, where *Crassostrea* cover increased through May 1989 (Figure 7.32).

Covers of foliose algae have been almost identical at oiled and unoiled sites in 1988-1989; these results were similar to those for long-term census roots. Species composition of roots was a subset of that found on long-term census roots (Tables 7.4, 7.14, 7.15, 7.17). As on long-term census roots, *Caloglossa letrieurii* was rare and *Caulerpa verticillata* common at oiled sites. Rarefaction estimates (Table 7.18) suggest few differences between oiled and unoiled sites. One site stands out from the others. Payardi Channel East had higher numbers of both species and occurrences than the other oiled sites; this was also similar to results for long-term census roots.

Recruitment

Artificial roots differed widely in percent cover from one sample date to the next (Figures 7.22-7.30). Only one large settlement of bivalves occurred in oiled and unoiled channels (August 1987, Figures 7.22, 7.23). *Mytilopsis*, which is normally not abundant in channels (Figure 7.23) covered up to 60% of the artificial roots. This settlement did not persist; *Mytilopsis* cover did not subsequently increase on roots in these channels. There were large differences between oiled and unoiled sites in percent cover of some groups at some sampling dates. In 1988-1989, *Balanus* recruitment was dense at unoiled sites, but was rare at oiled sites (Figure 7.25), while the opposite was true of foliose algae (Figure 7.26). The same species and groups of foliose algae were found at oiled and unoiled sites (Table 7.19); rarefaction estimates were slightly higher for oiled sites (5.2 ± 0.5 species/20 dowels at oiled sites, 4.6 ± 0.6 species/20 dowels at unoiled sites). These data illustrate the patchiness of settlement processes, but their contribution to the recovery process cannot yet be evaluated.

Channel and Lagoons -- Summary

Channels and lagoons still showed effects of oiling in May 1989. However, variation among control sites (particularly the two Margarita sites, where recruitment for most of the species normally found in channels and lagoons has failed) tended to decrease the magnitude of oiling effects. *Crassostrea* and *Balanus* have not recovered former population levels, although percent cover showed an increasing trend. Oil still covered ~18% of the roots in May 1989. Sessile animals were present, but major groups differed in abundance between oiled and unoiled areas. Algal cover was equivalent in oiled and unoiled areas, but species composition still differed in May 1989. Prospects for further recovery of this habitat depend on the tolerance of taxa like *Crassostrea* to reoiling by residual oil leaking from sediments as well as recovery of the roots themselves.

Drainage Streams and Rivers -- Unoiled Sites

Long-term Census

A bivalve mollusc, the false mussel *Mytilopsis sallei*, was the most abundant organism in drainage streams (25-65%, Figure 7.33). *Crassostrea*, the most common species in channels, was rare (<1.5% cover). The barnacle *Balanus improvisus* was common through August 1987, but has been virtually absent since (Figure 7.34). Diatoms covered 10-26% of root surfaces in 1986-1989 (Figure 7.35) and foliose algae covered 5-11% during the same period (Figure 7.36). Bare space averaged 15-34%, similar to its abundance on open coasts (Figure 7.37). Sessile invertebrates, excluding barnacles and bivalves, showed strong variation among years (Figure 7.38). All groups were rare until February 1988 when cover increased to 8%; percent cover of sessile invertebrates has remained above 5% since that time. Rare space occupants, including amphipod tubes, vermetids, another species of barnacle, dead mussel shells and empty barnacle tests accounted for the remainder.

Mussel cover differed among the four rivers sampled starting in August 1987. In particular, Quebrada Las Mercedes averaged ~20% cover of *Mytilopsis* while the other three rivers have had covers greater than 40% (Figure 7.39). Mussels were clustered at intermediate depth on roots at Quebrada Las Mercedes; differences in the physical structure of this site may explain its relatively low mussel cover.

There were two differences between samples from 1981-1982 and those collected starting in 1986. At one unoiled river site (Hidden River), diatom cover was <5% in 1981-1982. Since then, the lowest percent cover of diatoms at Hidden River has been 12% (range 12-32%). Diatom cover has been >6% at all sites in all samples taken since 1986 (Figure 7.34). Sessile animals were not recorded in 1981-1982 samples; most groups were rare until 1988 (Figure 7.38).

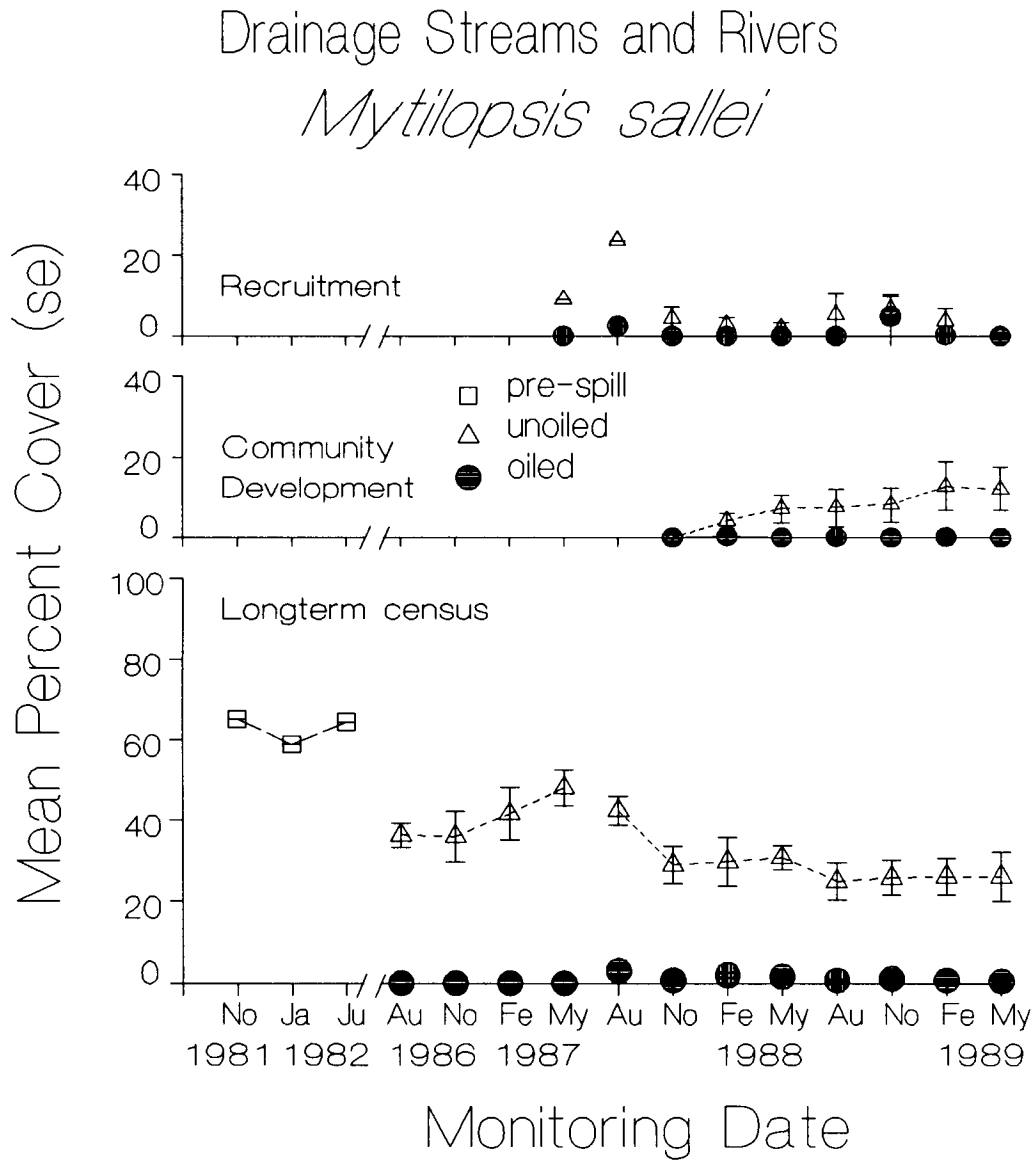


Figure 7.33 Abundance of *Mytilopsis salleii* on roots and dowels sampled from drainage streams and rivers. Symbols represent among site means of percent cover with standard error bars. N=1-4 sites/date. See text for details of sample sizes.

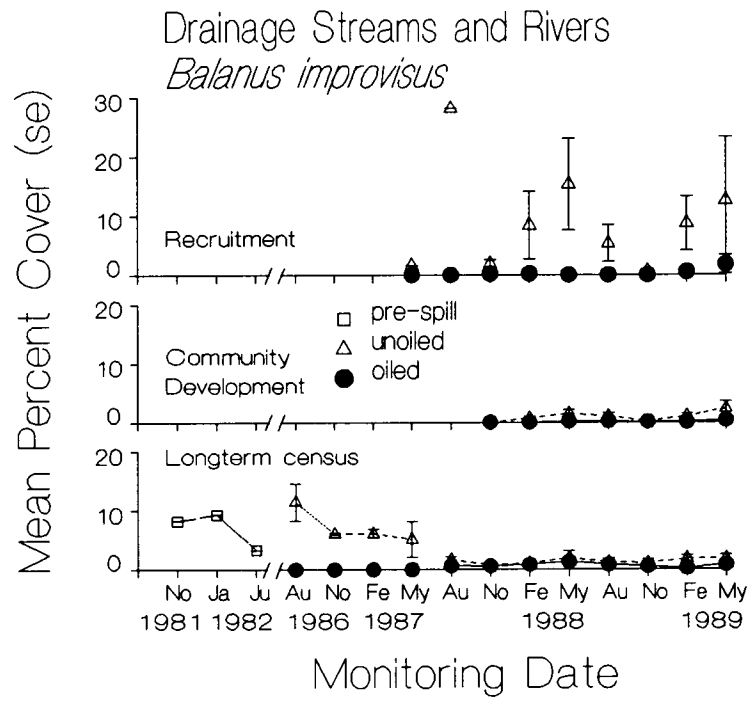


Figure 7.34 Abundance of *Balanus improvisus* on roots or dowels from drainage streams and rivers. Symbols represent among site means with standard error bars. N=1-4 sites/date. See text for details.

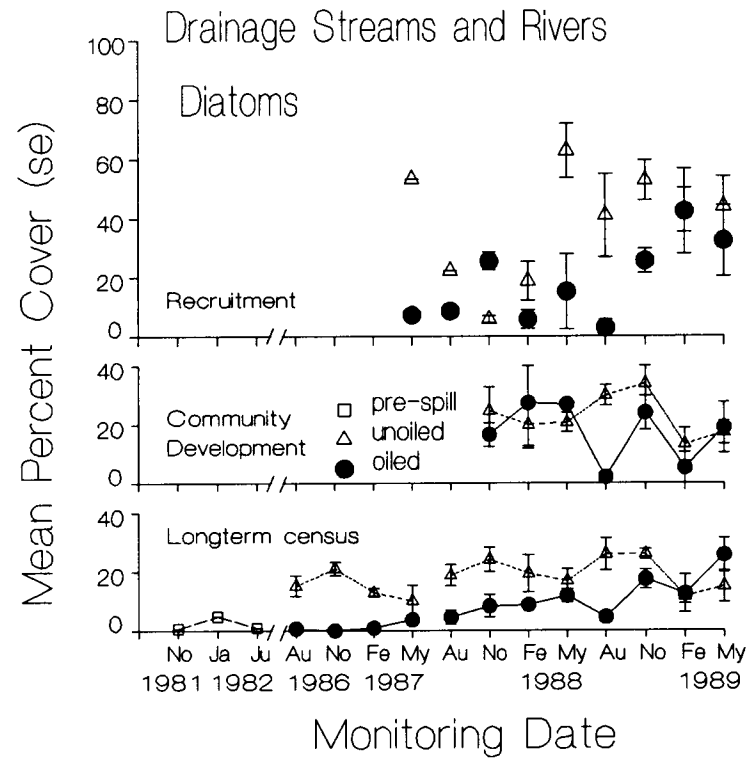


Figure 7.35 Abundance of diatoms on roots or dowels from drainage streams and rivers. Symbols represent among site means with standard error bars. N=1-4 sites/date. See text for details.

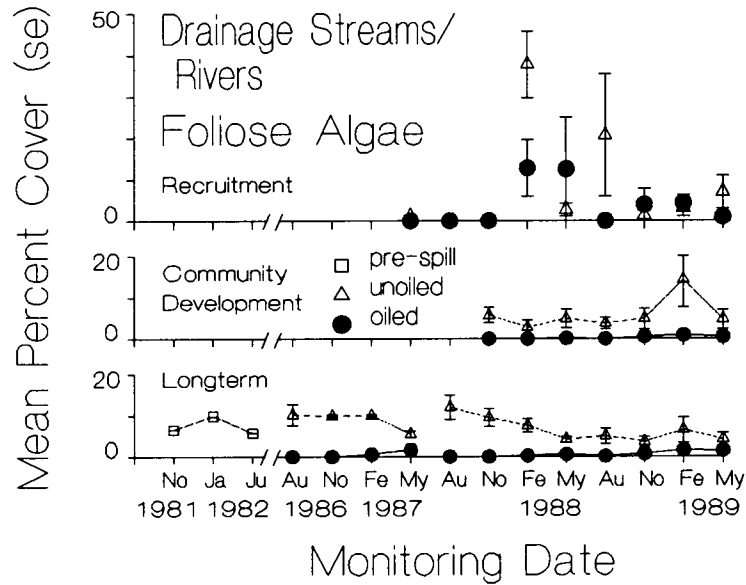


Figure 7.36 Abundance of foliose algae on roots or dowels from drainage streams and rivers. Symbols represent among site means with standard error bars. N=1-4 sites/date. See text for details.

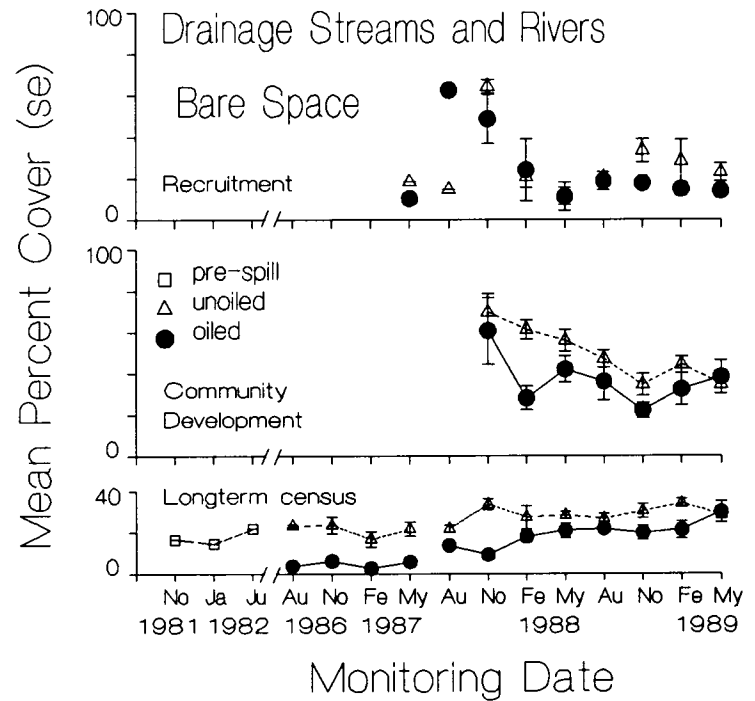


Figure 7.37 Abundance of bare space on roots or dowels from drainage streams and rivers. Symbols represent among site means with standard error bars. N=1-4 sites/date. See text for details.

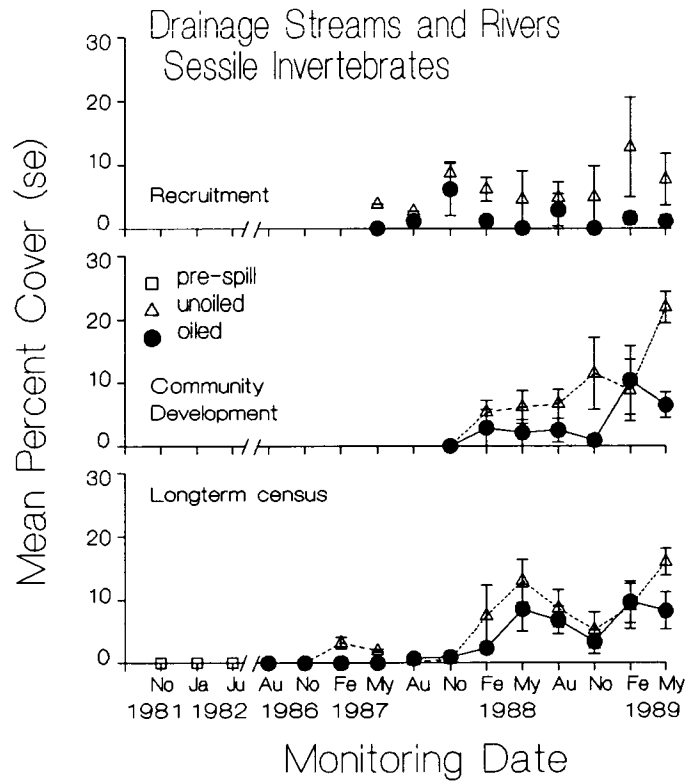


Figure 7.38 Abundance of sessile invertebrates excluding barnacles and bivalves on roots or dowels from drainage streams and rivers. Symbols represent among site means with standard error bars. N=1-4 sites/date. See text for details.

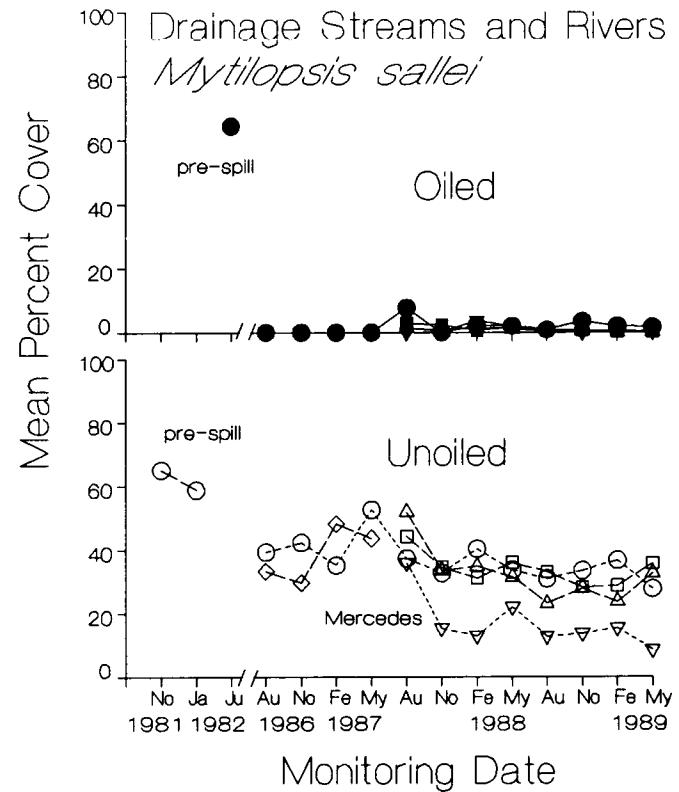


Figure 7.39 Abundance of *Mytilopsis sallei* on roots in the long-term censuses by site from drainage streams and rivers. Symbols represent site means of percent cover, n=16-50 roots each data point. See text for details.

Foliose algae covered ~10% of root surfaces in 1981-1982 (Figure 7.36); mixed algal turfs, *Chaetomorpha* sp. and *Polysiphonia* sp. 1 were recorded. Compared to channels and especially the open coast, fewer species were collected (Tables 7.4, 7.22, 7.23). *Polysiphonia* spp., *Caloglossa letrieurii* and *Bostrychia* spp. were common in drainage streams. Five to six species of foliose algae were expected on a sample of twenty roots using rarefaction estimates (Table 7.24).

Community Development

Community development was still incomplete almost two years after new roots entered the water. Almost 40% of the root surface was bare, although bare space on roots had declined over time (Figure 7.37). *Mytilopsis* increased in abundance through May 1989, but was still less than half as common as on long-term census roots (Figure 7.33). Coverage of *Balanus* was low (Figure 7.34), but its abundance was also low on randomly sampled roots during the same period. Diatoms and foliose algae were close to their abundance on long-term census roots (Figures 7.35, 7.36). Sessile invertebrates increased in abundance over time and were more abundant on community development than long-term census roots in May 1989 (Figure 7.38).

Species of foliose algae on community development roots were a subset of those recorded on long-term census roots (Tables 7.22, 7.23, 7.25). At Hidden River and Río Alejandro, slightly more species of algae were found on community development than long-term census roots (rarefaction estimates, Tables 7.24, 7.26). The opposite was true at the other two streams.

Recruitment

Appearance of different groups on artificial roots was patchy over time. Diatoms were present at each monitoring, and cover was usually >40% (Figure 7.35). Foliose algae were abundant in late 1987 and early 1988, but rare at other times (Figure 7.36). Mixed algal turfs were the most abundant type of foliose algae (Table 7.27). *Balanus* occurred in most monitorings in highly variable abundance (Figure 7.34). Although a few mussels were found at each monitoring, *Mytilopsis* was only common in August 1987, when it also settled heavily in oiled channels (Figures 7.23, 7.33). Sessile invertebrates were present in most monitorings, usually covering ~10% of the dowel surface (Figure 7.38).

Table 7.22 Distribution of algal species or categories in drainage streams, long-term census, second year after the spill. Data for August 1987-May 1988. N=85 roots/site, 340/habitat type, site codes as in Table 7.1. Sum = total occurrences/category in oiled or unoiled streams. ".": taxon did not occur.

Category	Oiled					Unoiled				
	LRRS	PAYR	PMRE	PMRW	Sum	ALER	HIDR	MERR	UNR	Sum
<i>Amphiroa ?fragilissima</i>	3	6	3	.	12
<i>Bostrychia tenella</i>	4	3	.	2	9
<i>Bostrychia</i> sp.2	2	2	1	3	8
<i>Caloglossa letrieurii</i>	.	.	.	2	2	13	15	13	15	56
<i>Caulerpa fastigata</i>	1	.	.	.	1	1	.	.	.	1
<i>Caulerpa verticillata</i>	.	1	.	.	1
<i>Ceramium</i> sp.	1	.	1
<i>Chaetomorpha</i> sp.	1	.	.	.	1	.	.	1	.	1
<i>Cladophora</i> sp.	2	.	.	.	2
? <i>Griffithsia</i> sp.	1	.	.	.	1
<i>Herposiphonia secunda</i>	2	.	.	.	2
<i>Heterosiphonia</i> sp.	1	.	23	10	34
<i>Polysiphonia</i> sp. 1	.	.	1	1	2	17	15	7	10	49
<i>Polysiphonia</i> sp. 2	1	.	2	1	4	4	5	11	4	24
<i>Polysiphonia</i> sp. 3	6	4	1	5	16
Total taxa:	4	1	2	3	7	11	7	9	7	13
Total occurrences:	5	1	3	4	13	54	50	61	49	214

Table 7.23 Distribution of algal species or categories in drainage streams, long-term census, third year after the spill. Data for August 1988 - May 1989. N=80 roots/site, 320/habitat type, site codes as in Table 7.1. Sum=total occurrences/category at oiled or unoiled sites. ".": taxon did not occur.

Category	Oiled					Unoiled				
	LRRS	PAYR	PMRE	PMRW	Sum	ALER	HIDR	MERR	UNR	Sum
<i>Bostrychia tenella</i>	2	.	.	2
<i>Bostrychia sp.2</i>	3	3	.	5	11
<i>Caloglossa letrieurii</i>	16	11	3	14	44
<i>Caulerpa verticillata</i>	.	1	.	3	4
<i>Chaetomorpha sp.</i>	1	.	1
<i>Cladophora sp.</i>	13	.	.	2	15	1	.	.	.	1
? <i>Gelidium pusillum</i>	3	.	3
<i>Champia parvula</i>	1	.	11	.	12
<i>Halimeda opuntia</i>	.	.	.	1	1
Mixed algal turf	6	.	.	.	6	.	13	.	.	13
<i>Polysiphonia sp. 1</i>	.	.	4	4	8	11	7	.	6	24
<i>Polysiphonia sp. 2</i>	.	1	.	.	1	8	13	1	1	23
<i>Polysiphonia sp. 3</i>	6	4	.	3	13
Total taxa:	2	2	1	4	6	7	7	5	5	11
Total occurrences:	19	2	4	10	35	46	53	19	29	147

Table 7.24 Rarefaction estimates of the number of taxa of foliose algae expected in a sample of 20 roots: drainage streams, long-term census.

	Mean \pm 95% Confidence interval		
	8-86 - 5-87	8-87 - 5-88	8-88 - 5-89
A. Oiled			
Largo Remo River South	0.1 \pm 0.2	1.2 \pm 0.4	2.1 \pm 0.3
Largo Remo River North	1.0 \pm 0.2	-	-
Punta Muerto River East	-	0.7 \pm 0.3	0.7 \pm 0.2
Punta Muerto River West	-	0.9 \pm 0.4	1.7 \pm 0.4
Payardi River	-	0.3 \pm 0.2	0.5 \pm 0.3
B. Unoiled			
Hidden River	5.9 \pm 0.4	5.3 \pm 0.4	5.5 \pm 0.4
Río Coco Solo	5.5 \pm 0.4	-	-
Río Alejandro	-	6.4 \pm 0.6	2.6 \pm 0.4
Quebrada las Mercedes	-	5.4 \pm 0.6	4.8 \pm 0.4
Unnamed River	-	5.4 \pm 0.4	3.4 \pm 0.4

Table 7.25 Distribution of algal species or categories in drainage streams, community development, third year after the spill. Data for August 1988-May 1989. N=40 roots/site. Site codes as in Table 7.1. Sum=total occurrences/category at oiled or unoiled sites. ".": taxon did not occur.

Category	Oiled				Unoiled				Sum
	PAYR	PMRE	PMRW	Sum	ALER	HIDR	MERR	UNR	
<i>Bostrychia tenella</i>	1	.	.	1
<i>Bostrychia</i> sp.2	3	.	.	3
<i>Caloglossa letrieurii</i>	8	4	.	13	25
<i>Caulerpa fastigata</i>	.	1	.	1
<i>Champia parvula</i>	1	.	9	.	10
<i>Herposiphonia secunda</i>	1	.	.	1
Mixed algal turf	7	.	.	7
<i>Polysiphonia</i> sp. 1	.	.	2	2	6	7	.	.	13
<i>Polysiphonia</i> sp. 2	.	.	1	1	5	3	.	2	10
<i>Polysiphonia</i> sp. 3	3	.	.	.	3
Total taxa:	0	1	2	3	5	7	1	2	9
Total occurrences:	0	1	3	4	23	26	9	15	73

Table 7.26 Rarefaction estimates of the number of taxa of foliose algae expected in a sample of 20 roots: drainage streams, community development, 1988-1989.

Mean \pm 95% Confidence interval	
A. Oiled	
Largo Remo River South	no roots marked
Punta Muerto River East	0.5 \pm 0.2
Punta Muerto River West	1.3 \pm 0.3
Payardi River	0 \pm 0
B. Unoiled	
Hidden River	5.7 \pm 0.4
Quebrada las Mercedes	1.0 \pm 0.01
Río Alejandro	4.4 \pm 0.3
Unnamed River	1.8 \pm 0.3

Table 7.27 Distribution of algal taxa in drainage streams, recruitment substudy, third year after the spill. Data for August 1988-May 1989. Site codes as in Table 7.1. Sum=total occurrences/category at oiled or unoiled sites. ".": taxon did not occur.

Category	Oiled					Unoiled				
	LRRS	PAYR	PMRE	PMRW	Sum	ALER	HIDR	MERR	UNR	Sum
<i>Caloglossa letrieurii</i>	2	.	.	2
<i>Caulerpa verticillata</i>	.	.	1	.	1
<i>Cladophora</i> sp.	1	.	.	1
Mixed algal turf	3	2	4	2	11	6	6	2	4	18
<i>Polysiphonia</i> sp. 1	1	.	2	.	3
Total taxa:	2	1	3	1	3	1	3	1	1	3
Total occurrences:	4	2	7	1	15	6	9	2	4	21
Sample size:	17	18	16	18	69	18	16	18	18	70

Drainage Streams and Rivers -- Oiled Sites

Long-term Census

The epibiota of mangrove roots in streams near Isla Payardi was almost completely eliminated after the spill and showed practically no signs of recovery through May 1989. All ordinarily abundant groups were absent or rare at oiled sites in August 1986, and all were absent in November 1986. Instead, oil (Figure 7.4) and a bacterial slime covered virtually all space. The remaining space in both months consisted of dead mussels and barnacles too covered by oil and bacteria to identify. Oil still covered ~28% of the roots in May 1989 (Figure 7.4) and leached from the sediments whenever the surface was disturbed. There were dramatic differences in algal abundance (Figure 7.36) and species occurrence (Tables 7.4, 7.22, 7.23) between oiled and unoiled streams. *Mytilopsis* was still essentially absent in May 1989 (Figure 7.33). A few sessile invertebrates, excluding barnacles and bivalves, were found starting in 1988 (Figure 7.38, Tables 7.28, 7.29), but anemones and tunicates were still completely absent from oiled streams in May 1989.

Table 7.28 Distribution of groups of sessile organisms in drainage streams, long-term census, second year after the spill. Data for August 1987- May 1988. N=85 roots/site, 340/habitat type. Site codes as in Table 7.1. Sum=total occurrences/group in oiled or unoiled streams. " Arb.": arborescent. ".": taxon did not occur.

Category	Oiled					Unoiled				
	LRRS	PAYR	PMRE	PMRW	Sum	ALER	HIDR	MERR	UNR	Sum
Anemones	2	.	.	.	2
Encrusting bryozoans	.	.	1	.	1	6	1	.	3	10
Arb. hydroids, bryozoans	.	28	17	17	62	37	10	37	29	113
Sponges	.	15	8	4	27	8	.	21	22	51
Solitary tunicates	4	1	.	5	10

Table 7.29 Distribution of groups of sessile organisms in drainage streams, long-term census, third year after the spill. Data for August 1988 - May 1989. N=80 roots/site, 320/habitat type. Site codes as in Table 7.1. Sum=total occurrences/group in oiled or unoiled streams. "Arb": arborescent. ".": taxon did not occur.

Category	Oiled					Unoiled				
	LRRS	PAYR	PMRE	PMRW	Sum	ALER	HIDR	MERR	UNR	Sum
Encrusting bryozoans	7	.	.	.	7	2	.	1	3	6
Anemones	1	3	.	1	5
Arb. hydroids, bryozoans	4	46	33	32	115	49	21	53	54	177
Sponges	1	20	23	19	63	5	2	26	18	51
Colonial tunicates	14	14
Solitary tunicates	9	1	.	4	14

Community Development

Roots that entered the water in August 1987 showed major differences between oiled and unoiled streams. Diatoms and oil covered the most space in oiled streams (Figures 7.4, 7.35); almost no bivalves, barnacles or foliose algae were found (Figures 7.33, 7.34, 7.36). Species of foliose algae in oiled streams were a subset of those recorded at unoiled streams (Table 7.25), but far fewer species were expected to occur on a sample of 20 roots (rarefaction estimates, Table 7.26). Sessile invertebrates were present starting in 1988, but only began increasing in abundance in 1989 (Figure 7.38).

Recruitment

Oil covered >40% of dowel surfaces in oiled streams through May 1989 (Figure 7.4). Almost no bivalves or barnacles recruited in oiled streams (Figures 7.33, 7.34), while at least some settlement of *Balanus* and *Mytilopsis* occurred each monitoring in unoiled streams. Overall, sessile invertebrates were rare and always less abundant than in unoiled streams (Figure 7.36). Foliose algae were only common in early 1988 (Figure 7.38, Table 7.27), while diatoms were patchily abundant, especially in 1989 (Figure 7.35). These data further suggest that recovery in oiled streams will be very slow.

Drainage Streams and Rivers -- Summary

These results show that the oil spill has had devastating effects in drainage streams. When data on root condition and percent cover are considered together (Figures 7.11, 7.12, 7.33-7.39), we predict that recovery will be a prolonged process, dependent upon the re-establishment of healthy prop roots and the gradual leaching of oil trapped in the sediments.

7.5 Conclusions

Despite limited data prior to the oil spill, several conclusions can be drawn. First, there is a distinct biological community on submerged *Rhizophora* roots in each of the three habitats examined. Mangrove roots on the open coast have the highest species richness. A moderate amount of bare space is available, and no single species dominates space. Foliose and crustose algae combined are the most abundant taxon. Roots in channels and lagoons are chiefly covered with bivalve molluscs, especially *Crassostrea rhizophorae*, and barnacles. *Mytilopsis sallei* is the most common species in drainage streams and rivers; barnacles, algae and diatoms are also abundant.

Second, oil had immediate effects on many organisms in all three habitats. Within three to nine months after the spill, the most common species or taxa were

less abundant in oiled than unoiled areas (*Crassostrea* in channels, foliose algae on the open coast) or had virtually disappeared (*Mytilopsis* in oiled streams). Foliose algae rot quickly and left little evidence (other than lowered abundances) of direct mortality three months after the spill. However, *Mytilopsis* (which persists dead until the byssal attachment deteriorates) and *Crassostrea* (where the ventral valve remains cemented to the root after death) were found in August 1986 and offer such evidence. In oiled streams, dead *Mytilopsis* were found on 61 of the 100 roots sampled (mean percent cover of 10.7%), while in unoiled streams dead mussels were found on 39 of 100 roots and cover averaged 1.3% (n=100). In oiled channels and lagoons, dead *Crassostrea* averaged 22.1% cover and occurred on 83 of 100 roots sampled, compared to 2.2% cover, present on 58 of 100 roots at unoiled sites.

Many less common species also showed relative decreases in abundances correlated with oiling soon after the event. On the open coast, these included crustose algae, sponges, arborescent hydroids and bryozoans, and other sessile invertebrates. In channels, they included *Balanus*, *Mytilopsis*, and other sessile invertebrates. In oiled streams, they included diatoms, foliose algae, and *Balanus*. A few species or taxa showed no early, post-oiling differences in abundance between oiled and unoiled areas. These included barnacles on the open coast, diatoms in channels, and sessile invertebrates (other than barnacles and bivalves) in streams. Of these, barnacles and sessile invertebrates were very rare (<5% cover), while diatoms averaged $\leq 20\%$ cover. One taxon, diatoms, bloomed ephemerally at oiled, open coast sites from three to six months after the spill.

Third, oil persisted in mangroves through May 1989. Initial oiling was heavy - nearly 100% of all roots sampled in each habitat had a measureable amount of oil on them in the first year after the spill. The proportion of sampled roots that had oil on them remained near 100% in streams and slowly decreased to about 70% in channels and on the open coast through May 1989. The percent cover of oil on randomly-sampled roots showed a similar pattern over time. Oil, while slowly becoming less abundant in all habitats, continued to cover more space in streams than in channels or on the open coast. Decreases in oil coverage resulted from weathering, microbial degradation, loss of bark (and thus of oil on bark), loss of organisms coated with oil and overgrowth of oil by epibiota.

There has been considerable reoiling of mangrove roots. Data from both community development roots (which entered the water 15-18 months after the original spill) and recruitment dowels (which were set out fresh and collected after three months) suggest that roots in oiled streams were nearly constantly reoiled, while those in channels and on the open coast experienced episodic reoiling, particularly during rainy seasons. In streams, all community development roots were oiled as soon as they entered the water. The fraction of such oiled roots first increased over time in channels and on the open coast, then fell beginning in late 1988. The percent cover of oil on community development roots remained $\leq 10\%$ in channels and on the open coast, but was as high as 55% in streams.

Results were similar when recruitment dowels are considered. Between 80-100% of all dowels from oiled streams had a trace or more of oil through May

1989; percent cover of oil ranged from 20-80%. The proportion of oiled dowels in channels varied from ~10-100%, first decreasing and then increasing after February 1988. Percent cover was <15% except in the two 1989 monitorings, when it exceeded 30%. On the open coast, fewer than 20% of dowels were oiled, except in May 1987, August 1988 and May 1989. The percent cover of oil was always <20%.

Reoiling appeared chiefly due to the emergence of oil from sediments, especially following heavy rains (personal observations). One control site was lightly oiled between May and August 1988 (Hidden Channel); preliminary data show this oil to be identical to that originally spilled (J. MacPherson, personal communication). This supports our observations on the source of reoiling. Analysis of sediment cores and of quarterly collections of sentinel organisms should provide more information on sources of reoiling, as well as the degree of degradation of oil in sediments over time.

Fourth, measurable differences remained in the epibiota of oiled and unoled sites in May 1989. Long-term effects of oiling, and recovery from those effects, varied both among habitats and among taxa. Changes in abundances of organisms from each habitat during the first three years after the spill illustrate some of this variability and suggest possible causes.

On the open coast, foliose and crustose algae on randomly censused roots showed higher percent cover, occurred on a greater number of roots, and had a higher species richness at unoled than oiled sites through May 1989. All sessile invertebrates except barnacles remained rare (or absent) on roots at oiled relative to unoled areas, and the increase in barnacle cover at oiled sites was correlated with the disappearance of a grazing snail. Recruitment onto dowels placed on the open coast was highly variable over time, but similar between oiled and unoled areas. Recruitment data showed that competent sporelings of foliose algae existed in both oiled and unoled areas, and that they could settle and grow rapidly. No other organisms (except diatoms) recruited to a measureable level in either oiled or unoled open areas. Foliose algal abundance on community development roots quickly reached levels similar to those in long-term censuses within treatments (oiled, unoled). However, percent cover of foliose algae at oiled sites on community development roots did not reach levels found on similarly-aged roots at unoled sites. This suggests post-settlement mortality or reduced growth or both of algae at oiled sites, three years after the spill.

Any discussion of results from channels and lagoons is complicated by several factors, including variation in the amount of original oiling among sites, differences in physical characteristics among sites (particularly the two control sites in Margarita Lagoon), and the secondary oiling of one control site (Hidden Channel). *Crassostrea*, the dominant species in 1981-2, remained more abundant at unoled than oiled sites through November 1987; abundances then converged. A slow increase in oyster abundance at oiled sites was partly responsible. At three of five oiled sites, *Crassostrea* abundance increased steadily and fairly rapidly, at a fourth it remained

low, and at a fifth it increased slowly, through May 1989. However, a decrease in *Crassostrea* abundance at unoiled sites throughout the same period also contributed to the convergence between oiled and unoiled sites. It is at present impossible to say whether the decline of this oyster at unoiled sites is part of a longer-term, natural cycle, or whether it will continue to decline. The addition of two control sites in Margarita Lagoon (in August 1987) further influenced an overall decrease in oyster abundance at unoiled areas. These sites showed the sharpest declines in *Crassostrea* cover of all unoiled sites and had little or no oyster cover on recruitment dowels or community development roots through May 1989. More physical and biological data are needed before conclusions can be reached regarding recovery of this species from oiling.

In contrast, the barnacle *Balanus improvisus* remained more abundant on randomly surveyed roots in unoiled channels than in oiled ones throughout the study. Recruitment was greater at unoiled sites only during 1987 and at oiled sites in 1988-9. However, slow increases in barnacle abundance over time on community development roots at unoiled sites were not equalled at oiled sites, where *Balanus* remained rare. The false mussel *Mytilopsis* recruited only once (August 1987, oiled sites) and remained rare to absent on community development roots at both oiled and unoiled sites. On long-term census roots, this bivalve tended to be slightly more abundant at unoiled sites through 1987 and early 1988, but did not differ between oiled and unoiled sites in the last year. This convergence in abundance between oiled and unoiled areas was due to decreases in mussel abundance at unoiled sites rather than increases at oiled sites. As with *Crassostrea*, conclusions regarding recovery await more data on relative rates and amounts of future change in *Mytilopsis* cover.

Other less abundant taxa, including diatoms, foliose algae and rare sessile invertebrates showed little or no differences in percent cover between oiled and unoiled channels over the last two years of the study. Differences in species occurrence did persist, however, for four of the six types of rare sessile invertebrates.

In summary, there has been partial recovery in channels and lagoons. Only *Balanus* still showed clear reductions in abundance on long-term, oiled roots relative to those on unoiled roots through May 1989. Results for the normally dominant *Crassostrea*, and especially for *Mytilopsis*, were equivocal (with convergence due partly or wholly to decreases in abundance in unoiled areas rather than increases in oiled ones). Some rare groups or species differed little or not at all between oiled and unoiled areas during the last two years.

Oiled drainage streams remained severely affected through May 1989. The normally dominant species, *Mytilopsis*, was not found on long-term census roots at oiled sites. Despite several minor episodes of recruitment (in August 1987 and November 1988), the false mussel did not increase in abundance on oiled community development roots. This contrasted sharply with unoiled sites, where *Mytilopsis*

remained the most abundant organism, showed minor recruitment in eight of nine quarterly intervals since 1987, and increased slowly and steadily on community development roots. The abundance of *Balanus* on long-term census roots in oiled areas converged with those at unoiled areas by mid-1987 because of decreases in abundance at unoiled sites. Its recruitment continued to differ between oiled and unoiled streams through May 1989, with little or none recorded at oiled sites and high variability at unoiled sites. Foliose algae showed a slow decline in abundance on unoiled, long-term census roots and continued to be absent to very rare on oiled roots through May 1989. Recruitment was variable and patchy everywhere, with no difference between oiled and unoiled streams by the third year after the spill. Post-settlement survival did differ, though, since only in unoiled streams did any algal cover develop on community development roots. Further, both the number of algal species found and the relative occurrence of algae on roots remained much higher at unoiled sites than in oiled streams through the entire period.

Diatoms in oiled streams increased in abundance from 1987 through May 1989 to roughly the same levels as those at unoiled sites; recruitment was variable, but greater at unoiled sites in six of nine instances. Diatom cover on community development roots was similar at oiled and unoiled sites. Rare sessile invertebrates showed similar abundances on long-term census roots in oiled and unoiled streams, but generally had higher percent coverage both on unoiled dowels and unoiled community development roots than on oiled ones, through May 1989.

All three habitats showed differences in mangrove root epibiota between oiled and unoiled areas over the three years of this study. Foliose and crustose algae, sponges, rare invertebrate groups, and hydroids and bryozoans (from the open coast), *Balanus* (in channels and lagoons), and *Mytilopsis* and diatoms (in streams) all were more abundant on long-term census roots in unoiled sites than in oiled ones through May 1989. Abundances of several species, including *Mytilopsis* (in channels and lagoons) and foliose algae and *Balanus* (in streams) converged in abundance between oiled and unoiled sites over time, primarily because of decreases in average cover at unoiled sites. Other groups, including diatoms (on the open coast), foliose algae and *Mytilopsis* (in channels and lagoons) also converged at oiled and unoiled sites during the period of the study, but from a combined increase at oiled sites and decrease at unoiled sites.

A fifth and final conclusion is that roots themselves differed in several ways between oiled and unoiled areas. In the second and third years after the spill, the length of healthy, long-term census roots was greater in unoiled streams, channels and on the open coast than in the corresponding oiled habitats. For roots that entered the water after the spill (community development), root length was greater on unoiled open coast and in unoiled streams, but longer in oiled than unoiled channels.

The physical condition of roots in oiled and unoiled areas also differed. Prop roots were naturally damaged from logs washing ashore or downstream, from treefalls, and from attack by herbivorous isopods and crabs (unpublished data). At any given time, of roots sampled in the long-term census ~3-9% at unoiled open

coast sites, 0-3% in channels and 1-10% in streams were damaged and appeared dead. Root mortality at oiled sites during the same period was higher (24-43% at open coast sites, 18-46% in oiled channels and lagoons, and 43-64% in streams). Community development roots were neither exposed to initial, heavy oiling nor to residual oil for as long. As expected, mortality was lower than in long-term census roots. However, results were similar. Oiled areas showed greater mortality of community development roots than unoiled ones in all three habitats.

The oil spill at Isla Payardi, Panama, is a continuing opportunity to gain insight into the effects and potential management of oil spills in an ecosystem not only important on a worldwide scale, but also a major component of the Southeastern coast of the United States. Three years after the original spill, full recovery has not occurred in any of the three habitats examined. Roots in streams have suffered the most serious effects and have experienced the poorest recovery. Their normally dominant species, *Mytilopsis*, disappeared and has remained absent; most other, relatively rare species have at best partially recovered. Reoiling has been nearly constant from oil trapped in bank sediments; reduced recruitment relative to other oiled habitats suggests oil levels may have continued to be at lethal levels for larvae. The epibiota of *Rhizophora* roots on open, wave exposed shores appeared superficially less affected by May 1989. However, most species, including dominant groups, still showed major differences between oiled and unoiled areas, despite (1) observed high rates of turnover for many open-coast species and (2) nearly constant washing by waves during dry season. Although results to date in channels have been the most equivocal, some recovery has clearly occurred. The oyster *Crassostrea*, dominant before the spill, has increased in abundance over time at four of five oiled sites. Less common species have shown mixed results through May 1989. However, the high degree of variation among sites, and especially the puzzling decreases in several species at unoiled sites, indicate more data are needed before final conclusions are drawn regarding effects and recovery in this habitat.

Both our data and observations of the poor state of trees and roots, particularly in channels and drainage streams, suggest recovery of the mangrove root epibiota may not occur for several more years. At sites where oiling was heaviest, recovery of root epibiota is intimately tied to the recruitment and survival of mangrove trees (e.g., on substratum availability for the epibiota), as well as on factors examined in this study. Additional long-term monitoring will provide data needed to assess more fully the effects of the April 1986 oil spill on this fragile ecosystem.

7.6 Acknowledgments

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Chapter 8 Subtidal Seagrass Communities

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8.1 Introduction

Seagrass meadows along the central Caribbean coast of Panama are usually located between small fringing reefs and extensive mangrove forests. The largest of these grassbeds are near the Atlantic entrance of the Panama Canal and the refinery at Bahía Las Minas. A large oil spill, which oiled several of these lagoonal grassbeds, occurred at Bahía Las Minas during April 1986 (Chapter 1).

Beds of seagrasses and their associated animal communities are very susceptible to many types of man-induced (Jacobs 1980; Phillips 1980; Williams 1988; Thayer *et al.* 1975; Zieman *et al.* 1986) and natural (Stauffer 1937; Thomas *et al.* 1961) environmental perturbations. The response of seagrass communities to oiling, although not well described (Zieman *et al.* 1986; Marshall *et al.*, 1990), appears to be highly dependent on the position of grassbeds in the subtidal and intertidal zones and to local climatic and oceanographic conditions at the time of oil exposure. Many other factors also undoubtedly play a role in the response to oiling and any subsequent recovery processes. Sediment texture, degree of exposure to waves and currents, tidal flushing, seasonal timing of recruitment, suspended sediment loads, initial seagrass density, etc. may all have a role in determining the nature of the recovery process. The grassbeds on the Caribbean side of Panama are located along a very complex coastline that is broken up by embayments at river mouths; there also are numerous small islands within the bays. This complexity has resulted in the development of seagrass meadows under various combinations of environmental conditions. The April 1986 spill at Bahía Las Minas, Panama (Cubit *et al.* 1987), oiled nearshore environments, including many hectares of seagrass meadows, from the Atlantic entrance of the Panama Canal to María Chiquita (Figure 8.1).

Some seagrass meadows elsewhere in the tropics and in the subtropical and temperate zones have been contaminated by oil from tanker wrecks, the release of oil from tankers, and an oil rig blowout (Zieman *et al.* 1986). Despite these cases, omnipresent threats to seagrass meadows, and the current world-wide decrease in seagrass acreage, little is known regarding the ability of seagrass meadows to recover after oiling (Zieman *et al.* 1986; Thorhaug and Marcus 1987), and nothing is known concerning the resistance of the resident animal assemblages to oil effects. The

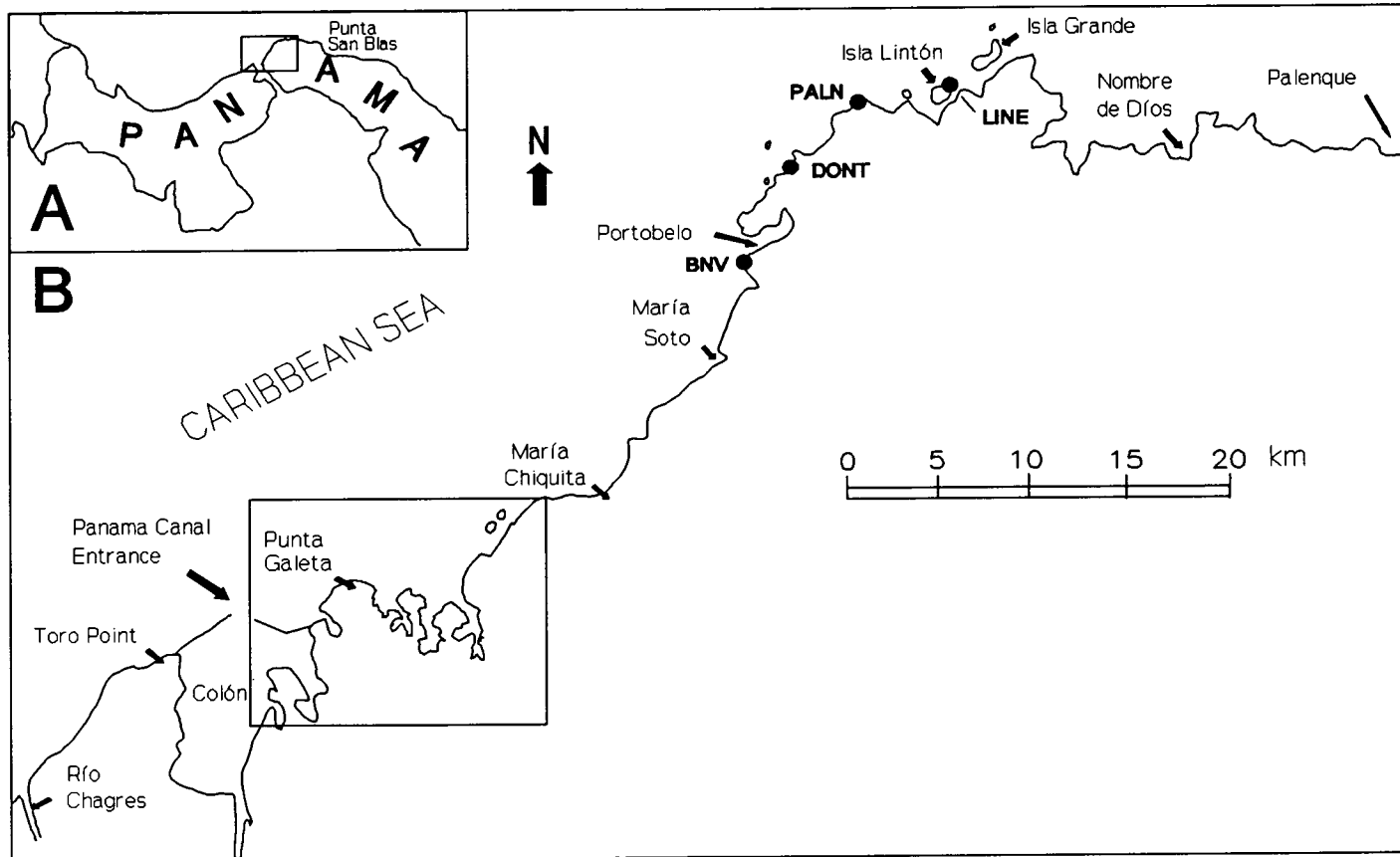


Figure 8.1 Map of the study sites included in this report. Site codes are: LRS = Largo Remo South, LREN = Largo Remo Entrance, PGN = Peña Guapa Norte, MINN = Mina Norte, BNV = Buenaventura, DONT = Doncella *Thalassia*, PALN = Palina Norte, and LINE = Lintón East. Filled circles = oiled sites, open circles = unoiled sites. See Figure 1.1 for further details.

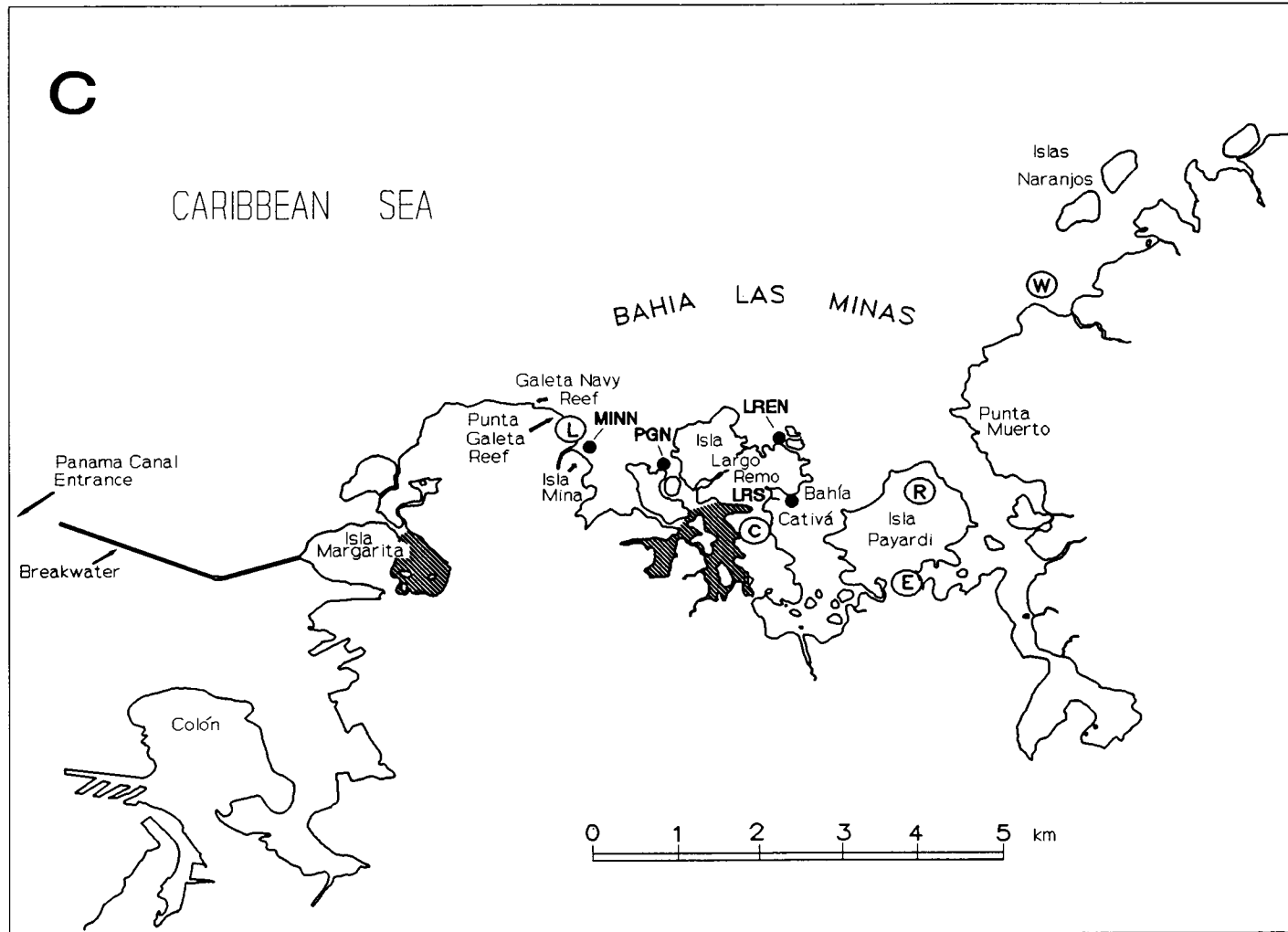


Figure 8.1 Map of the study sites included in this report (continued).

Bahía Las Minas refinery spill in Panama presented an opportunity to observe both the amount of damage done by various levels of oiling to numerous seagrass meadow communities and their resilience to this type of damage under a wide range of environmental conditions.

The ecological roles of seagrasses have been documented by numerous authors (e.g., Kikuchi and Peres 1977; Zieman 1982). Seagrass beds are important as nursery grounds for many species of fish and large crustaceans; they stabilize sediments, act as sediment traps, provide substrata for many species of epiphytes and epifauna, and provide food as both green leaves and detritus for many small invertebrates and juvenile fish species. Thus, damage to or the loss of seagrass meadows may have ecological effects that extend well beyond the boundaries of those meadows.

This report describes the preliminary results of a three-year oil spill recovery study in Panamanian seagrass meadows. To date, animals found in benthic core samples collected during September 1986, November 1986, January 1987, and January 1988 have been sorted to major taxonomic categories. The polychaetes of the September 1986 core samples have been sorted to species where possible. Seagrasses from these samples were dried and weighed. Pushnet samples have also been collected quarterly at each grassbed site from November 1986 until April 1988. Pushnet samples from November 1986, January 1987, April 1987, and July 1987 have been sorted to major taxa. Caridean shrimp, typically the most abundant group in the pushnet samples, have also been identified to species. Complete listings of all species from both pushnet and core samples will be compiled to describe and quantify effects of the oil spill on seagrass meadow infaunal, epifaunal, and floral communities. This study should provide much-needed information about the effect of oil on seagrass biomass and the abundance of the associated epifauna and infauna.

8.2 Methods

8.2.1 The Study Sites

Seagrass meadows from Isla Margarita, near the Atlantic entrance of the Panama Canal, to Isla Lintón (near Isla Grande) were chosen as study sites (Figure 8.1). All grassbeds were adjacent to mangrove shorelines and were subtidal, but shallow enough to be sampled without SCUBA. Depth profiles were produced for each site based on three transects extending from the shore to the seaward edge of each grassbed. Depth was measured at 1 m intervals along each transect. All sites were located behind small patch or barrier reefs in shallow lagoons. Deeper grassbeds do not exist along this section of the Panamanian coast. Physical parameters measured at each site are summarized in Table 8.1.

Table 8.1 Environmental characteristics of oiled and unoiled grassbed sites. Means and standard deviations, (in parentheses), are given for all characteristics except "oil seen." Dates data were collected: September 1986 - April 1987 for observations of oil, October 1987 for sediment characteristics, April 1987 - July 1988 for salinity, and July 1988 for depth.

Sites	Oil Seen	Sediment Characteristics		Salinity (o/oo)	Depth(cm)
		%Silt-Clay	%Organic		
Oiled					
LRS	4/4x	3.72 (1.21)	3.72 (0.19)	30.1 (5.9)	53.6 (14.3)
LREN	4/4x	7.82 (1.56)	4.61 (0.22)	33.0 (3.3)	64.1 (17.3)
PGN	4/4x	5.03 (1.72)	4.14 (0.74)	33.4 (3.6)	49.3 (16.0)
MINN	4/4x	25.44 (1.79)	13.80 (3.15)	31.6 (5.1)	45.0 (7.4)
Unoiled					
BNV	0/4x	17.43 (5.06)	8.89 (3.23)	32.9 (4.1)	50.0 (6.3)
DONT	0/4x	11.34 (6.79)	5.04 (1.61)	35.0 (2.0)	11.3 (5.0)
PALN	0/4x	6.45 (3.96)	4.89 (1.24)	34.0 (4.1)	25.0 (3.9)
LINE	0/4x	4.33 (2.07)	4.80 (2.18)	35.0 (2.5)	45.0 (5.0)

8.2.2 Infaunal Sampling

Samples of seagrasses, algae, and benthic fauna were collected with a PVC coring tube (10.16 cm inside diameter). Each sample consisted of three pooled cores totaling 0.024 m². Eight samples were taken at each of eight grassbed sites (samples were collected at 12 sites in September 1986 and at 16 sites in November 1986, but data from only eight of those sites are included in this report). Initially, sample locations within each grassbed were haphazardly selected except that patches of bare sand and of pure *Syringodium filiforme* were avoided. After the first two collection dates, September 1986 and November 1986, a grid was used to locate replicate core positions in each grassbed. Permanent corner markers were placed at each site and a set of grid block positions, based on measurements made from the corner posts, was randomly selected each sampling date.

Five surface-scrapes for sediment grain-size analyses were also taken at each station. Five additional sediment scrape samples were collected at each site in hexane rinsed, hydrocarbon-free glass containers. These samples are stored frozen in Panama.

Upon collection, core samples were sieved through 500 μm mesh screen and preserved in buffered 10% formalin with rose bengal. In the laboratory, samples were gently washed with tap water to remove formalin. Seagrass blades and roots were separated from the samples and dried at 90°C for five days. Prior to drying, blade and root masses were carefully examined for hidden animals and attached epiphytic macroalgae were removed. After drying, blades and roots were weighed separately. Macroalgae in the samples were also dried and weighed. Only the seagrass data were analyzed for this report.

Animals were sorted to the taxa indicated in Table 8.2 for the first collection of core samples; both epifauna and infauna were processed (September 1986). For this report, abundance patterns of a few selected taxa, those that are usually abundant in tropical and temperate grassbeds, were graphed. Warwick (1988) demonstrated that little information is lost in benthic studies when animals are enumerated to major taxa only. Accurate identification to species requires the expertise of taxonomic specialists. This is particularly true in the tropics where many benthic invertebrates are undescribed. The taxonomic levels selected for analyses in this report reflect 1) the relative importance of various groups in tropical seagrass beds, 2) the principal investigator's taxonomic specialties (polychaetes and caridean shrimp), and 3) taxa that can easily be identified. Further species identifications are underway by taxonomic specialists at the Florida Museum of Natural History (fish) and the U.S. National Museum of Natural History (polychaetes and isopods). Polychaetes from November 1986, January 1987, and January 1988 are being sorted to families.

Sediment samples collected for textural analyses were processed by wet sieving through a standard (Wentworth) series of brass mesh sieves. Organic content was determined by combustion. Data from the most completely analyzed set of sediment samples are summarized in Table 8.1. Due to time constraints, the remaining sediment samples will not be processed unless a major change in seagrass biomass or other possible sediment-affecting changes are detected. Sediments collected for hydrocarbon analyses were frozen. No seagrass sediments other than those collected during the first months of the project have been analyzed for hydrocarbon content (Chapter 9).

Table 8.2 List of taxa from core collections made during September 1986.
Additional species will be added as they are identified.

Phylum Mollusca	Family Poecilochaetidae
Class Gastropoda	<i>Poecilochaetus johnsoni</i>
<i>Acmaea pustulata</i>	Family Cirratulidae
<i>Cerithium eburneum</i>	<i>Cirriformia punctata</i>
<i>Cerithium variabile</i>	<i>Cirriformia luxuriosa</i>
<i>Diodora</i> sp. A	<i>Cirratulus cirrata</i>
<i>Fasciolaria tulipa</i>	<i>Cirratulus</i> sp. A
<i>Polinices lacteus</i>	<i>Cirratulus</i> sp. B
<i>Polinices hepaticus</i>	<i>Cirratulus</i> sp. C
<i>Smaragadia viridis</i>	<i>Dodecaceria concharum</i>
Nudibranch species	
Class Bivalvia	Order Capitellida
<i>Anadara notialis</i>	Family Capitellidae
<i>Arcopsis adamsi</i>	Capitellid species
<i>Chione cancellata</i>	Family Maldanidae
<i>Codakia orbicularis</i>	<i>Chymenella</i> sp.
<i>Corbula</i> sp. A	Order Opheliida
<i>Diplodonta punctata</i>	Family Opheliidae
<i>Tagelus divisus</i>	<i>Armandia bioculata</i>
<i>Tellina alternata</i>	<i>Trachytyrpane</i> sp. A
Phylum Sipuncula	Order Phyllodocida
Sipunculid species	Family Phyllodocidae
	<i>Eualia myriacyclum</i>
Phylum Annelida	Phyllodocid sp. A.
Class Polychaeta	Family Polynoidae
Order Orbiniida	Polynoid species
Family Orbiniidae	Family Sigalionidae
Orbiniid sp. A	<i>Psammolyce spinosa</i>
Orbiniid sp. B	Family Syllidae
<i>Scoloplos</i> sp. A	Syllid species
<i>Scoloplos</i> sp. B	Family Nereidae
<i>Scoloplos rubra</i>	<i>Ceratonereis mirabilis</i>
Family Paraonidae	<i>Nereis riisei</i>
<i>Aricidea suecia</i>	<i>Platynereis dumerili</i>
Order Spionida	Family Glyceridae
Family Spionidae	<i>Glycera robusta</i>
<i>Spio</i> sp. A	Family Goniadidae
<i>Prionospio cirrifera</i>	<i>Goniada aciculata</i>
<i>Prionospio heterobranchia texana</i>	<i>Goniada</i> sp. A

Table 8.2 List of taxa from core collections made during September 1986 (continued).

Order Amphinomida	Order Sabellidae
Family Amphinomidae	Family Sabellidae
<i>Chloeia viridis</i>	<i>Podochela riisei</i>
<i>Eurythoe complanata</i>	<i>Hypsicomus phaeotania</i>
Order Eunicida	<i>Sabella melanostigma</i>
Family Eunicidae	Sabellid sp. A
<i>Eunice websteri</i>	Sabellid sp. B
<i>Eunice</i> sp. A	<i>Chone</i> sp. A
<i>Lysidice ninetta</i>	<i>Megalomma bioculatum</i>
<i>Marphysa sanguinea</i>	<i>Megalomma vesiculosum</i>
<i>Nematonereis unicornis</i>	
Family Onuphidae	Phylum Arthropoda
<i>Kinbergonuphis</i> sp.	Class Ostracoda
Family Lumbrinereidae	Ostracod species
<i>Lumbrinereis treadwelli</i>	Class Malacostraca
Family Arabellidae	Order Stomatopoda
<i>Arabella mutans</i>	Stomatopod species
Arabellid sp. A	Order Decapoda
<i>Drilonereis nuda</i>	Family Sicyonidae
Family Dorvilleidae	<i>Sicyonia laevigata</i>
<i>Dorvillea rubrovittatus</i>	Family Palaemonidae
<i>Schistomeringos longicornis</i>	<i>Periclimenes americanus</i>
Order Oweniida	Family Alpheidae
Family Oweniidae	Alpheid species
<i>Owenia collaris</i>	Family Hippolytidae
Order Flabelligerida	<i>Hippolyte zostericola</i>
Family Flabelligeriidae	<i>Latreutes fucorum</i>
<i>Pherusa inflata</i>	<i>Thor floridanus</i>
Order Terebellida	<i>Trachycaris restrictus</i>
Family Pectinariidae	Family Processidae
<i>Pectinaria gouldii</i>	<i>Ambidexter symmetricus</i>
Family Ampharetidae	<i>Processa fimbriata</i>
Ampharetid sp. A	Family Callianassidae
<i>Isolda bipinatta</i>	Callianassid sp. A
Family Terebellidae	Family Upogebidae
<i>Eupolyμία nebulosa</i>	<i>Upogebia affinis</i>
<i>Pista fasciata</i>	Family Diogeniidae
<i>Pista</i> sp. A	<i>Clibinarius antillarum</i>
Terebellid species	Family Calappidae
	<i>Hepatus pudibundus</i>

Table 8.2 List of taxa from core collections made during September 1986
(continued).

Family Majidae
<i>Pitho laevigata</i>
<i>Pitho aculeata</i>
<i>Pitho</i> sp. A
Family Xanthidae
Xanthid species
Family Pinnotheridae
<i>Pinnixia</i> sp. A
Order Mysidacea
Mysid species
Order Cumacea
Cumacean species
Order Tanaida
Tanaid species
Order Isopoda
Isopod species
Order Amphipoda
Amphipod species
Phylum Echinodermata
Class Echinoidea
<i>Echinometra lucunter</i>
<i>Lytechinus variegatus</i>
Class Holothuroidea
Holothuroid sp. A
Holothuroid sp. B
Class Ophiuroidea
Ophiuroid species

8.2.3 Epifaunal Sampling

Mobile epifauna were collected, starting in November 1986, by pushnet sampling along 10 m transects. Initially, these positions were also haphazardly located but were later positioned randomly within the grid system (see Chapter 8.2.2). The pushnet frame was 0.75 m wide and 0.35 m high, with a mesh of 1.0 mm Nytex screen. Five replicate samples were collected at each site on each collection date. Animals collected by pushnet were treated with the same preservatives and dye used for core samples. Table 8.3 lists the taxa identified and enumerated from the first four pushnet collections. Additional epifaunal collections are being processed.

8.2.4 Size-frequency Measurements

Size-frequency data and other statistics on various life-history attributes are also being collected on selected species from the oiled and unoiled sites. To date, measurements of carapace length have been completed on the two most abundant species of caridean shrimps collected by pushnet in November 1986. Animals from later collections are being measured. These data will be analyzed to detect possible sublethal effects of oiling on size, sex ratios, and reproductive response.

8.2.5 Data Analyses

Data on abundant epifaunal and infaunal taxa were graphed for each site and collection date. Epifaunal data, which are more complete, were also used in similarity analyses using a frequency transformation and Czekanowski's Quantitative Index. The dendrogram was produced with a group-average sorting strategy. As described by Bloom (1981), there are many alternative data transformations and similarity indices that can result in differing patterns when applied to the same data set. Czekanowski's Quantitative Index is thought to be the most conservative similarity index (Bloom 1981) and, for that reason, was selected for use here. The similarity analysis and the dendrogram were produced with a software package (CAS by Stephen A. Bloom) developed for community data. Further analyses (as developed by Bloom 1980) of the patterns seen in this dendrogram will be undertaken later.

Table 8.3 Epifaunal taxa collected from November 1986 through October 1987. Additional species will be added as they are identified.

Phylum Platyhelminthes	Family Processidae
Platyhelminth species	<i>Processa fimbriata</i>
	<i>Ambidexter symmetricus</i>
Phylum Mollusca	Family Diogeniidae
Class Gastropoda	<i>Clibanarius antillarum</i>
Gastropod species	Family Paguriidae
Class Polyplacophora	Pagurid species
Polyplacophoran species	Family Parthenopidae
	Parthenopid species
Phylum Arthropoda	Family Xanthidae
Subphylum Chelicerata	Xanthid species
Class Pycnogonida	Family Pinnotheridae
Pycnogonid species	<i>Pinnixia</i> sp. A.
Subphylum Crustacea	Family Majidae
Class Ostracoda	Majid species
Ostracod species	Family Portunidae
Class Malacostraca	Portunid species
Order Stomatopoda	Order Mysidacea
Stomatopod species	Mysid species
Order Decapoda	Order Cumacea
Family Penaeidae	Cumacean species
<i>Penaeus duorarum</i>	Order Tanaidae
Family Sicyonidae	Tanaid species
<i>Sicyonia laevigata</i>	Order Amphipoda
Family Gnathophyllidae	Amphipod species
<i>Gnathophyllum americanum</i>	
Family Palaemonidae	Phylum Echinodermata
<i>Leander tenuicornis</i>	Class Echinoidea
<i>Palaemon northropi</i>	<i>Echinometra lucunter</i>
<i>Periclimenes longicaudatus</i>	<i>Lytechinus variegatus</i>
<i>Periclimenes americanus</i>	Class Holothuroidea
<i>Typton tortugae</i>	Holothuroid sp. A
Family Alpheidae	Class Ophiuroidea
Alpheid species	Ophiuroid species
Family Hippolytidae	
<i>Hippolyte zostericola</i>	Phylum Chordata
<i>Latreutes fucorum</i>	Fish species
<i>Latreutes parvulus</i>	
<i>Tozeuma carolinense</i>	
<i>Trachycaris restrictus</i>	
<i>Thor manningi</i>	

8.3 Results

8.3.1 Epifauna

Caridean shrimp species, by far the most abundant major taxon in the epifaunal study, showed a varied response to oiling. *Hippolyte zostericola* (Figure 8.2) was generally more abundant at the oiled sites than at the unoiled sites. In contrast, *Latreutes fucorum* (Figure 8.3) was always uncommon at the oiled sites, as was *Periclimenes americanus* (Figure 8.4). The commercially important penaeid shrimp *Penaeus duorarum*, like *Hippolyte*, was generally more abundant at the oiled sites (Figure 8.5).

Small fish collected by pushnet (Figure 8.6) were much more abundant in the unoiled areas than in the oiled areas. Most of the fish collected were small parrotfish (Scaridae), gobies (Gobiidae), and blennies (Blenniidae). Other larger, more mobile fishes generally escaped the slow-moving pushnet. Presumably, the fishes that were caught by pushnet hauls were those that cannot easily emigrate from oiled to unoiled seagrass meadows and that would not survive emigration through deeper offshore areas.

Majid crabs (Figure 8.7) showed no discernible differences in abundance between oiled and unoiled sites. Amphipods (Figure 8.8) and mysid shrimp (Figure 8.9) both showed increases in abundance after the second sampling date at Largo Remo Entrance (LREN). Fluctuations in abundances of these two small crustaceans can be seen at the other three oiled sites. Virtually no amphipods were found at Peña Guapa North (PGN), a heavily oiled site.

A greater abundance of hermit crabs (Figure 8.10) was apparent at Mina North (MINN) and PGN (both oiled sites) until July 1987 when hermit crab abundances also increased at the unoiled sites. Hermit crabs are being identified to species, but these data are not complete enough to be included in this report.

Echinoderms, including Holothuroidea, Stellerioidea, and Echinoidea, were not abundant at any oiled or unoiled site until a sudden increase in abundance of Ophiuroidea occurred at Doncella (DONT) during July 1987 (Figure 8.11).

8.3.2 Infauna

Tanaids (Figure 8.12) were completely absent from the oiled stations from September 1986 through January 1988. The patterns of amphipod abundance from core samples (not shown) followed those seen in the epifaunal collections.

Bivalves (Figure 8.13) were virtually absent from both the oiled and unoiled beds when the first post-spill samples were taken, but increased similarly thereafter. Infaunal Ophiuroidea (Figure 8.14) were absent at all oiled sites on the first post-spill date and were still almost nonexistent in January 1988, more than 1.5 years after the spill. They were also not abundant at any unoiled site in the September 1986 samples, but their abundances later increased at some of the unoiled sites.

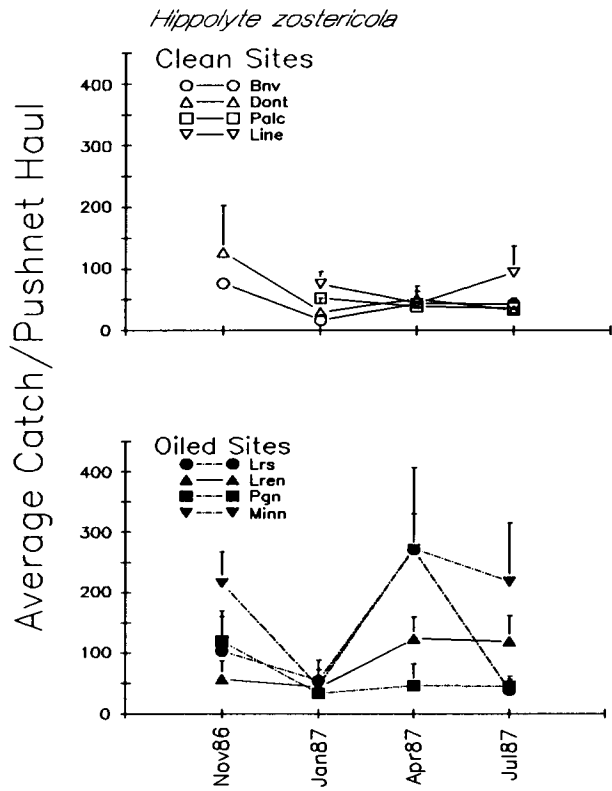


Figure 8.2 Abundance of *Hippolyte zostericola* in pushnet samples from oiled and unoled (=clean) seagrass meadows. Vertical bars represent one standard deviation. Site codes are those in Figure 8.1.

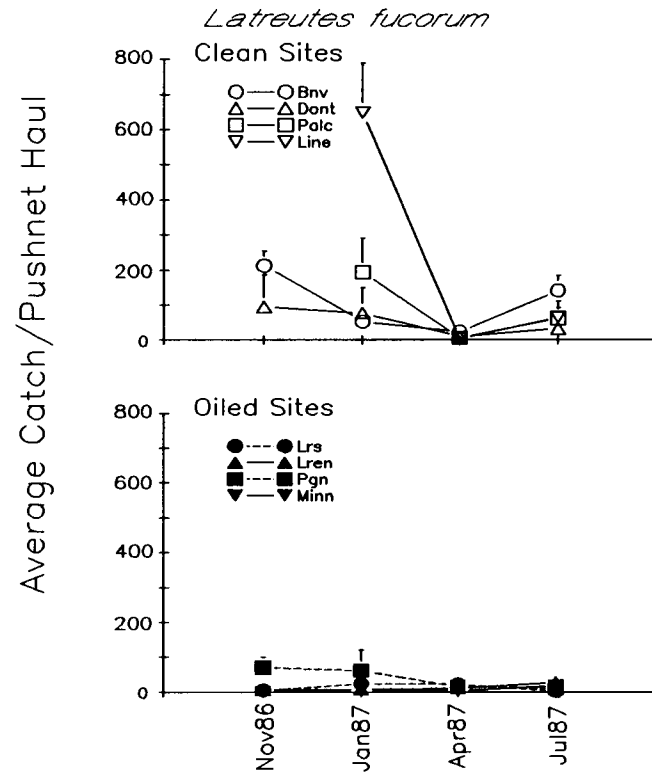


Figure 8.3 Abundance of *Latreutes fucorum* in pushnet samples from oiled and unoled (= clean) seagrass meadows. Vertical bars represent one standard deviation.

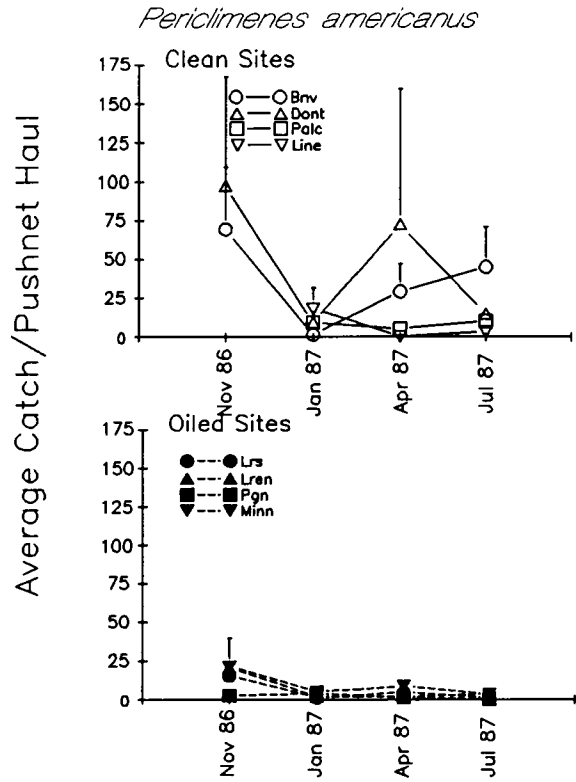


Figure 8.4 Abundance of *Periclimenes americanus* in core samples from oiled and unoiled (= clean) seagrass meadows. Vertical bars represent one standard deviation.

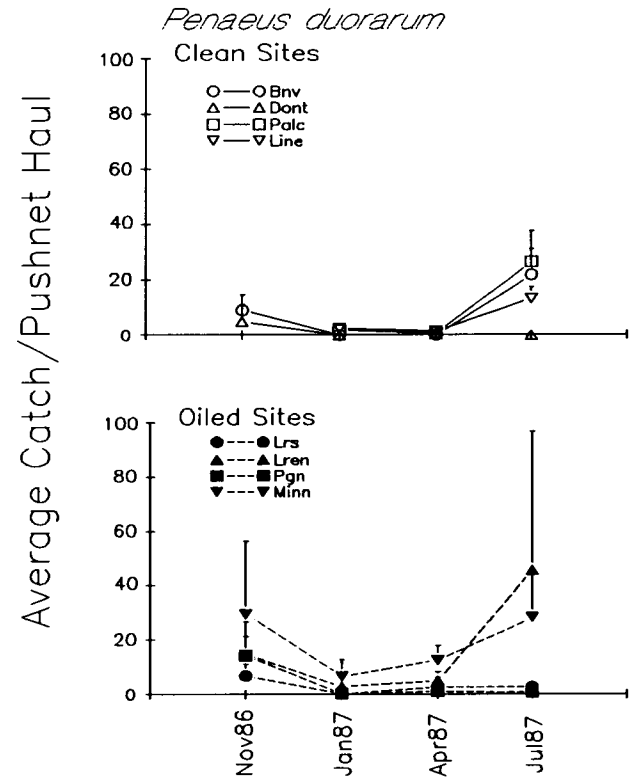


Figure 8.5 Abundance of *Penaeus duorarum* in pushnet samples from oiled and unoiled (= clean) seagrass meadows. Vertical bars represent one standard deviation.

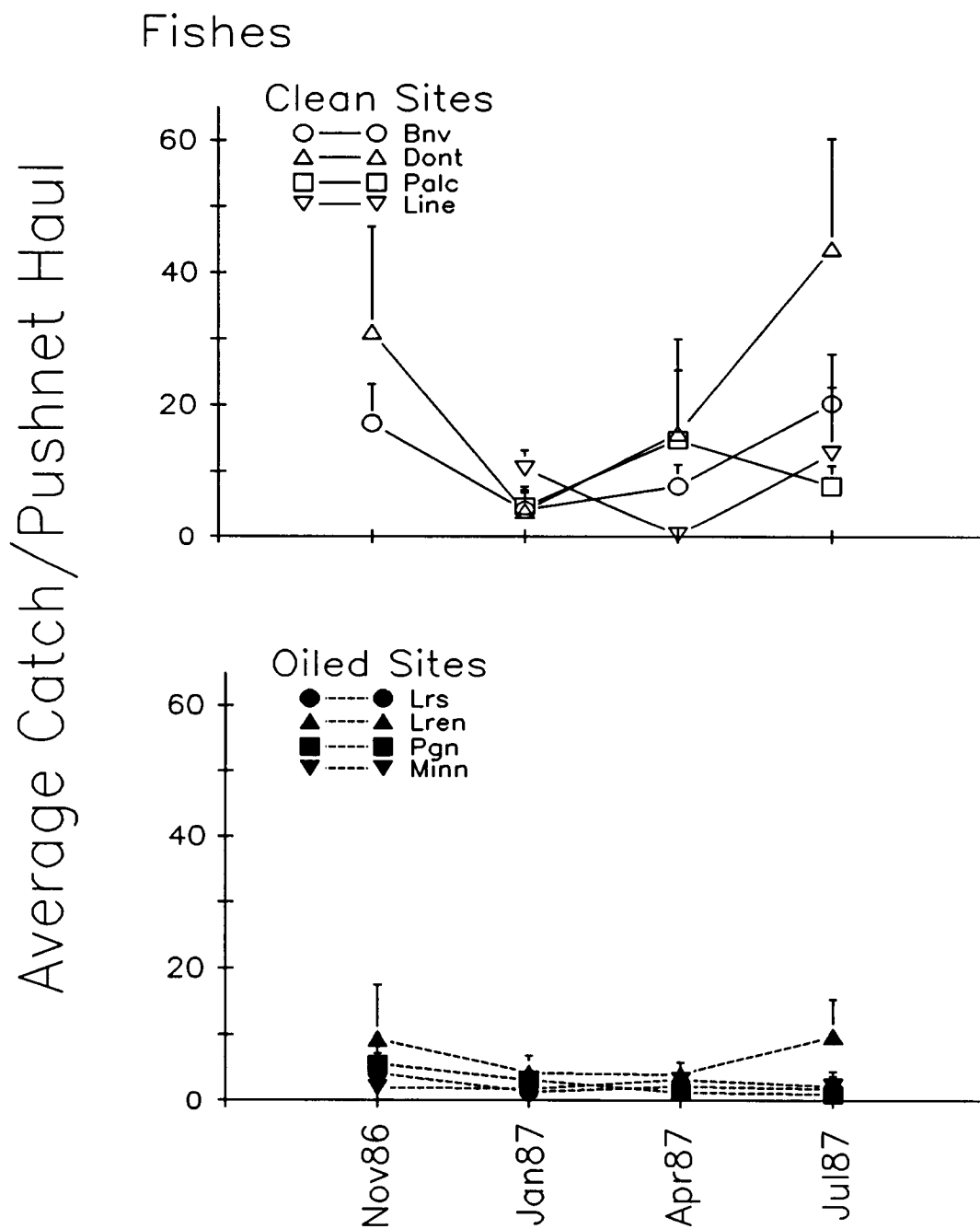


Figure 8.6 Abundance of fishes in pushnet samples from oiled and unoled (= clean) seagrass meadows. Vertical bars represent one standard deviation.

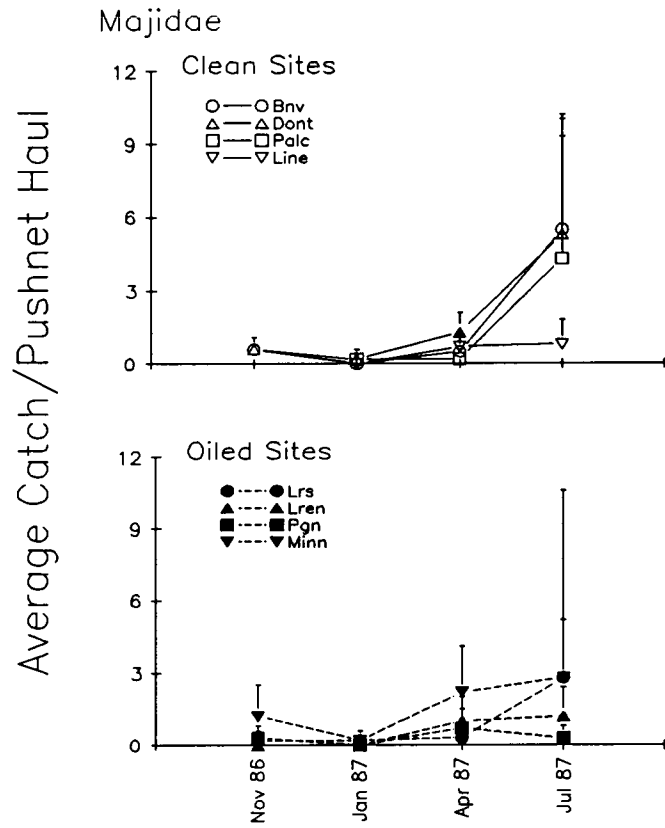


Figure 8.7 Abundance of Majidae in pushnet samples from oiled and unoled (= clean) seagrass meadows. Vertical bars represent one standard deviation.

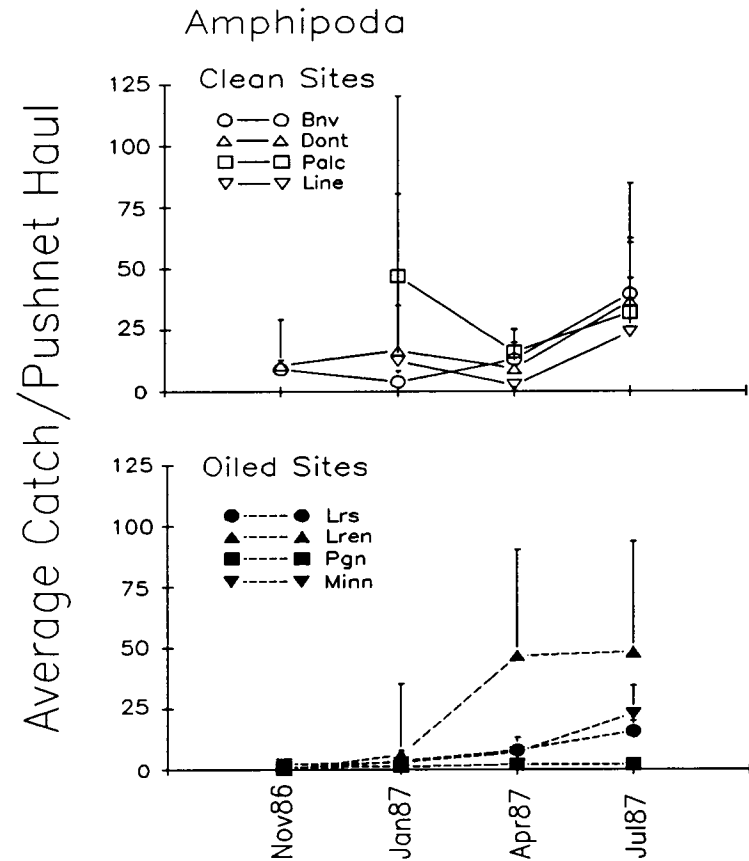


Figure 8.8 Abundance of Amphipoda in pushnet samples from oiled and unoled (= clean) seagrass meadows. Vertical bars represent one standard deviation.

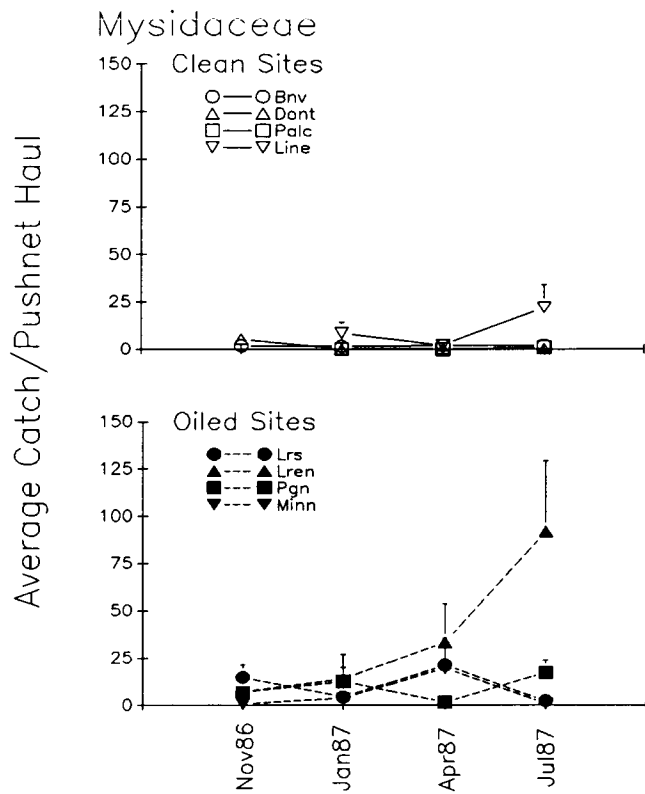


Figure 8.9 Abundance of Mysidaceae in pushnet samples from oiled and unoled (= clean) seagrass meadows. Vertical bars represent one standard deviation.

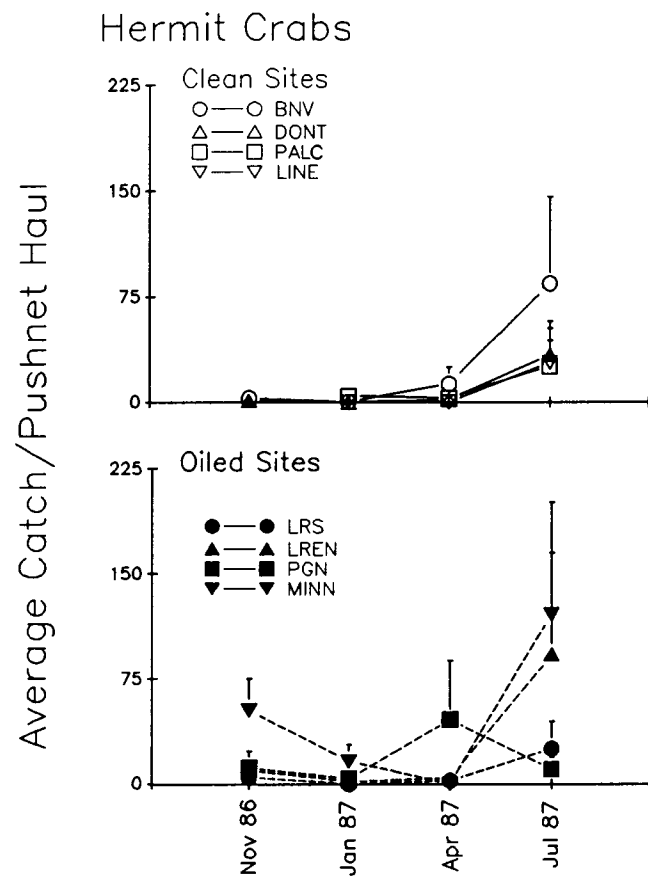


Figure 8.10 Abundance of hermit crabs in pushnet samples from oiled and unoled (= clean) seagrass meadows. Vertical bars represent one standard deviation.

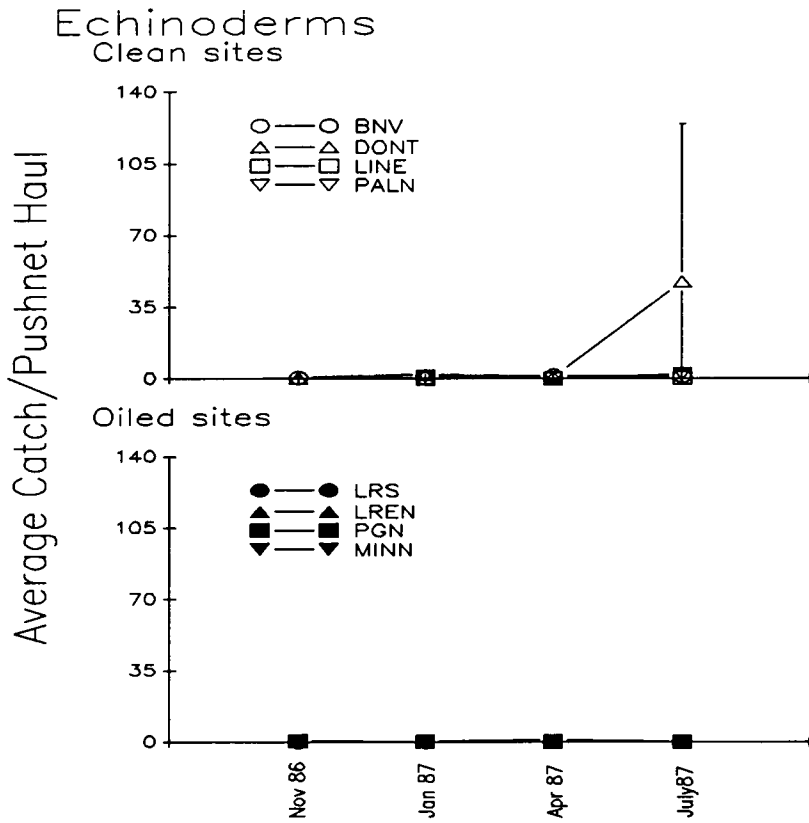


Figure 8.11 Abundance of echinoderms in pushnet samples from oiled and unoled (= clean) seagrass meadows. Vertical bars represent one standard deviation.

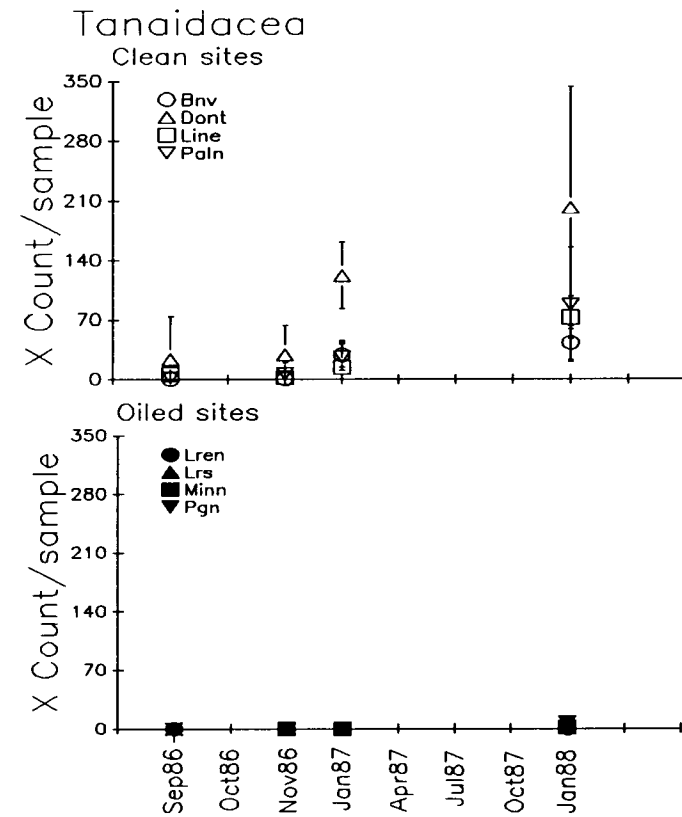


Figure 8.12 Abundance of Tanaidacea in core samples from oiled and unoled (= clean) seagrass meadows. Vertical bars represent one standard deviation.

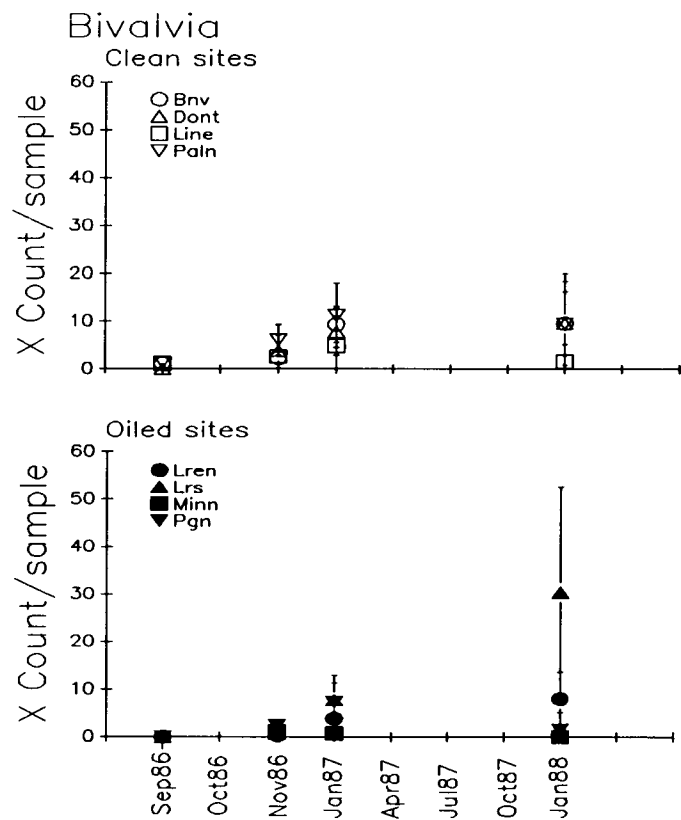


Figure 8.13 Abundance of *Bivalvia* in core samples from oiled and unoled (= clean) seagrass meadows. Vertical bars represent one standard deviation.

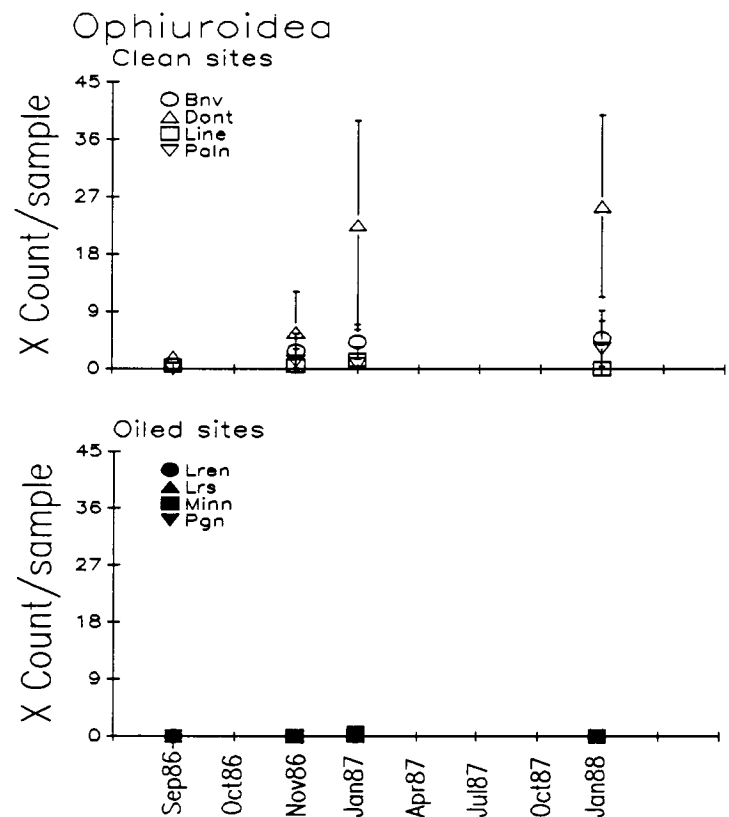


Figure 8.14 Abundance of *Ophiuroidea* in core samples from oiled and unoled (= clean) seagrass meadows. Vertical bars represent one standard deviation.

Polychaetes were omitted from this set of graphs because they are being identified to families and have not yet been counted for all collection dates.

8.3.3 Size-frequency Analyses

The abundance of males, females, and juveniles of the caridean shrimp *Hippolyte zostericola* at oiled and unoiled grassbed sites in November 1986 suggests that juvenile shrimp were adversely affected at all the oiled sites (Figure 8.15). Juveniles of *Latreutes fucorum* were similarly affected, but not at all sites. Further analyses of these data and completion of measurements of more species should indicate whether oil impact was life-stage dependent.

8.3.4 Multivariate Analyses

Sites from pushnet collections from November 1986 through January 1988 formed groupings (Figure 8.16) that could be attributed to the effects of oil or to differing environmental parameters (e.g., those listed in Table 8.1). Group "A" samples were from unoiled sites, with the exception of Margarita 2 (MAR2) which was moderately oiled. Group "B" included 7 oiled and 2 unoiled samples, group "C" included 9 oiled and 1 unoiled sample, group "D" included 4 unoiled and 1 oiled sample, group "E" included 8 unoiled sites, and group "F" included 10 oiled sites. No clusters were based entirely on collections at one site or collections made on a single sampling date. These patterns suggest that no "recovery" has occurred, since all clusters are entirely or almost exclusively composed of either oiled or unoiled sites. A second hypothesis to explain the observed cluster pattern is that the oiled sites were different from the unoiled sites prior to the oil spill. If this is the case, initial impacts and "recovery" cannot be evaluated through post-spill comparisons of oiled and unoiled "control" sites using this kind of analysis.

8.3.5 Seagrasses

Depth profiles (Figure 8.17) show the topographic features and grassbed widths at the eight sites monitored after January 1987. All grassbeds were separated from the shoreline by a thin band of sand or muddy sand. The seagrass meadows terminate in deeper water in zones of coral rubble, rock, or sand/mud flats. The grassbeds are of varying width; the narrowest bed is at Largo Remo South and the widest is at Palina North. These differences are attributable to variation in bottom slopes and reef development.

Total seagrass biomass (Figure 8.18) and blade biomass (Figure 8.19), as percent of the total biomass, did not vary much between oiled and unoiled sites, except for the relatively low total biomass at oiled sites in September 1986, the first collection after the spill. Oiled seagrass beds, at later sampling dates, appeared to be receding from the shoreline (Figure 8.20). This change in meadow edge positions may have started at a much earlier date; monitoring of this phenomenon did not

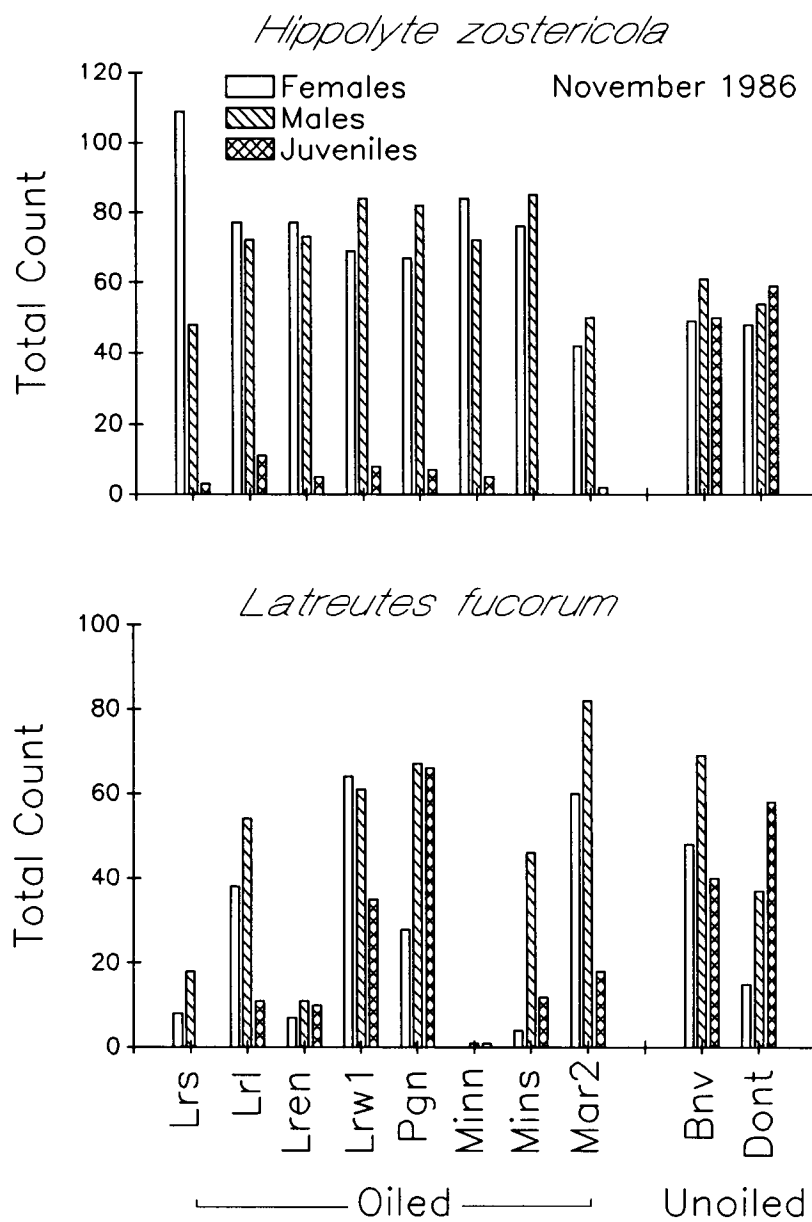


Figure 8.15 Distribution of male, female, and juveniles of the two caridean shrimps, *Hippolyte zostericola* and *Latreutes fucorum* in oiled and unoiled grassbeds. Site codes are given in the legend to Figure 8.1.

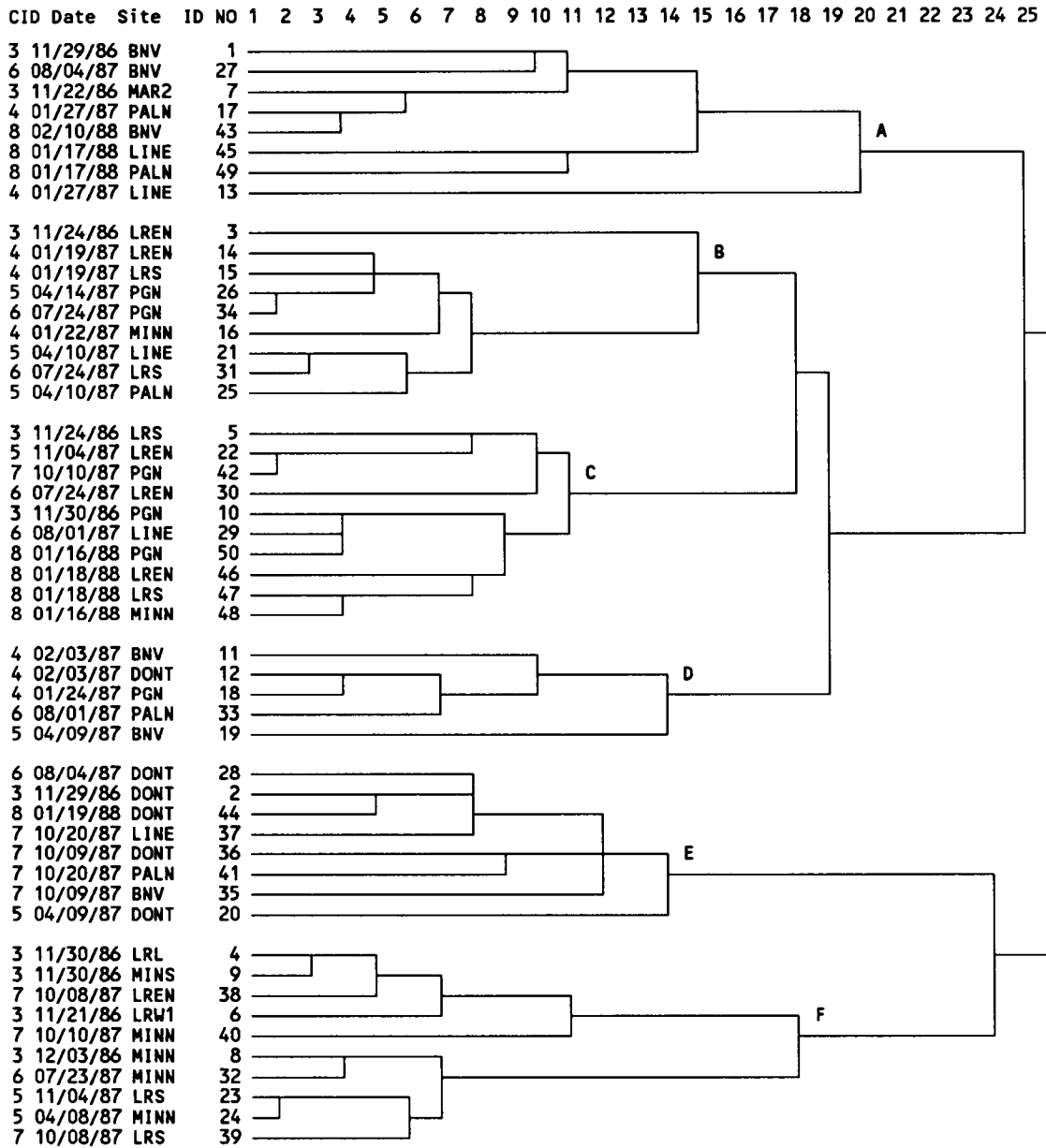


Figure 8.16 Dendrogram resulting from group-average cluster analysis of pushnet samples taken from control and oiled sites over the first six collection dates of the epifaunal study. Collection identification codes (CID) correspond to the collection dates (CID3 = November 1986, CID4 = January 1987, CID5 = April 1987 and CID6 = July 1987, CID7 = October 1987, and CID8 = January 1988).

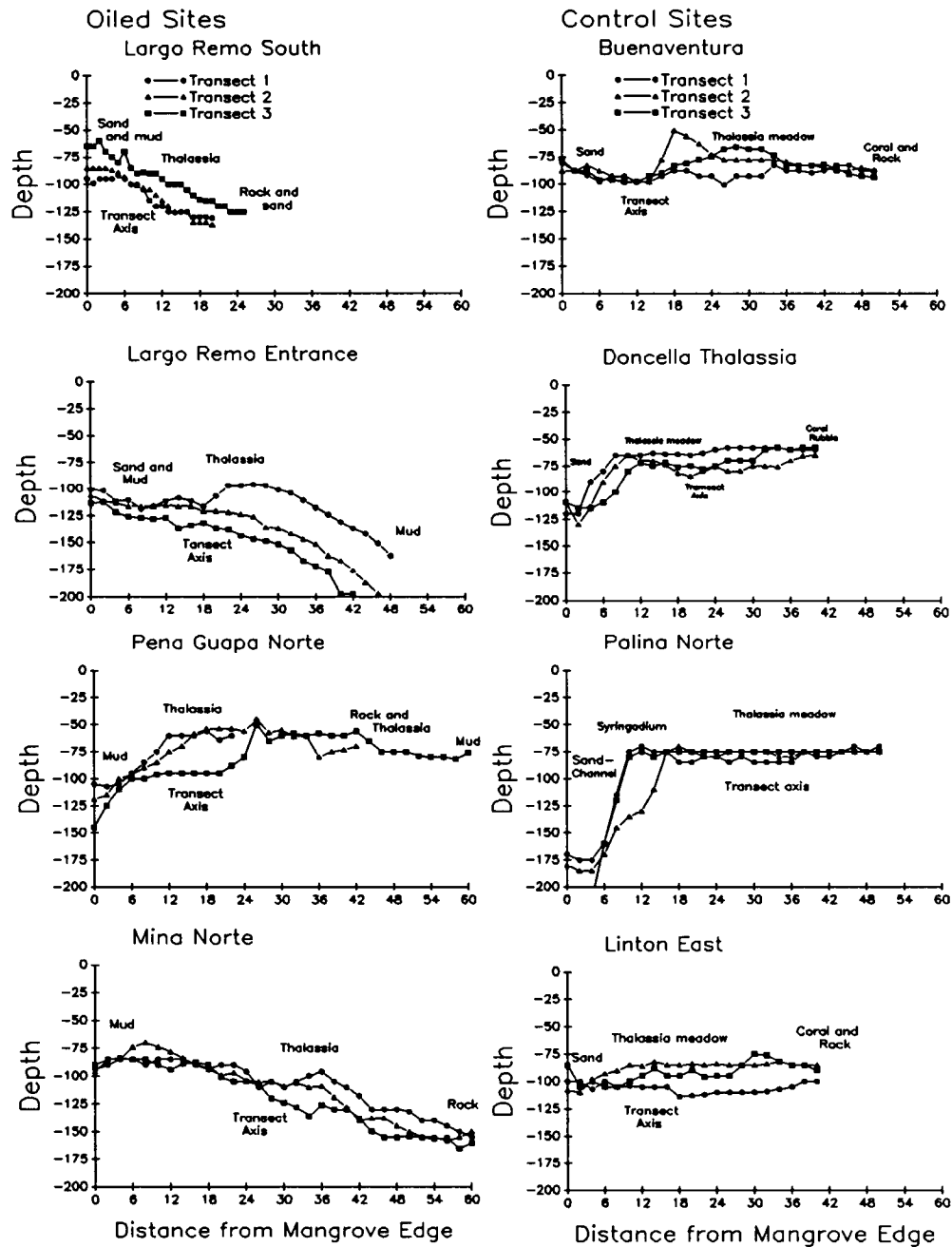


Figure 8.17 Depth (cm) against distance (m) from mangroves at each site.

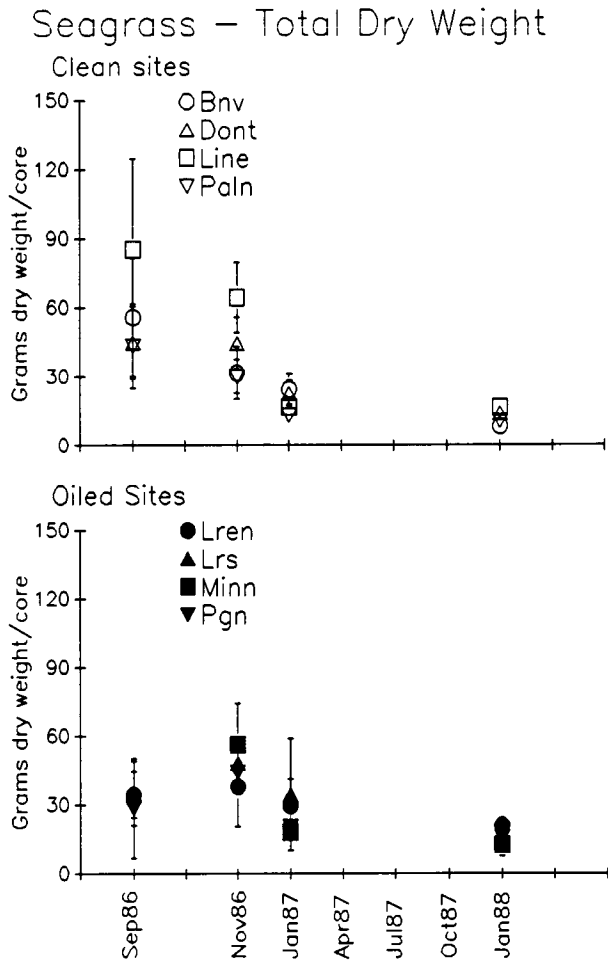


Figure 8.18 Total seagrass biomass at unoiled (= clean) and oiled seagrass meadows.

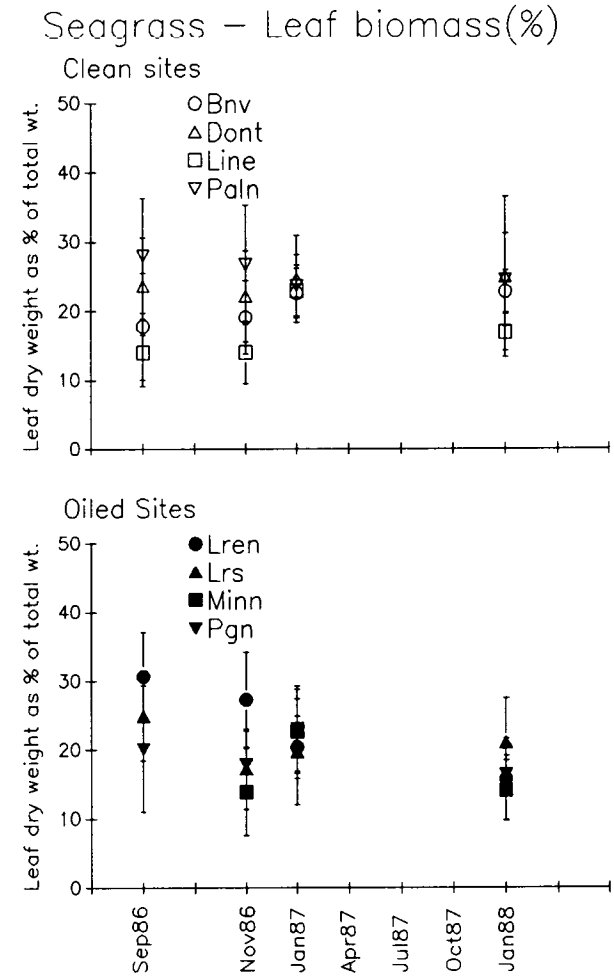


Figure 8.19 Seagrass blade biomass (as percentage of total biomass) at unoiled (= clean) and oiled seagrass meadows.

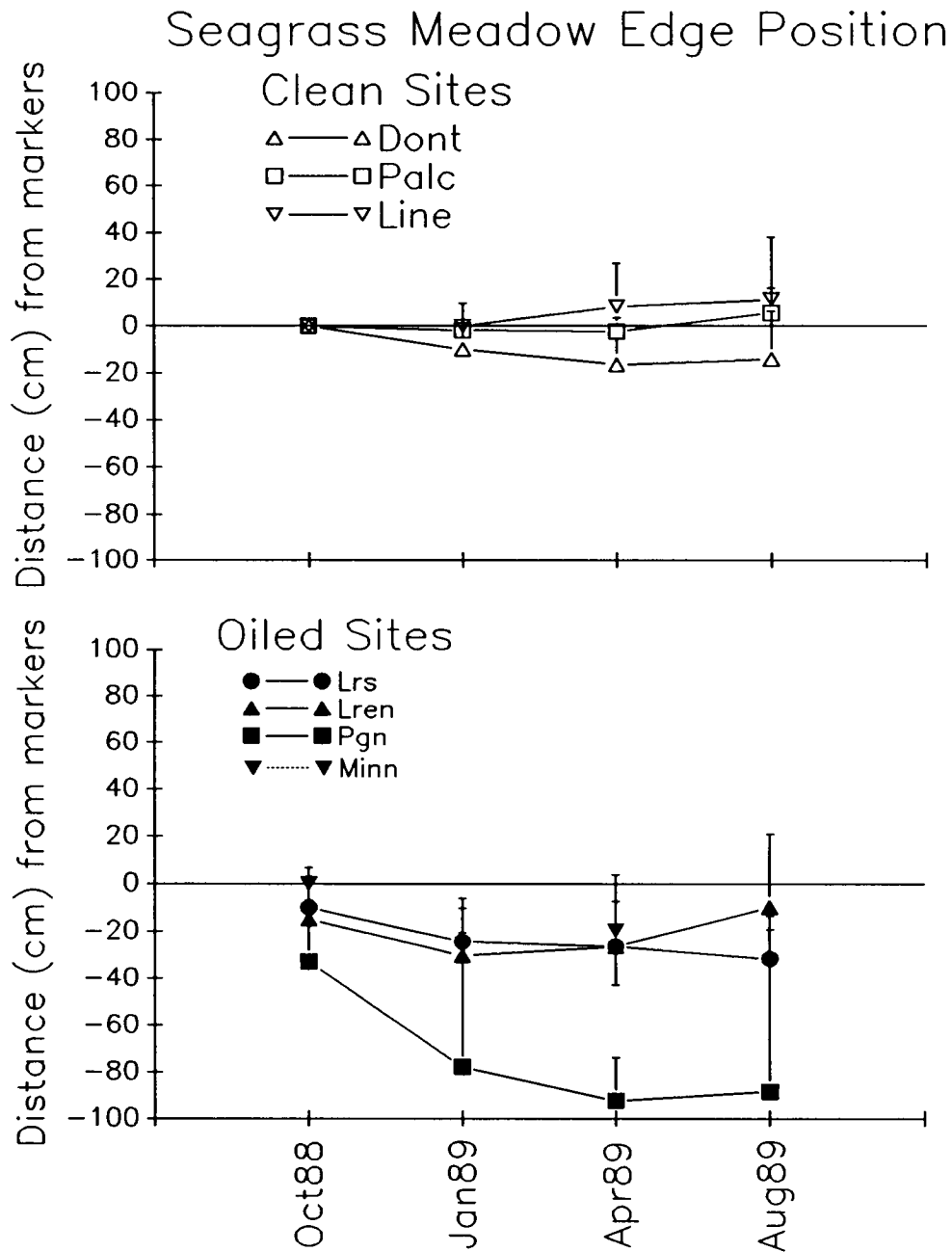


Figure 8.20 Cumulative changes in the location, with respect to marker posts, of the shoreward most edge of unoiled (= clean) and oiled seagrass meadows. All markers were repeatedly taken at the fourth unoiled site (BNV).

begin until October 1988 when we first noticed large changes in the position of the shoreward edge of the grassbed at LRS.

8.4 Discussion

The graphs presented here represent only some of the taxa collected at the oiled and unoiled seagrass meadows. It would be difficult to make any type of summary statement regarding the initial effects of the oil spill on seagrass meadow fauna based on these figures. A complete set of graphs for all infaunal and epifaunal taxa would obviously be even more difficult to interpret in terms of a hypothesized recovery process. More comprehensive analyses, utilizing MANOVAs and classification analyses, of the patterns shown by these figures are underway. The dendrogram shown in this report for the epifaunal assemblage suggests that classification techniques might be useful in analyzing the large data set that will result from the seagrass study. Date and site groupings in this dendrogram suggest that oiling produced major changes in animal distribution patterns; 46 of the 51 analyzed pushnet samples separated into clusters based on oiling. However, other explanations of this pattern are possible.

One of the most interesting aspects of this study, at the present time, is the apparent impact of oil on juvenile caridean shrimp. The few juvenile *Hippolyte zostericola* seen at oiled sites suggests that either recruitment at these sites was affected by oil exposure or that reproductive output is less when adults are exposed to oil. The variable effect of oil on juvenile *Latreutes fucorum* is more difficult to explain. Additional measurements of these two species collected at later dates are underway and analyses of these data will be presented later. It will be important to determine additional life history characteristics of each species in order to better interpret these data.

Oil may have had a negative initial effect on seagrass biomass, as suggested by the samples collected nearly five months after the spill (Figure 8.18). The seagrass in one heavily oiled intertidal meadow was killed and washed away along with grassbed sediments (Chapter 4). The receding shoreward edges, observed later in this study, of grassbeds at oiled sites might be due to chronic oiling from the heavily oiled mangrove forests at each of these sites. Much oil was buried in the forests and it may continue to affect seagrasses as it slowly washes out.

Chapter 9 Hydrocarbon Analyses

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Bermuda Biological Station for Research

9.1 Introduction

In April 1986 more than 50,000 barrels of medium-weight crude oil spilled from a ruptured storage tank into a complex region of mangroves, seagrasses and coral reefs on the Caribbean coast of Panama just east of the entrance to the Panama Canal (see Chapter 1). This spill, which is the largest recorded into coastal habitats in the tropical Americas, happened close to the Smithsonian Tropical Research Institute's marine laboratory at Galeta. As populations of plants and animals in both oiled and unoled areas had been previously documented, the study of this spill affords a unique opportunity to assess the effects of oil on tropical coastal ecosystems. Investigations were begun immediately and initial ecological effects were summarized by Jackson *et al.* (1989). Intertidal mangroves, seagrasses, algae and associated invertebrates were covered by oil and died soon after. There was also extensive mortality of subtidal reef corals and infauna of seagrass beds. Such strong subtidal effects stand in sharp contrast to conclusions based on laboratory dosing experiments and small-scale field experiments. These kinds of studies indicated that corals suffer only transient physiological effects from exposure to oil and should not be expected to exhibit mortality associated with spills unless the oil is dispersed into subsurface waters (Knap 1987; Ballou *et al.* 1987). Small amounts of dispersants were sprayed from aircraft over offshore areas and in restricted channels. Cubitt *et al.* (1987) reported that refinery officials estimated less than 21,000 l of Corexit 9527 were used. This amount of dispersant and the limited areas over which it was sprayed would have been inadequate to disperse the large amount of oil spilled. Thus chemical dispersion would not account for the mortality seen in subtidal corals over the extended areas documented in the study of this spill (see Chapter 5).

Chapter 9.2 summarizes results of the analysis of samples collected in 1986 for the short-term assessment of the impact of the Bahía Las Minas oil spill. The initial

results of this collection were used to validate the initial assessment of biological damage associated with oiling (see Jackson *et al.* 1989; Burns and Knap 1989). Ultraviolet fluorescence (UVF) spectral data and gas chromatography (GC) results are presented here. These and subsequent data sets will form the basis for correlation of the chemical and biological studies in the long-term damage and recovery assessment. Chapter 9.3 describes the second major sampling effort conducted in 1988 and 1989, details the inventory of samples archived for analysis, and presents observations on the condition of mangrove roots in sediment core samples. Chapter 9.4 is a description of the analytical quality assurance procedures used in the program.

9.2 Petroleum Hydrocarbons in Sediments, Corals, Seawater and Bivalves Collected in September 1986

9.2.1 Methods

Sampling sites

The coastline is convoluted, with strips of beach bordering exposed areas and mangroves lining sheltered areas. Offshore, there are subtidal seagrass beds with coral reefs to seaward. Photographs of the affected coastline are in Jackson *et al.* (1989) and Figure 9.1 is a map of the study area.

Samples of coral and associated sediments were collected from reefs where coral growth and mortality were being measured. Samples were taken from the same reefs, close to, but just outside the areas marked for assessment studies. Palina West (PALW; Figure 9.1) is an unoiled control reef in a relatively pristine area of the coast approximately 40 km northeast of the spill site. No oil from the 1986 spill was seen along this region of the coast during the spill, but frequent shipping traffic to and from the Panama Canal is an occasional source of tar balls and other contamination. Naranjos South (NARS) is located about 18 km upwind of the site of the spill and was visually categorized as moderately oiled. Sediments in small depressions on these two reefs were coarse coral rubble, indicating high-energy sorting despite the proximity of landward seagrass beds. Galeta Channel (GALC), about 20 km downwind of the spill site, and Payardi North (PAYN), at the spill site, were heavily oiled. Their associated sediments contained a large portion of small particles, indicating they retained more of the fine sediments eroded from seagrass and mangrove areas landward. No oil slicks were observed on the days corals were collected, although on other occasions slicks were observed to emanate from highly polluted mangrove areas due to runoff during heavy rains.

Subtidal sediments were collected in seagrass beds marked for biological assessment studies. Areas that had been marked for biological studies of mangrove mortality, leaf growth and litter production were sampled for hydrocarbon contents in intertidal sediments. Bivalves were sampled for use as biological indicator organisms, where possible.

Collections

Siderastrea siderea is a common massive coral; *Agaricia tenuifolia* forms thin plate-like colonies that project out from the reef surface. Samples of these scleractinian corals were collected by a diver at 3 to 6 m depth using hammer and chisel. Each piece was brought to the surface, using care not to touch the living portions. The corals were sealed in solvent-washed aluminum foil. Surface sediments were collected by a diver from small depressions on coral reefs and from seagrass beds using solvent-cleaned glass jars as scoops. These were sealed with screw cap lids lined with foil.

High-volume seawater samples were taken in areas of mangroves at Isla Largo Remo (Largo Remo Mangrove = LRM; Figure 9.1) and at Galeta Sand (GALS) by passing water through a glass column packed with Amberlite XAD-2 resin. Columns were attached to mangrove roots with the intakes positioned below the estimated low tide mark. Sampling took approximately 5 hours at flow rates of 150 to 400 ml/min over bed volumes of 100 ml resin. Upon retrieval, columns were sealed with glass caps and wrapped in foil.

At each mangrove study site, one deep core was taken by pounding an 8 cm diameter aluminum tube into the substratum with a sledge hammer. A machete was used to cut the core out of the dense mangrove roots. Since the holes filled quickly with oil, the bottoms of the cores, which could have been contaminated with surface oil during the sampling operation, were extruded and discarded. One deep layer at approximately 15 to 20 cm below the surface was then extruded onto a jar lid overlain with clean foil. The core edges were cut away and the remaining sample sealed in a glass jar. A core plunger was then inserted into the core bottom and the top 5 cm of sediment extruded. This was treated in the same manner as the deep section of the core and stored in a separate glass jar.

Triplicate surface samples of mangrove sediments were collected within a 1 m circle of the cores. To prevent cross contamination of samples, the sampling gear was cleaned with detergents followed by rinses with analytical grade acetone and hexane. Unoiled sites were sampled first, followed by lightly and finally heavily oiled sites based on visual assessment. The sediments in all of the mangrove areas consisted of densely compacted mangrove root material with fine sediment interspersed. Surface scrapes were biased toward loose sediment on the surface. Samples were transported on ice to the laboratory at Galeta where all but the water samples were frozen at -20°C. Coral and sediment samples and the refrigerated water sample columns were packed in ice chests and flown to the Bermuda Biological Station for Research (BBSR) for chemical analysis.

Analysis

Corals were thawed and tissue was blown off the skeleton with compressed air into clean glass beakers (Knap and Sleeter 1984). This technique removes most of

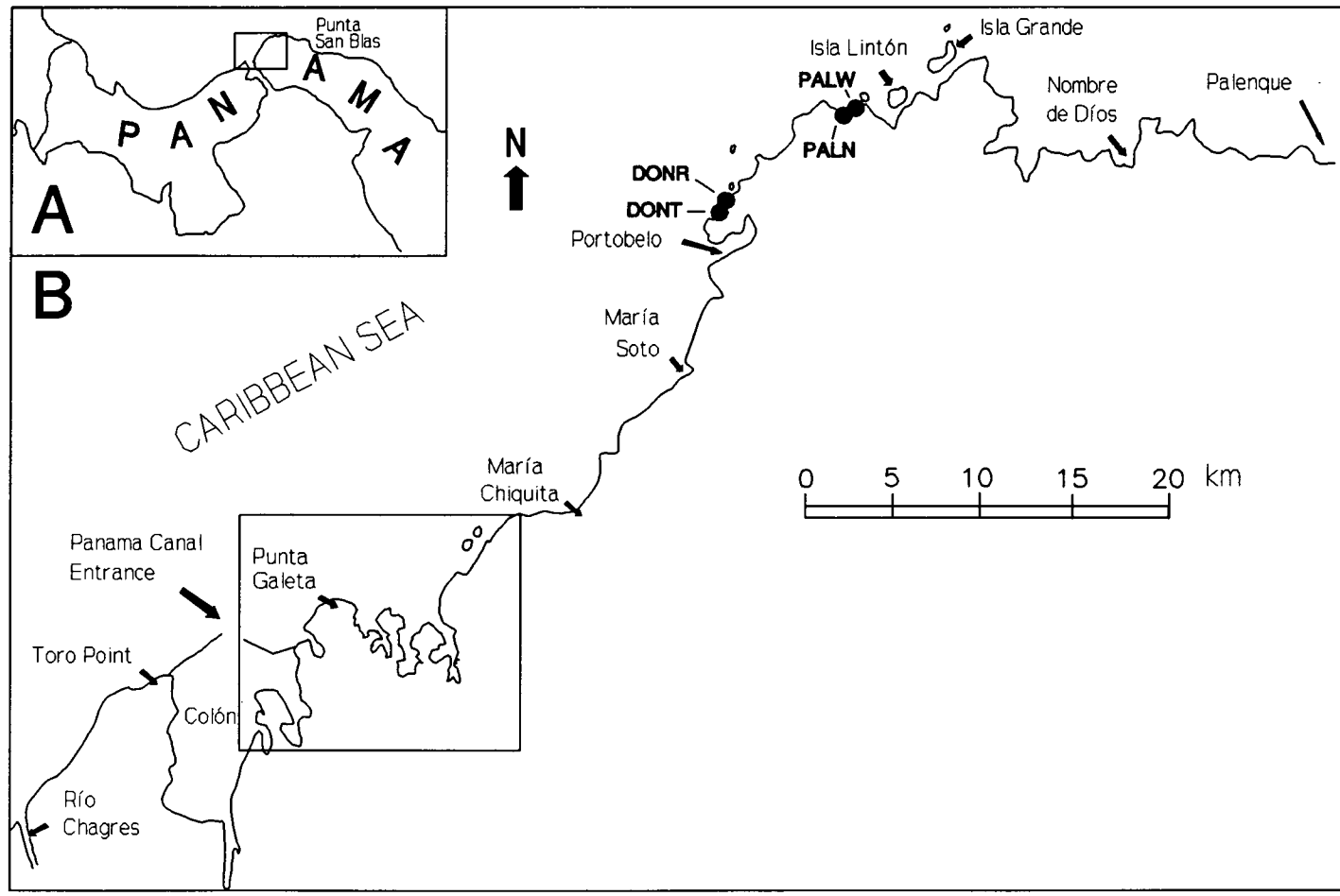


Figure 9.1 Map of sampling sites, September 1986. Refer to Figure 1.1 for details.

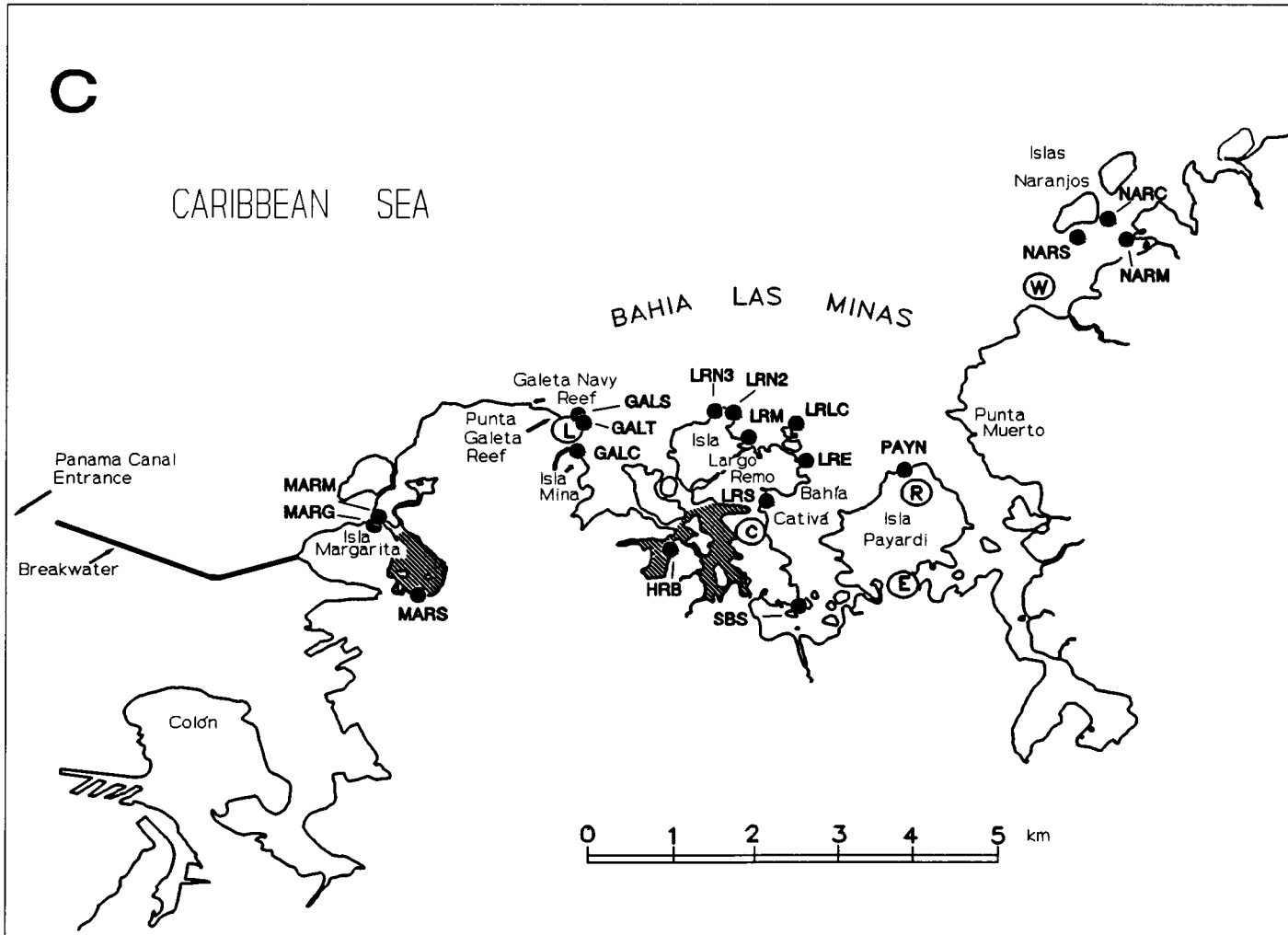


Figure 9.1 Map of sampling sites, September 1986 (continued).

the soft tissue of these species, but it was found in subsequent work on other species that scraping coral surfaces with stainless steel scalpels increases tissue recovery. Enough pieces of *S. siderea* were available for triplicate analyses, but *A. tenuifolia* had to be processed as single composite samples from each station. Because fragments of coral skeleton were included in the samples, it was not possible to determine an accurate wet weight of the tissues. Tissues were ground with precombusted Na_2SO_4 into a homogeneous paste. Protein determinations were made on aliquots of the sample paste as a measure of tissue mass using the Folin-phenol procedure (Lowry *et al.* 1951). The aliquots were diluted sufficiently to preclude interference by Na_2SO_4 , as determined by including the same amount of salt in the standards and blanks.

The three bivalve samples and four of the coral reef sediment samples were thawed. Large rocks and plant matter were removed from sediments and the samples were spooned into 500 ml round-bottom flasks and extracted for 8 hours using a hydrolysis reflux procedure with NaOH/MeOH as the extraction solvent (UNESCO 1982). Extracts were separated into non-saponifiable (NSL) and saponifiable (SL) lipid fractions, dried with Na_2SO_4 , and concentrated to 1 ml by rotary evaporation. Remaining sediment extractions were done by mixing the sediments with Na_2SO_4 to bind water followed by direct extraction with MeCl_2 (UNESCO 1990).

Total extractable lipid determinations were made by evaporating 10 μl aliquots of the extracts onto the pan of a microbalance. For saponified samples extractable organic matter (EOM) is the sum of the saponifiable and non-saponifiable lipid fractions. Elemental sulfur was removed from sediment extracts by percolating them over a small column of activated copper. Water samples were extracted in a specially designed continuous extractor using acetone with 10% water (Ehrhardt 1987). Total lipid extracts from the water samples or the NSL extracts from sediments and corals were separated into "saturated" and "unsaturated" hydrocarbon fractions by adsorption chromatography using alumina and silica gels or by normal phase high performance liquid chromatography (HPLC) (UNESCO 1990).

Hydrocarbons were analyzed by capillary gas chromatography (GC) with flame ionization detection (FID) on SE 52 fused silica columns with H_2 carrier gas. N-octadecene was added to extracts as an internal GC quantification standard. Compounds resolving into single peaks were quantified by means of response factors calculated from external standards over the GC elution range of C_{12} through C_{34} and naphthalene through picene. Unresolved components, typical of petroleum residues, were quantified by determining the GC areas gravimetrically and relating them to an average response factor over the appropriate elution range. Aromatic fractions were also analyzed by ultraviolet fluorescence (UVF) using hexane dilutions of samples of the spilled oil to generate the standard response curve. Extracts were made to appropriate dilutions to be read on the linear range of fluorescence emission versus concentration at 310 nm and 360 nm for the excitation and emission monochromators respectively. Further compositional detail was obtained by coupled excitation-emission spectra with wavelengths set 23 nm apart (UNESCO 1990).

After extraction of the lipids, residual sediments were dried at 90°C and sieved to determine the size distribution of particles. Sieves were stainless steel of standard mesh sizes 63 μm , 125 μm , 250 μm , 500 μm and 1000 μm . For mangrove cores, part of the 0-5 cm sections of the cores were dried, lightly ground with a mortar and pestle to break up peat and root material, and then sieved.

9.2.2 Uptake of Oil by Corals

Quantitative

Results of triplicate analyses of the *S. siderea* samples are shown in Table 9.1. The triplicates were collected over an area of several tens of square meters at depths of 3 to 6 m. The data include two estimates for the content of "petroleum" hydrocarbons in the tissue samples. These are the amount of unresolved compounds (URE) as determined by GC and the UVF oil equivalents of the aromatic fractions. The GC estimates expressed as mg URE g⁻¹ protein break the data into two groups: low and high contamination areas (Table 9.1). The same data expressed as mg URE g⁻¹ lipid break the data into low, medium and high contamination areas. The UVF oil equivalents g⁻¹ lipid break the data into unoiled, low, medium and high contamination areas. In all cases, the relative ranking of contamination is from the unoiled control at Palina West, through light to moderate levels at Naranjos South, moderate to high levels at Galeta Channel, to very high levels at Payardi North. This pattern is repeated in the quantitative results for *A. tenuifolia*. The results of the composite analyses for this species along with the means of the data for *S. siderea* and the overall evaluation of oiling severity based on oil concentrations in coral tissues are given in Table 9.2. Also listed in Table 9.2 are the protein to lipid ratios for both species at each site. This ratio appears to increase in the highly oiled samples.

Qualitative

Representative chromatograms of the saturated hydrocarbon fractions from corals at control and oiled reefs are shown in Figure 9.2. These may be contrasted with the chromatograms of the original oil and the seawater extracts shown in Figure 9.3. The hydrocarbons in the originally spilled oil span the C₁₀ to C₃₆ elution range as resolved on our system with the n-alkane peaks predominant over an unresolved mixture (URE). The carbon preference index (CPI), defined as the sum of odd carbon chain length n-alkanes divided by the sum of even carbon chain length n-alkanes over the C₁₀ to C₃₆ elution range is close to 1. The oil type was 70 % Venezuelan crude, 30 % Mexican Isthmus crude, with specific gravity 27° API at 15.6°C, and density of 0.89 gm cm⁻³.

Table 9.1 Concentrations of hydrocarbons in *Siderastrea siderea* expressed as mg g⁻¹ lipid or mg g⁻¹ protein as indicated. The horizontal lines mark significantly different sites (ANOVA, p<0.05).

Reef	Saturates			Unsaturates	
	Biogenics ¹ (mg g ⁻¹ lipid)	URE ² (mg g ⁻¹ lipid)	URE (mg g ⁻¹ protein)	UVF ³ oil (mg g ⁻¹ lipid)	URE (mg g ⁻¹ lipid)
Palina West	1.4	1.8	0.40	0.2	2.8
	0.3	0.6	0.11	>0.1	1.8
	0.2	0.6	0.15	>0.1	0.2
Naranjos South	2.4	1.6	0.30	1.8	2.5
	0.6	1.2	0.29	0.9	0.5
	0.6	1.3	0.40	1.0	0.6
Galeta Channel	3.8	2.6	0.82	5.8	2.5
	2.1	2.5	0.58	4.4	2.6
	1.5	2.5	0.71	2.7	1.9
Payardi North	2.8	4.6	0.72	23.6	2.8
	3.8	3.9	0.93	21.3	4.0
	3.3	6.8	0.63	41.4	4.3

¹Biogenics are the sum of multiple peaks with relative retention indices (RRI) near 1500, 1700 and 1900, which are common biogenic hydrocarbons produced by marine algae.

²URE is the GC signal generated by the complex mixture of hydrocarbon residues that cannot be resolved into individual peaks. This is a conservative estimate of "petroleum" hydrocarbons in samples.

³UVF oil units were determined against a standard curve made of the originally spilled oil.

Table 9.2 Concentrations of petroleum hydrocarbons and overall assessment of the severity of reef oiling. Hydrocarbons are expressed as mg URE g⁻¹ lipid and mg g⁻¹ protein. Also shown are protein to lipid ratios, and oiling severity based on tissue analysis of the corals.

Reefs	URE (mg g ⁻¹ lipid)	URE (mg g ⁻¹ protein)	UVF (mg g ⁻¹ lipid)	Protein Lipid	Oiling Severity
Palina West					
<i>Siderastrea</i>	1.0	0.2	0.1	4.6	none
<i>Agaricia</i>	0.3	0.1	>0.1	2.5	
Naranjos South					
<i>Siderastrea</i>	1.4	0.3	1.3	4.3	light - moderate
<i>Agaricia</i>	0.4	0.1	0.4	2.6	
Galeta Channel					
<i>Siderastrea</i>	2.5	0.7	5.0	4.2	moderate - heavy
<i>Agaricia</i>	2.8	1.0	3.1	4.5	
Payardi North					
<i>Siderastrea</i>	5.1	0.8	25.3	6.1	very heavy
<i>Agaricia</i>	8.3	1.2	49.7	6.4	

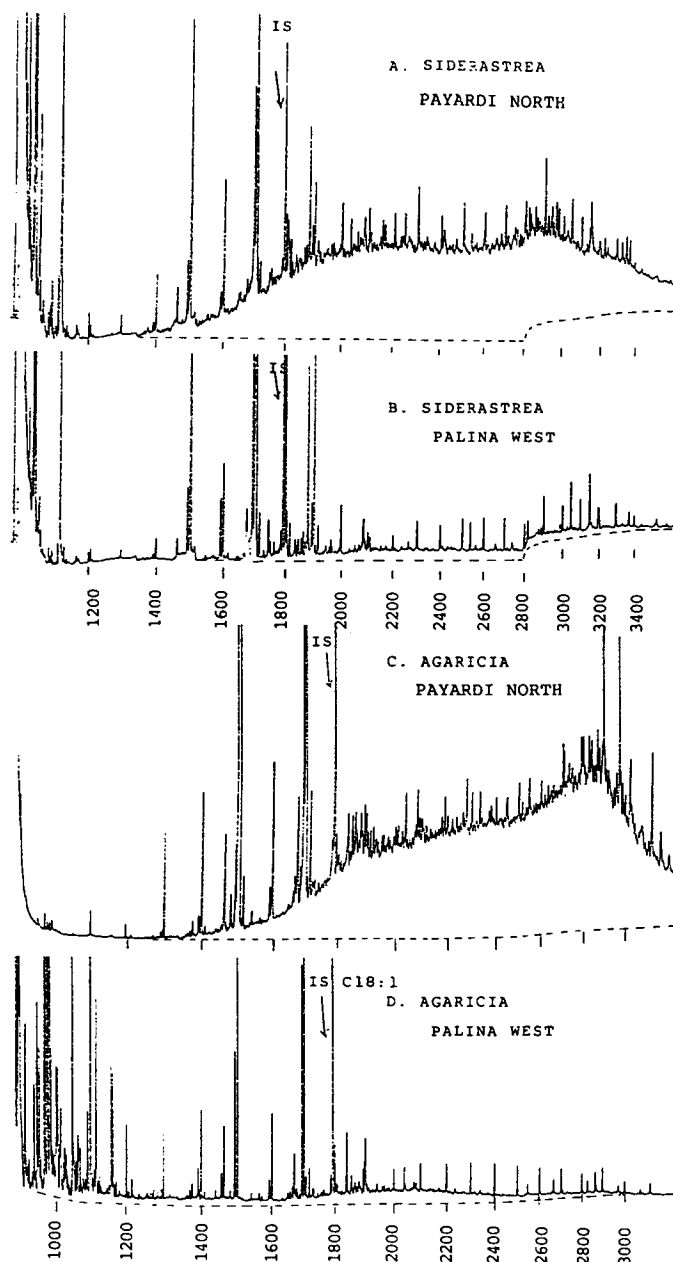


Figure 9.2 Gas chromatograms (with flame ionization detection) of saturated hydrocarbons from corals. Payardi North and Palina West are the oiled and unoled reefs, respectively. Shown are the elution positions of n-alkane standards used to compute relative retention indices of resolved peaks on SE-52 fused silica capillary columns. "IS" is n-octadecene added as internal standard. Dashed line is the column bleed signal. See Table 9.2 for concentrations of petroleum hydrocarbons.

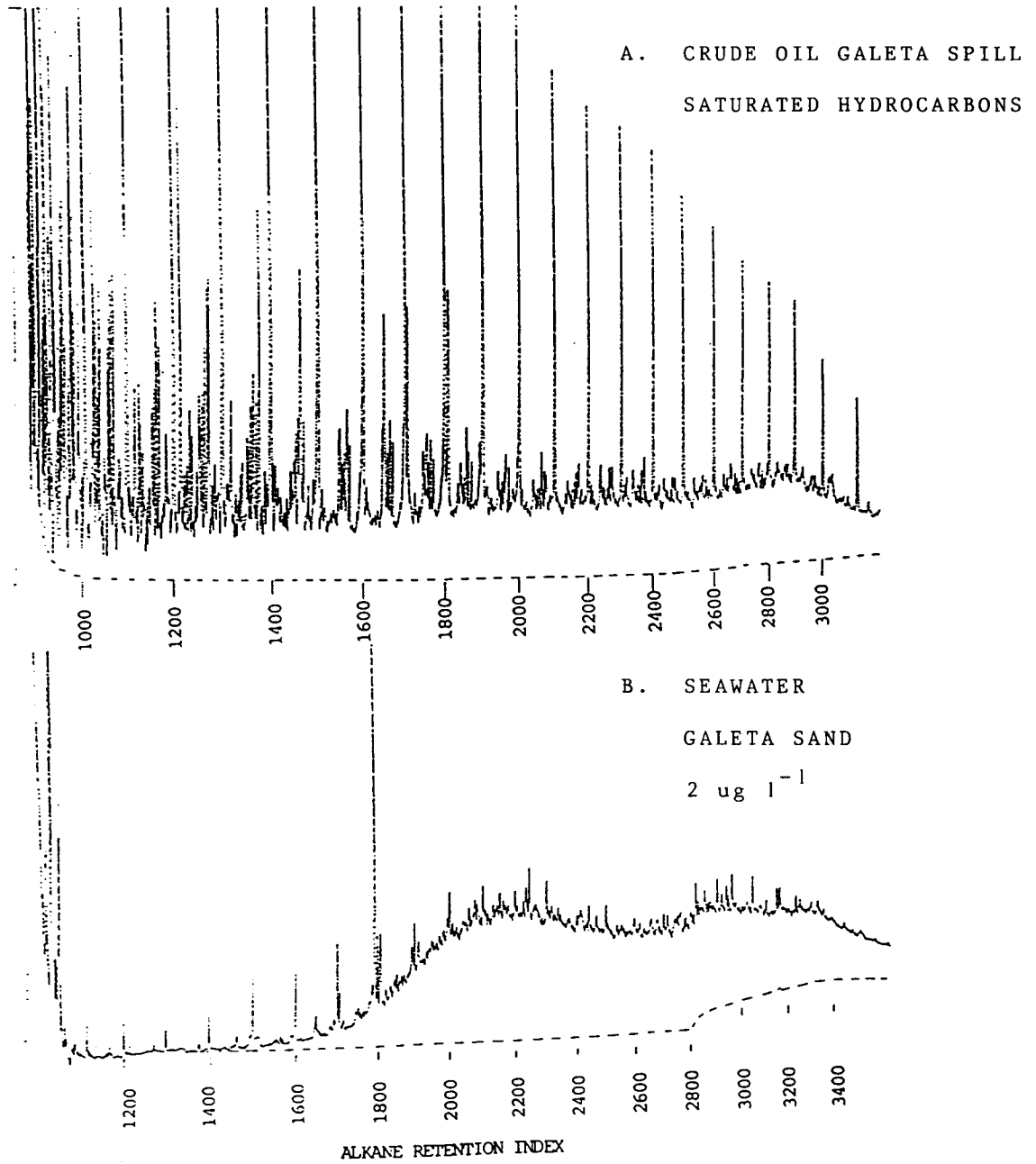


Figure 9.3 Gas chromatograms of saturated hydrocarbons in VMIC oil and suspended in seawater near mangroves at Galeta Sand (Figure 9.1), September 1986.

Characterization of this oil through our adsorption chromatography procedure showed it was 47.4% saturates, 6.4% light aromatic, 4.8% heavy aromatic, 18.9% more polar aromatic hydrocarbons that required MeCl₂ for elution, and 22.5% that remained on the column and was unrecovered. The corals from unoiled reefs show clusters of discrete peaks at relative retention indices (RRI) typical of the patterns of biogenic hydrocarbons synthesized by marine algae (RRI around 1500, 1700, and 1900) (Blumer *et al.*, 1971). These were likely to have been synthesized by the dinoflagellate symbionts in the coral tissues. Some of the other discrete peaks may also be biogenics. There is negligible contribution from the higher plant waxes in the C₂₁-C₃₁ elution range, which would be expected if the corals accumulate biogenic hydrocarbons from the surrounding seagrass and mangrove ecosystems. Corals from the oiled areas show similar patterns of biogenics, but also the accumulation of hydrocarbons representative of a highly degraded oil. The oil pattern is similar to that seen in the water samples collected at the oiled mangrove area (Figure 9.3). Compared with the original oil, the volatile and low boiling components are missing as are most of the n-alkanes and other resolved hydrocarbons. This emphasizes the residual unresolved pattern of hydrocarbons typical of highly degraded oil residues.

These data document the uptake of dissolved or dispersed hydrocarbons by subtidal corals as a result of this large oil spill in Panama. The corals appear to take up the hydrocarbons primarily from the water column as indicated by the GC patterns. With similar levels of oil in the sediments but a significantly lower concentration of hydrocarbons in coral tissues at Payardi North than at Galeta Channel (when expressed per gram lipid), it would appear that the oil in the sediments is not necessarily available for uptake by corals. This conclusion is suggested by the lack of biogenic hydrocarbons typical of seagrasses or mangroves in the corals, despite the location of reefs in areas where they would be expected to receive significant input of detritus from these contiguous ecosystems. High levels of oil hydrocarbons were suspended in seawater at oiled mangrove sites (Table 9.3). This indicates that the highly oiled mangrove sediments may be a source of contaminated water for extended periods of time after the original spill. Coral mortality, as shown by decreases in coral cover, ranked the severity of oiling in the same order as that determined by analyzing the hydrocarbon composition of the surviving corals. The coral data in Jackson *et al.* (1989) showed percent cover decreased by 79% at 3 m depth and 68% at 9 to 12 m depth on the Galeta Channel reef. On this basis, Jackson *et al.* (1989) ranked this reef as heavily oiled. Reductions in coral cover were significantly less on light to moderately oiled reefs and were not observed on unoiled reefs.

Table 9.3 Concentrations of petroleum hydrocarbons in water samples from two oiled mangrove sites. Determination by GC analysis of saturated fractions and UVF analysis of aromatic fractions.

Site	Volume (l)	URE ($\mu\text{g l}^{-1}$)	UVF ($\mu\text{g l}^{-1}$)
Largo Remo Mangrove	53	4.5	15.1
	45	6.7	29.1
Galeta Sand	115	1.9	4.8

Samples collected by pumping seawater through glass columns filled with Amberlite XAD 2 resin. Columns were tied to the roots of mangrove trees with intakes approximately 0.5 m below the water surface.

Corals have been shown to incorporate petroleum hydrocarbons into their tissues as a result of dosing in experimental systems (Peters *et al.* 1981; Knap *et al.* 1982). In addition to documenting the incorporation of petroleum hydrocarbons into coral tissue and demonstrating a positive correlation between tissue burden and mortality, the data from this field study also suggest a possible modification in the protein to lipid ratios of corals heavily stressed by oil. Although the sample size is too small to show a statistically significant change in this ratio, the trend is consistent with earlier laboratory studies in which corals were stressed by additions of dispersed oil. Cook and Knap (1983) reported a temporary reduction in photosynthetic rate and a decrease in lipid biosynthesis, with wax ester and triglyceride synthesis reduced 97% and 95%, respectively, relative to undosed control corals. Fatty acid, sterol and polar lipid synthesis rates were reduced 83% to 21% when compared with the controls. While these effects were only visible for a few hours in the laboratory tank experiments, the data from the Galeta Sand samples collected five months after the oil spill indicate such physiological effects may be chronic in highly contaminated reef systems. Rinekevich and Loya (1979) reported that corals on reefs subject to chronic oil pollution showed decreased reproductive success. Peters *et al.* (1981) observed impaired development of reproductive tissues, degeneration and loss of symbiotic zooxanthellae, and atrophy of mucus secretory cells and muscle bundles in corals exposed to oil in long-term tank experiments. In a detailed study of the effects of petroleum hydrocarbons on the lipid metabolism of larval lobsters, Capuzzo *et al.* (1984) demonstrated that oil exposure lowers levels of triglycerides and raises levels of sterols compared to controls. These authors also noted that fatty acid stores were altered, indicating decreased storage in energy reserves and decreased mobilization of essential fatty acids into phospholipid pools. These alterations in lipid metabolism were related to alterations in larval development, metamorphosis and growth rates (Capuzzo *et al.* 1984). Alterations in lipid metabolism would also be expected to have profound effects on the physiology and reproductive success of corals.

A large portion of the energy fixed in the algae-coral photosynthesis process goes into mucus production (Crossland *et al.* 1980). This mucus, which is rich in wax esters, triglycerides and other lipids, is an energy-rich link in the coral reef food chain (Benson and Muscatine 1974). Thus, if the lipid synthesis system is disrupted in corals heavily stressed by petroleum hydrocarbons, not only would coral physiology be adversely affected, but the impact would cascade to other components in the ecosystem that are dependent on the corals for food and substrata.

With this rationale, the impact of oil on the lipid composition of coral tissues will be assessed in addition to oil contents as part of the long-term study.

9.2.3 Oil Contamination in Coastal Sediments

The objectives of the analysis of the small volume surface sediment samples were 1) to estimate mean oil levels in sediments for establishing dose-response relationships with the biological assessment studies, 2) to assess variability due to spatial and temporal heterogeneity of oil distributions within a particular study site, and 3) to document special features of the oil distribution as visible when samples were collected. UVF analyses showed the relative distribution of oil in surface sediments and were useful for confirming the relative severity of oiling at the various sites. However, calculated concentrations were an overestimate of the actual amount of oil in the sediments as measured gravimetrically or by GC. This is because UVF analysis is sensitive only to fluorescent aromatic hydrocarbons. The light aromatics are water soluble and subject to rapid removal from environmental samples by dissolution and evaporation processes. Residual higher molecular weight hydrocarbons generally have higher fluorescent intensities than the low molecular weight hydrocarbons. Thus, to improve the quantitative accuracy of the UVF analysis, the weathered oil extracted from the heavily contaminated mangrove sediment at station SBS (Figure 9.1) was used to generate a new response graph for the UVF analyses. The 98 surface sediment samples were then recalculated to yield oil estimates in units of degraded Venezuelan/Mexican Isthmian crude (D.VMIC). Table 9.4 contains the UVF data for all of the 1986 sediment samples tabulated according to ecosystem type. Sampling sites are shown in the map in Figure 9.1.

Table 9.4 Oil content of sediments collected in 1986. Concentrations of oil were estimated by UVF analysis using degraded VMIC as the reference standard. Also shown are sediment water content and size composition, and UVF spectrum type. See Figure 9.1 for site locations.

Depth (cm)	% Water	Area Sampled	µg oil/ g dry wt.	% >1000µ	% >125µ	% <125µ	UVF spectrum
Mangrove sediments							
Heavily oiled							
0-1	84%	LRM	37,881				
0-1	82%	(Surface,	17,037				
0-1	86%	seaward front)	50,283				VMIC
0-5	85%	LRM Core	1,830	62%	38%	0%	VMIC
15-20	79%	LRM Core	1,003	10%	78%	13%	VMIC
0-1	83%	LRM	63,581				VMIC
0-1	86%	(Surface,	49,275				
0-1	85%	mid-grove)	123,421				
0-5	81%	LRM Core	35,195	16%	67%	17%	VMIC
10-15	84%	LRM Core	513	62%	38%	0%	VMIC
0-1	93%	LRM	87,735				VMIC
0-1	89%	(Surface,	8,840				VMIC
0-1	91%	high berm)	372,856				
0-1	79%	SBS	66,848				VMIC
0-1	72%	(Surface,	72,463				VMIC
0-1	70%	seaward front)	12,439				VMIC
0-5	74%	SBS Core	256,000	11%	88%	1%	VMIC
15-20	79%	SBS Core	210	24%	76%	0%	VMIC
0-1	78%	SBS	189,962				
0-1	80%	(Surface,	92,228				VMIC
0-1	82%	mid-berm)	159,513				VMIC
0-5	81%	SBS Core	120,254	42%	50%	8%	VMIC
15-20	82%	SBS Core	30	35%	60%	5%	Tr.VMIC
0-1	83%	SBS	42,967				VMIC
0-1	81%	(Surface,	15,198				VMIC
0-1	83%	inland grove)	21,824				VMIC
0-5	84%	SBS Core	6,929	8%	75%	16%	VMIC

Table 9.4 Oil content of sediments collected in 1986 (continued).

Depth (cm)	% Water	Area Sampled	µg oil/g dry wt.	% >1000µ	% >125µ	% <125µ	UVF spectrum
Mangrove sediments							
Moderately oiled							
0-2	87%	MARS 7/86 HC	1,091	42%	50%	7%	L. Fuel
0-2	82%	MARS 7/86 HC	1,976				L. Fuel
0-1	79%	MARM	2				Biog
0-1	79%	(Surface,	4				Biog
0-1	81%	seaward of boom)	6				Tr.
0-5	81%	MARM Core	650	13%	73%	14%	VMIC
15-20	79%	MARM Core	1,589	42%	50%	8%	D.H.Fuel
Lightly oiled							
0-5	47%	NARM Core	182	17%	77%	6%	VMIC
15-20	67%	NARM Core	12	9%	76%	15%	VMIC
0-1	78%	MARS	5				Biog
0-1	79%	(Surface,	5				Biog
0-1	78%	inside of boom)	3				Biog
0-5	79%	MARS Core	250	19%	70%	10%	Tr.VMIC
15-20	73%	MARS Core	18	39%	53%	8%	H.Fuel
Unoiled							
0-1	90%	HRB	151				
0-1	91%	(Surface,	181				VMIC
0-1	90%	seaward front)	138				
0-5	88%	HRB Core	85	23%	67%	10%	W.H.Fuel
0-1	91%	HRB	46				
0-1	89%	(Surface,	30				W.Crude
0-1	89%	mid-grove)	38				
0-5	86%	HRB Core	70	32%	60%	9%	W.L.Fuel
15-20	84%	HRB Core	36	52%	44%	5%	W.L.Crude
0-1	90%	HRB	53				W.Crude
0-1	89%	(Surface,	67				
0-1	91%	inland grove)	47				W.Crude
0-5	87%	HRB Core	131	17%	74%	9%	W.L.Crude

Table 9.4 Oil content of sediments collected in 1986 (continued).

Depth (cm)	% Water	Area Sampled	µg oil/ g dry wt.	% >1000µ	% >125µ	% <125µ	UVF spectrum
Seagrass sediments							
Heavily oiled							
0-2	68%	LREN	7,274	5%	59%	36%	VMIC
0-2	65%	Surface	11,070	7%	54%	40%	
0-2	64%		7,132	5%	76%	19%	VMIC
0-2	67%		24,555	4%	68%	28%	
Moderately oiled							
0-2	40%	LRS	335	30%	67%	2%	VMIC
0-2	39%	Surface	97	29%	69%	2%	VMIC
0-2	39%		331	33%	65%	2%	
0-2		NARC	150	4%	54%	42%	VMIC
0-2	41%	Surface	163	2%	61%	36%	VMIC
0-2	52%		48	7%	57%	36%	VMIC
0-2	47%	MARG (6/86 HC)	18	7%	59%	34%	VMIC
0-2	70%	Surface	144	100%	0%	0%	VMIC
0-2	48%		104	3%	69%	28%	W.VMIC
0-2	47%		59	9%	70%	21%	VMIC
0-2	48%	MARG (6/86 HC)	63	4%	63%	33%	VMIC
0-2	48%	Surface	28	2%	65%	33%	
0-2	49%		81	3%	66%	31%	H.Fuel
0-2	47%		60	2%	69%	29%	H.Fuel
0-2	39%	LRN2	4	33%	57%	10%	
0-2	40%		7	30%	64%	6%	Biog
0-2	41%		5	21%	68%	11%	Biog
0-2	37%		350	28%	68%	5%	
0-2	36%		35	33%	62%	5%	
0-2	37%		8	50%	47%	4%	
0-2	41%		2	44%	50%	6%	
0-2	37%		20	32%	64%	4%	VMIC
0-2	39%		14	25%	72%	3%	
0-2	39%		7	14%	82%	4%	Biog
0-2	39%		500	21%	73%	6%	

Table 9.4 Oil content of sediments collected in 1986 (continued).

Depth (cm)	% Water	Area Sampled	µg oil/ g dry wt.	% >1000µ	% >125µ	% <125µ	UVF spectrum
Seagrass sediments							
Lightly oiled							
0-2	30%	LRLC	7	20%	74%	7%	VMIC
0-2	29%		13	21%	72%	7%	
0-2	29%	Offshore	4	20%	74%	6%	
0-2	29%	Lagoon	4	22%	74%	5%	
0-2	44%	100 m off western	3	33%	65%	3%	
0-2	42%	edge of Largo Remo	5	53%	46%	1%	Biog
0-2	51%	Lagoon shore LRN3	8	35%	59%	7%	
Unoiled							
0-2	37%	PALN	0	45%	52%	3%	H.Fuel
0-2	36%		0	48%	49%	3%	
0-2	39%		0	44%	53%	3%	
0-2	39%		216	43%	56%	1%	H.Fuel
0-2	39%	DONT	0	21%	71%	8%	H.Fuel
0-2	37%		0	37%	59%	4%	
0-2	37%		0	32%	65%	4%	
0-2	37%		0	31%	64%	5%	--
Coral reef sediments							
Heavily oiled							
0-2	39%	PAYN	193	49%	44%	6%	
0-2	36%		197	60%	36%	3%	VMIC
0-2	43%		217	48%	47%	5%	VMIC
0-2	40%		79	51%	47%	2%	VMIC
0-2	52%	GALC	221	13%	70%	17%	
0-2	47%		364	21%	64%	15%	VMIC
0-2	48%		136	16%	69%	15%	
0-2	48%		715	16%	78%	6%	VMIC

Table 9.4 Oil content of sediments collected in 1986 (continued).

Depth (cm)	% Water	Area Sampled	µg oil/ g dry wt.	% >1000µ	% >125µ	% <125µ	UVF spectrum
Coral reef sediments							
Moderately oiled							
0-2	47%	LRE	41	21%	79%	0%	VMIC
0-2	44%		54	29%	71%	1%	
0-2	44%		44	27%	72%	1%	
0-2	44%		19	27%	72%	0%	VMIC
Lightly oiled							
0-2	29%	NARS	2	52%	47%	0%	
0-2	27%		15	64%	36%	0%	
0-2	30%		28	60%	39%	1%	H.Fuel
0-2	29%		25	66%	34%	1%	H.Fuel
Unoiled							
0-2	24%	DONR	0	90%	10%	0%	Tr.H.Fuel
0-2	20%		0	94%	5%	1%	--
0-2	19%		0	94%	6%	0%	--
0-2	18%		41	94%	6%	0%	H.Fuel
0-2	27%	PALW	0	77%	23%	0%	Tr.Fuel
0-2	23%		0	86%	14%	0%	Tr.Fuel
0-2	24%		0	82%	18%	0%	
0-2	24%		0	89%	11%	0%	Tr.Fuel
Reef Flat Sediments							
Moderately oiled							
0-2	43%	GALT	18	10%	86%	4%	
0-2	34%	(Seagrass bed	2	55%	45%	1%	H.Fuel
0-2	55%	on reef flat)	116	51%	47%	2%	VMIC
0-2	42%		52	57%	43%	0%	VMIC
0-2	41%		94	19%	80%	1%	VMIC
0-2	33%		60	66%	34%	0%	VMIC

1. Composition of oils based on UVF coupled excitation/emission spectra. "W." means weathered and missing portion of spectrum due to soluble light aromatics. VMIC = Venezuelan/Mexican Isthmian Crude, -- = flat trace, L. Fuel or H. Fuel refers to UVF from predominantly 1-2 and 1-3, and 4-ringed aromatics respectively.

"HC" = samples collected by Dr. H. Caffey in June or July 1986.

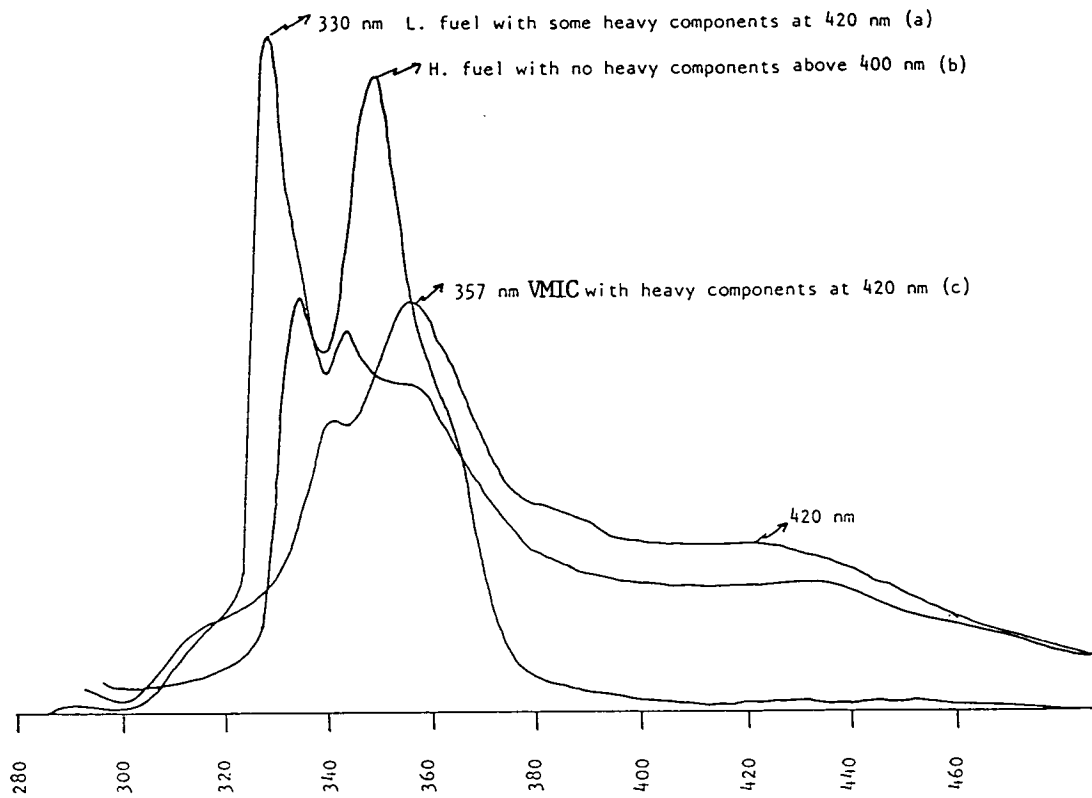
2. Oil content quantified by comparison with standard response graph made from dilutions of the weathered oil extracted from SBS mangrove sediment.

Mangrove Sediments

The data show that the control site at HRB (Figure 9.1) contained trace residues of oil, perhaps from earlier spills ($< 200 \mu\text{g/g}$). MARS and NARM were lightly oiled with VMIC. MARS surface sediments, sampled in July 1986, showed a high level of fuel oil (Figure 9.4), which was still visible in the deeper core sediments sampled in September. The surface of the core from this site also contained VMIC. This site was classified as moderately oiled for the July 1986 collection by its concentration of light fuel oil ($1,000 \mu\text{g/g}$). The SBS site was heavily oiled, with highest concentrations in the mid-berm area, and the LRM site was heavily oiled from the seaward front through the mid-grove high berm. (The berm is the local topographical high line due to accumulation of sediment and debris at the usual high tide mark). It was obvious by visual inspection that oil distribution was extremely patchy. This is reflected in the replicate surface sediment samples. The coefficient of variation, defined as the standard deviation divided by the average times 100, ranged from 14% to 56% within stations. To illustrate the extreme patchiness, three samples from the high berm area of LRM were taken from patches of oil visually ranked as shown in Table 9.4. The coefficient of variation in these three samples was 122% with oil levels ranging from 2 to 66% of the sediment dry weight. There is some disparity between the visual assessments of oiling severity and the amount of oil in sediments determined by chemical analysis in the areas initially classified as unoiled to lightly oiled. These samples, collected five months after the spill, demonstrate that oil had been transported in some cases into previously unoiled areas (e.g., MARM). Such movement is consistent with subsequent observations of slicks emanating from heavily oiled mangroves during storm events and redistribution of oil along the coast.

Seagrass and Coral Reef Sediments

Six seagrass beds were sampled in September 1986 where biological assessment studies were underway (LREN, LRS, NARC, LRLC, PALN and DONT; Figure 9.1). Samples from MARG were collected in June 1986 and stored in the freezer. The data identify heavily oiled ($> 1,000 \mu\text{g/g}$), moderately oiled (100 to $500 \mu\text{g/g}$) and lightly to unoiled ($< 100 \mu\text{g/g}$) areas (Table 9.4). One sample from the unoiled control sites showed the presence of tar balls in relatively pristine areas. This is expected on the Panama coast from ship traffic and previous contamination incidents. The UVF spectra for residues of the crude oil spilled in 1986 at Bahía Las Minas were distinguishable from highly weathered fuel residues in tar balls (Figure 9.4). This distinction may become less clear as weathering processes further change the composition of the VMIC. LREN sediments, which were composed of fine silty sand, had much higher levels of oil than areas with coarser sands, such as LRS, despite the extremely high levels of oil in the mangroves fronting both of these seagrass beds. Oil levels dropped with distance from shore (LRN2 compared to



- (a). Sample No. 83. Mangrove sediment. MARS.
(b). Sample No. 87. Reef sediment. DONR.
(c). Sample No. 85. Seagrass Sediment. MARG.

Figure 9.4 Examples of synchronous excitation/emission UVF spectra.

LRN3, and LREN compared to LRS). In general, chemical analysis agreed with visual observations, but some disparity is evident in the areas classified as lightly and moderately oiled.

The samples along the shore of Isla Largo Remo (LRN2) were taken in about 0.5 m of water at about 10 m intervals. Visual observations showed that in some areas where the tide was eroding the surface sand, layers of oiled sand were visible underneath. Thus, even in areas where the surface sands appeared to be unoiled, layers of oil persisted at varying depths depending on erosion and burial rates. It was not possible to take compacted cores for depth distribution in these dynamic environments. These erosion and redistribution processes account for some of the variability in the chemical measurements.

On the basis of sediment chemistry data, corroborated by coral mortality and the oil content in coral tissues, the PAYN and GALC reefs were ranked as heavily oiled, LRE as moderately oiled, NARS as lightly oiled and DONR and PALW as unoiled (Table 9.4). Unoiled reefs showed traces of highly weathered fuel oils visible in the UVF spectra. As with the seagrass sediments, the variability between samples may be a result of sediment redistribution processes.

9.2.4 Correlation of UVF and GC Oil Analyses

Thirty-six sediment samples consisting of surface and depth cores from mangrove areas and surface sediments from seagrass beds and coral reefs were Soxhlet extracted and subjected to complete analysis by UVF and GC. Oil content was estimated by two methods: by the UVF method in units of D.VMIC and by the unresolved (URE) signal of the saturated and light aromatic hydrocarbon fractions by GC. Figure 9.5 shows the graphs of the log of the average concentration of petroleum hydrocarbons determined by UVF plotted against the unresolved components in saturated and aromatic hydrocarbon fractions determined by gas chromatography. Four samples were processed through the high performance liquid chromatography (HPLC) class separation procedure, as developed in 1988, before the computer system on the HPLC failed. In order to complete the analyses on schedule, the separations were continued using traditional column chromatography separations on silica/alumina columns. This procedure is adequate to clean up samples for GC analysis, but separations are not consistent enough to be used for estimating the concentrations of classes of aromatic hydrocarbons and oxygenated reaction products as can now be done by HPLC/UVF. The GC analyses showed that the oil in all samples, including mangrove cores, was highly degraded (Figures 9.2, 9.3, 9.6, and 9.7). Most alkanes were missing, leaving primarily an unresolved signal. This was quantified by use of an average response factor over the appropriate elution range on the GC. The data provide a conservative estimate of the amount of residual oil in samples (Table 9.5).

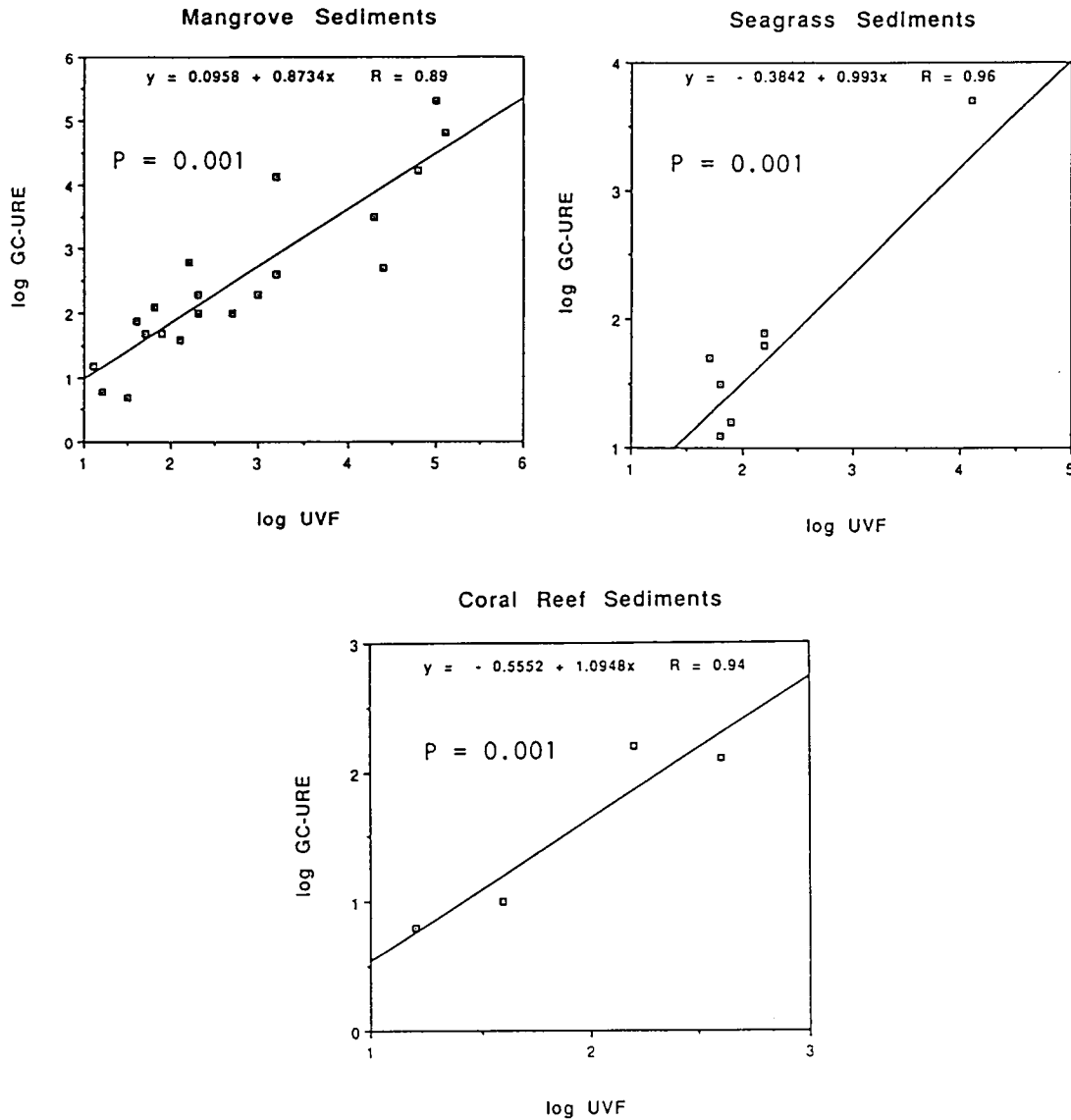


Figure 9.5 Correlations between the concentrations of petroleum hydrocarbons determined by ultraviolet fluorescence spectroscopy and by gas chromatography graphed as log ($\mu\text{g/g}$ dry wt).

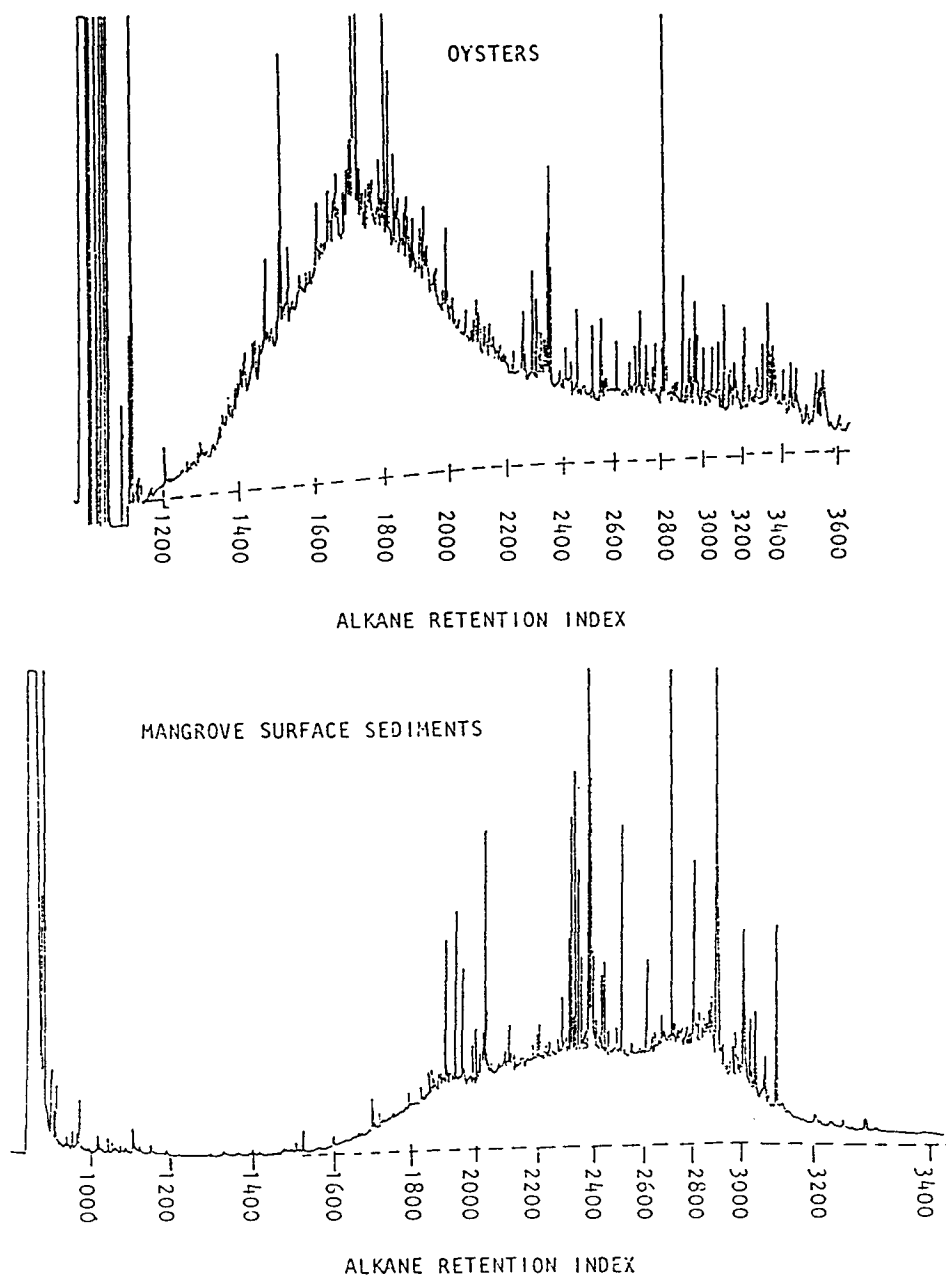


Figure 9.6 Saturated hydrocarbon gas chromatograph traces at mangrove site MARM.

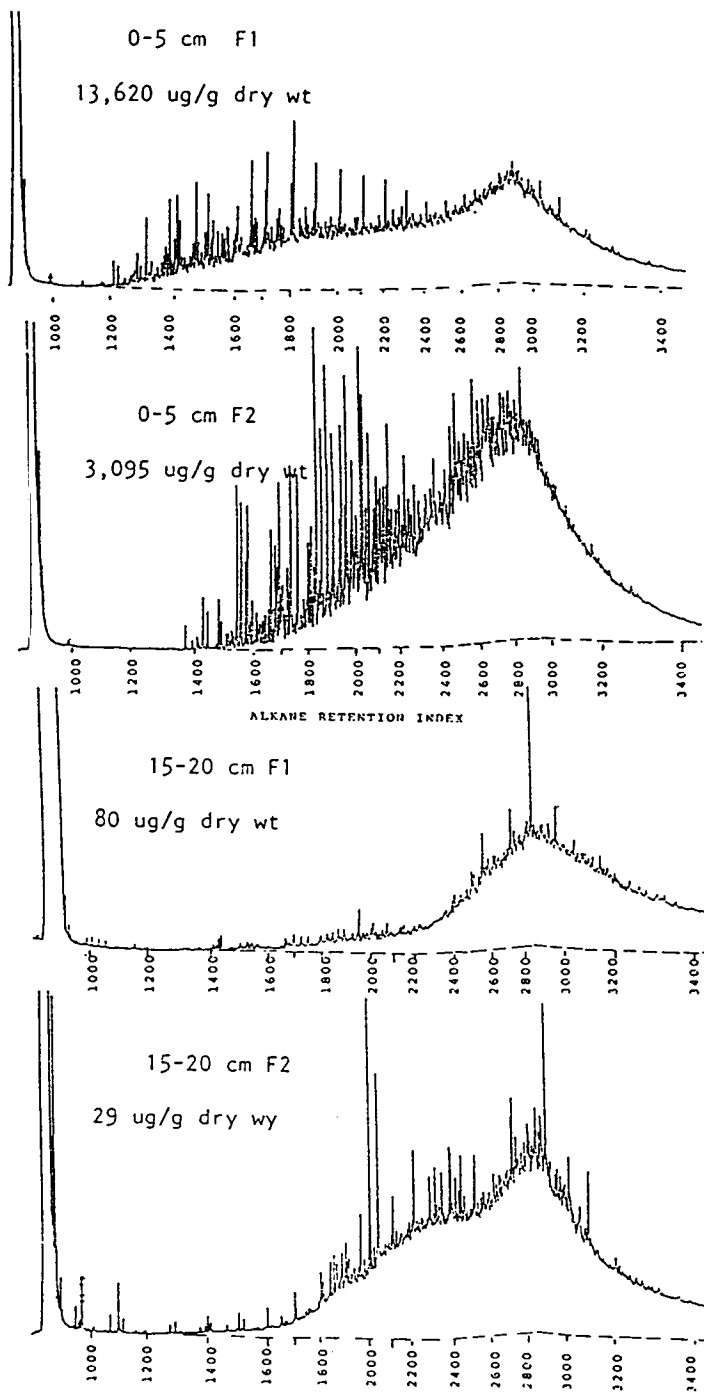


Figure 9.7 Gas chromatograms of F1 saturated and light aromatic hydrocarbons and F2 aromatic hydrocarbons in mangrove sediment cores. Station LRM, September 1986.

Table 9.5 Gas chromatographic analysis of sediment samples collected in 1986 and ratios indicative of stages of weathering.

Depth (cm)	% Water	Area Sampled	$\mu\text{g/g dry wt.}$		Phytane/ URE phyt.	URE C28/ URE C19	GC Evaluation
			F1	F2/F3			
Mangrove sediments							
Heavily oiled							
0-5	85%	LRM Core (seaward front)	439	30	0.7	1.6	D.VMIC
15-20	79%	LRM Core	127	81	0.1	9.7	D.VMIC
0-5	81%	LRM Core (mid-grove)	13,620	3,095	2.2	1.5	D.VMIC
10-15	84%	LRM Core	80	29	0.7	4.6	D.VMIC
0-5	74%	SBS Core (seaward front)	164,557	15,783	2.8	1.3	D.VMIC
15-20	79%	SBS Core	150	63	0.7	0.8	D.VMIC
0-5	81%	SBS Core (mid-grove)	48,611	21,034	3.1	0.7	D.VMIC
15-20	82%	SBS Core	2	3	0.9	0.9	Tr.
0-5	84%	SBS Core (inland grove)	2,109	974	0.9	1.2	D.VMIC
Moderately oiled							
0-5	87%	MARS (7/86 HC)	279	102	0.1	1.6	D.Fuel/Crude
0-5	81%	MARM Core (seaward of boom)	504	161	0.2	1.0	D.Fuel/Crude
15-20	79%	MARM Core	11,159	558	0.2	0.3	D.Fuel/Crude
Lightly oiled							
0-5	47%	NARM Core (small island)	87	13	0.3	2.1	D.VMIC
15-20	67%	NARM Core	10	5	1.0	3.5	Tr.
0-5	79%	MARS Core (inside boom)	104	31	0.1	1.5	Biog
15-20	73%	MARS Core	5	2	0.5	2.0	Biog
Unoiled							
0-5	88%	HRB Core (seaward front)	31	13	1.0	3.7	Biog
0-5	86%	HRB Core (mid-grove)	22	30	0.4	1.8	Biog
15-20	84%	HRB Core	40	35	1.0	2.6	Biog
0-5	87%	HRB Core (inland grove)	32	14	4.0	5.3	Biog

Table 9.5 Gas chromatographic analysis of sediment samples collected in 1986 and ratios indicative of stages of weathering (continued).

Depth (cm)	% Water	Area Sampled	$\mu\text{g/g dry wt.}$		Phytane/ URE phyt.	URE C28/ URE C19	GC Evaluation
			F1	F2/F3			
Seagrass sediments							
Heavily oiled							
0-2	68%	LREN	2,403	3,158	0.5	1.7	D.VMIC
Moderately oiled							
0-2	40%	LRS	74	14	0.9	1.7	D.VMIC
0-2	46%	NARC	53	14	0.7	2.6	D.VMIC
0-2	41%	NARC	49	19	0.7	1.5	D.VMIC
0-2	52%	NARC	28	19	0.4	1.7	D.VMIC
0-2	47%	MARG (6/86)	12	5	1.0	1.4	D.VMIC
0-2	48%	MARG (6/86)	28	6	1.0	2.7	D.VMIC
Lightly oiled							
0-2	30%	LRLC	6	8	0.6	1.7	D.VMIC
Unoiled							
0-2	37%	PALN	0	0	--	--	N.D.
0-2	39%	DONT	0	0	--	--	N.D.
Coral reef sediments							
Heavily oiled							
0-2	39%	PAYN	110	53	1.1	0.8	D.VMIC
0-2	52%	GALC	85	48	0.8	1.2	D.VMIC
Moderately to lightly oiled							
0-2	47%	LRE	9	1	0.5	0.8	D.VMIC
0-2	29%	NARS	4	2	4.3	2.5	Tr.
Unoiled							
0-2	24%	DONR	N.D.	N.D.	N.D.	--	N.D.
0-2	27%	PALW	1	N.D.	Biog	--	N.D.

"URE" is the unresolved mixture of saturated and light aromatic hydrocarbons quantified by means of an average response factor over the appropriate elution range on the GC. D.VMIC is degraded Venezuelan/Mexican Isthmian Crude oil, "Tr" is trace amount, "N.D." is not detectable. "Biog." is biogenic interference in the ratio or a predominance of biogenic hydrocarbons in the GC evaluation.

"F1" are the saturated and light aromatic hydrocarbons, F2/3 are the aromatic hydrocarbon fractions from separation on silica/alumina columns.

Samples collected September 1986 except MARS and MARG, which were collected June or July 1986. Mangrove samples were collected with hand coring device. Seagrass and reef samples were collected with jars used as scoops.

The ratios "Phytane/URE phyt." and "URE C28/URE C19" are meant to provide an estimate of the relative stage of degradation of the oil residues, as explained in the text.

Analysis of the results generated by these two methods for estimating oil content showed a highly significant correlation for all sediment samples (mangroves, seagrass and coral sediments) spanning six orders of magnitude with a slope nearly equal to one (Figure 9.5). Replicates for UVF analyses ranged from 1 to 4 determinations as listed in Table 9.4 and include 0 - 5 cm core sections and surface scrapes. The GC analyses were single samples from Table 9.5. The significance of the relationship remained despite the different sampling methods for mangrove, seagrass and coral reef sediments. Thus, the relative ranking of site contamination can be done on the basis of either analysis.

As would be expected, the chemical data generally corroborated the visual estimates of the severity of oiling. Since some areas are flushed more than others, the residual levels in light and moderately oiled areas may cause them to be modified from their initial classifications. Concentrations span several orders of magnitude and because dose/response relationships generally follow a loglinear relationship (Widdows *et al.* 1990), it should be possible to establish correlations for both lethal and sublethal biological effects with the chemical data. This will be the primary focus of the chemical distribution data in the context of the long-term assessment of effects of the spill on coastal ecosystems. To facilitate this interpretation, Table 9.6 lists the log of the mean of all replicate determinations of petroleum hydrocarbons at each station sampled during the 1986 collection by both UVF and GC methods.

9.2.5 Bioindicators

The case for using bivalves as indicator organisms for petroleum contamination in coastal waters was detailed by Burns and Smith (1981). Oysters were collected from three sites in the 1986 sampling effort. No living oysters or other obvious bivalves could be found in heavily oiled mangrove areas despite searching by the field collection crew. Oysters were collected from mangrove roots at the HRB unoiled reference site, and MARM moderately oiled and MARS lightly oiled sites (Figure 9.1). The results of UVF and GC analysis of these samples are shown in Table 9.7. Bivalves preferentially accumulate the more soluble, lower molecular weight hydrocarbons in an oil contaminated ecosystem. A comparison of the GC and UVF patterns between the oysters and mangrove sediments is given in Figures 9.6 and 9.8. GC quantification is accurate despite the different elution ranges. However, the UVF analyses of the oyster samples would be more quantitative if the fluorescence intensity were measured at the wavelengths optimum for diesel fuel (280/325 nm) rather than the crude oil (310/360 nm). This phenomenon has been further discussed in Burns *et al.* (1990). Uptake is related to the lipid content of the organisms in addition to the amount of oil in surrounding waters. Thus, expression of results as oil content per mg lipid provides an effective comparison between sites.

Table 9.6 Summary of concentrations of petroleum hydrocarbons in sediments collected in 1986 based on UVF and GC analysis. Values expressed as log of average concentrations in $\mu\text{g/g}$ dry weight. Composition was determined by UVF and GC analysis, and the initial ranking of oiling severity was made by visual observation.

Depth cm	Area Sampled	log ($\mu\text{g/g}$ dry wt.)		Composition (UVF & GC)	Visual Assess.
		UVF	GC		
Mangrove sediments					
Heavily oiled					
0-5	LRM seaward front	4.4	2.6	D.VMIC	Heavy
15-20	LRM	3.0	2.1	D.VMIC	
0-5	LRM mid-grove	4.8	4.1	D.VMIC	Heavy
10-15	LRM	2.7	1.9	D.VMIC	
0-5	LRM high berm	5.2		D.VMIC	Heavy
0-5	SBS seaward front	5.0	5.2	D.VMIC	Heavy
15-20	SBS	2.3	2.2	D.VMIC	
0-5	SBS mid-grove	5.1	4.7	D.VMIC	Heavy
15-20	SBS	1.5	0.3	Tr.VMIC	
0-5	SBS inland grove	4.3	3.3	D.VMIC	Heavy
Moderately oiled					
0-5	NARM small island	2.3	1.9	D.VMIC	Light
15-20	NARM	1.1	1.0	Tr.VMIC	
0-5	MARS 7/86	3.2	2.4	Mix	None
0-5	MARM seaward of boom	2.2	2.7	Mix	
15-20	MARM	3.2	4.0	Mix	
Lightly oiled					
0-5	MARS inside boom	1.8'	2.0	Tr.VMIC	None
15-20	MARS	1.2	0.7	Biog	
Unoiled					
0-5	HRB seaward front	2.1	1.5	Tr.Mix	None
0-5	HRB mid-grove	1.7	1.3	Tr.Mix	None
15-20	HRB	1.6	1.6	Tr.Mix	
0-5	HRB inland grove	1.9	1.5	Tr.Mix	None
Reef Flat sediments					
Moderately oiled					
0-2	GALT	1.8		D.VMIC	Heavy

Table 9.6 Summary concentrations of petroleum hydrocarbons in sediments collected in 1986 based on UVF and GC analysis (continued).

Depth cm	Area Sampled	log ($\mu\text{g/g}$ dry wt.)		Composition (UVF & GC)	Visual Assess.
		UVF	GC		
Seagrass sediments					
Heavily oiled					
0-2	LREN	4.1	3.4	D.VMIC	Heavy
Moderately oiled					
0-2	LRS	2.2	1.9	D.VMIC	Heavy
0-2	NARC	2.2	1.7	D.VMIC	Moderate
		2.2	1.7		plus
		1.7	1.4		Dispersants
0-2	MARG 6/86	1.9	1.1	D.VMIC	Light
0-2	MARG 6/86	1.8	1.4	D.VMIC	Light
0-2	LRN2	1.9		D.VMIC	Moderate
Lightly oiled					
0-2	LRLC	0.8	0.8	D.VMIC	Light
0-2	LRN3	0.7		Biog	Light
Unoiled					
0-2	PALN	N.D.*	N.D.	Tr.H.Fuel	None
0-2	DONT	N.D.	N.D.	Tr.H.Fuel	None
Coral reef sediments					
Heavily oiled					
0-2	PAYN	2.2	2.0	D.VMIC	Heavy
0-2	GALC	2.6	1.9	D.VMIC	Heavy
Moderately oiled					
0-2	LRE	1.6	0.9	D.VMIC	Heavy
Lightly oiled					
0-2	NARS	1.2	0.6	Tr.Mix	Light
Unoiled					
0-2	DONR	N.D.	N.D.	Tr.H.Fuel	None
0-2	PALW	N.D.	0.0	Biog	None

Summary data taken from Tables 9.1 and 9.2. Concentrations of oil listed as the log of the average oil concentration as determined by UVF and GC analysis of surface samples and depth cores.

*: only 1 out of 4 replicates showed any oil contamination.

"D.VMIC" is degraded Venezuelan/Mexican Isthmian Crude, "Mix" is a mixture of light and heavy crude, "Biog" is biogenic, "Tr" is trace amount, "N.D." is not detectable.

Table 9.7 Lipid weights and petroleum hydrocarbons in oysters. September 1986 collection.

Sample Station	F1 URE - GC		F2 UVF	mg lipid/ g dry wt	% NSL
	$\mu\text{g/g}$ dry wt	$\mu\text{g/mg}$ lipid	$\mu\text{g/g}$ dry wt		
P2-27 HRB	49	1.1	20	44	15%
P2-37 MARS	309	6.1	61	50	13%
P2-78 MARM	157	3.8	59	42	16%

Dry weight estimated as 15% of wet weight. Wet tissue extracted by hydrolysis procedure. "% NSL" is the percent of lipid in the non-saponifiable fraction. Petroleum content estimated as the unresolved GC signal (URE) of the saturated hydrocarbon fractions and the fluorescent intensity (UVF) of the aromatic fractions.

The maximum limit of accumulation in bivalves is generally up to approximately 30 μg oil/mg lipid. The Bahía Las Minas samples ranged from 1.1 $\mu\text{g/mg}$ lipid at the unoiled site to 3.8 and 6.1 $\mu\text{g/mg}$ at the sites classified as moderately and lightly oiled, respectively, on the basis of oil in surface sediments determined by GC and UVF analysis. These preliminary data confirm that the bivalves will be useful in establishing dose/response relationships for sublethal biological effects. However, the GC and UVF patterns must be interpreted in view of the preferential bioaccumulation of low molecular weight (LMW) compounds. These are the most soluble hydrocarbon fractions and the bivalves accumulate them from oil suspended in the water column. As these are also the most acutely toxic fractions of oil, the dose/response relationship should be further strengthened. However, it may be expected that doses estimated from bivalves may not exactly correspond with those estimated from sediments. Rather, they provide a time-integrated estimate of the degree of contamination in the surrounding water column.

As discussed above (Chapter 9.2.2), corals also take up hydrocarbons from surrounding waters. The analyses of the 1986 samples did not show the extreme bias toward LMW hydrocarbons in either the GC or UVF analysis as noted for the bivalves (Burns and Knap 1989). Rather, the pattern of oil in the corals resembled the total suspended fraction of hydrocarbons in surrounding waters. An experiment was conducted to determine the efficiency of the pressured-air method used to remove coral tissue from the skeletal matrix. The species of *Agaricia* used forms thin platelike colonies that are easily crushed. After the surface tissue was removed with the air method, the remaining skeleton was ground with Na_2SO_4 and extracted in a Soxhlet apparatus. The results are shown in Table 9.8 for unoiled and oiled corals. They indicate that the air method left about 90% of the lipid inside the skeletal matrix. However, oil concentrations were higher in the easily removed tissue, making it possible to rank levels of exposure.

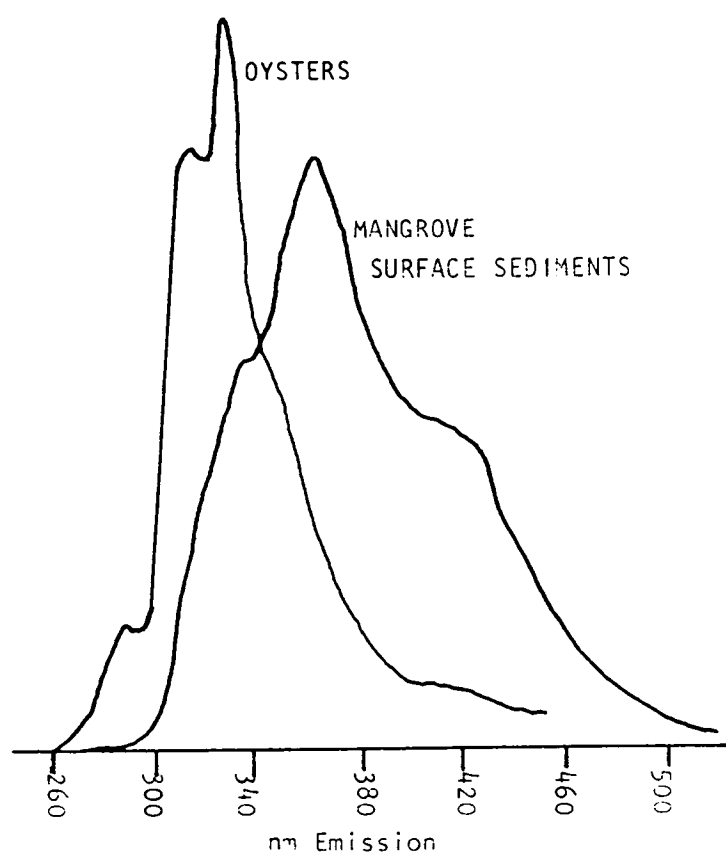


Figure 9.8 Synchronous excitation/emission fluorescence scans of aromatic hydrocarbons extracted from oysters and mangrove sediment at station MARM.

Table 9.8 Experiment to determine efficiency of air method in removing tissue from corals.

Sample	Total mg lipid	Total μg F1 URE	$\mu\text{g}/\text{mg}$	Total μg F2 UVF	$\mu\text{g}/\text{mg}$
PALW <i>Agaricia tenuifolia</i> :					
surface tissue	190	57	0.3	19	<0.1
deep tissue	1,849	68	<<0.1	12	<<0.1
PAYN <i>Agaricia tenuifolia</i> :					
surface tissue	118	979	8.3	5,865	50
deep tissue	1,091	6,343	5.8	5,491	5

Surface tissue was blown off coral skeleton with air pressure. Deep tissue extract was obtained by grinding remaining skeleton with Na_2SO_4 followed by hydrolysis extraction of the salt/tissue paste.

Because of this finding, the tissue preparation procedure was modified during the processing of the 1988/89 collection and tissue removal was aided by mechanical scraping.

9.2.6 Weathering Processes

Gas chromatograms of all the sediment samples showed significant alteration of the oil had occurred within five months after the spill, even in heavily oiled mangroves. Figure 9.3 shows the difference in saturated hydrocarbons of VMIC contrasted with spilled VMIC extracted from seawater near oiled mangroves. Most hydrocarbons lighter than C_{18} were missing. The resolved peaks in the oil due to n-alkanes and branched alkanes were degraded to the point that ratios of peaks are not useful as measures of the stage of degradation. The residual pattern emphasizes the unresolved signal in the C_{18} through C_{34} elution range. This pattern is similar to that seen in oiled seagrass and coral reef sediments. Some of the mangrove sediments showed high levels of less degraded oil. In assessing the GC and UVF patterns from the 1986 samples, strategies for interpreting the geochemical degradation patterns and for the integration of the chemical data with the biological studies must be further developed to incorporate the rapid rate of degradation as seen in this initial data set.

Evaporation/solution of Light Aromatic Hydrocarbons

The light aromatic and aliphatic hydrocarbons are generally the most acutely toxic fractions of oil. The HPLC separation procedure was calibrated to separate the aliphatic hydrocarbon plus benzene and naphthalene fraction (F1) from the heavier aromatics (F2 & F3). Five of the extracts from mangrove sediments from the 1986 collection and the Venezuelan/Mexican Isthmian crude oil have been subjected to class separation and estimation of the amount of fluorescence in each fraction. Table 9.9 expresses the data as percent of the total recovered UVF signal in each fraction, measured at 310/360 nm for excitation/emission (ex/em), respectively. As will be further discussed below, the degree of weathering can be related to a relative decrease in F1 UVF due to evaporation and solution and an increase in UVF intensity in the oxygenated reaction product fractions (F4 & F5). Because the spectral pattern within each fraction differs from the original oil, the UVF estimate is only semi-quantitative. Current efforts are being directed at developing a UVF protocol that will utilize calibration standards and ex/em wavelengths relevant to each fraction. In this way, the content of oxygenated reaction products will be more accurately estimated. A sample of 1-pyrenol has been obtained from the National Cancer Institute Standards Repository and will be used as a calibration standard for F4 and F5 fractions.

In addition to the semi-quantitative UVF estimate, selected extracts were analyzed for individual aromatics by GC/MS. Calibration of the instrument for LMW aromatics and selected ion scans for aromatic classes in the reference oils and selected environmental samples is underway. Data on the linearity of response factors are presented in Chapter 9.4. Table 9.10 shows the content of the light aromatic hydrocarbons in the Venezuelan/Mexican Isthmian crude oil and in Sunniland Formation oil. None of the volatile components was detectable in extracts of the sediment samples collected five months after the spill. Note that benzene and toluene could not be accurately quantified in this GC/MS procedure because it eluted with the hexane solvent front. A GC/MS protocol for determination of parent and alkyl polynuclear aromatic hydrocarbons has been developed and individual component data will be available. Characterization of the Sunniland oil is being conducted to provide initial prediction of the impact of a spill of this oil on similar tropical ecosystems.

Table 9.9 Percentage of fluorescent material in HPLC fractions of extracts of VMIC oil and mangrove sediments separated by HPLC. Sample numbers are given above each site; depth of samples in cores are given below each site.

Fraction	VMIC	P2-77 SBS (0-5 cm)	P2-85 LRM (0-5 cm)	P2-45 LRM (0-5 cm)	P2-46 LRM (15-20 cm)	P2-48 LRM (15-20 cm)
F1	11	18	12	20	9	9
F2	59	51	62	55	69	65
F3	28	25	23	19	16	21
F4	0	4	2	5	3	3
F5	1	2	1	2	2	1

F1 contains benzenes through naphthalenes, F2 contains phenanthrenes through benzopyrenes, F3 contains more polar aromatic derivatives, F4 and F5 contain the oxygenated derivatives (aromatic ketones, aldehydes and quinones). All determinations made using 310 nm excitation/360 nm emission using D.VMIC to construct the standard response curve. Note text explanation that these wavelengths result in underestimations in F4 and F5.

Table 9.10 Content of volatile aromatics in the ethyl benzene through decyl benzene elution range in Venezuelan/Mexican Isthmian Crude Oil and Sunniland Formation Crude Oil for comparison. None of these volatile benzene derivatives could be detected in sediment core samples containing VMIC collected in September 1986.

Compound	VMIC Oil µg/mg oil	Sunniland Oil µg/mg oil
benzene*		
toluene*		
ethyl benzene	0.9	1.2
m,p-xylene	2.2	1.5
o-xylene	1.1	1.1
isopropyl benzene	0.1	0.2
n-propyl benzene	0.2	0.4
mesitylene	0.4	0.2
p-cymene	0.9	1.2
n-butyl benzene	0.4	0.2
n-hexyl benzene	n.d.	n.d.
n-octyl benzene	n.d.	n.d.
n-decyl benzene	n.d.	n.d.

* Benzene and toluene could not be quantified due to coelution with the hexane used as the solvent for injection.

Microbial and Photochemical Degradation

Figure 9.7 shows the chromatograms of both saturated and aromatic hydrocarbon fractions from heavily oiled mangrove sediments. The top traces are from the 0-5 cm surface core from LRM mid-grove and the bottom traces are from the corresponding 15-20 cm depth core. The near-surface oil is modified from the VMIC, but is relatively less degraded than the residues of oil that had been transported to depth. The resolved alkanes in F1 and aromatics in F2 are missing from the deeper core. This pattern was repeated in all of the other depth series cores from mangroves. That is, with the exception of the high energy seaward front cores at station LRM, very little oil had been transported to deep sediment layers even in heavily oiled mangroves five months after the spill. The oil that was transported was relatively degraded compared to the oil in the surface cores, implying that degradation processes proceeded at depth. Because so many of the samples had no visible alkanes, traditional indices of degradation based on n-alkane to isoprenoid ratios are of limited use in this data set. Rather, ratios of isoprenoids to URE at the same retention index (Phytane/URE) and the ratio of URE at C₂₈ compared to URE at C₁₉ are more appropriate (Table 9.5). These ratios and the presence or absence of alkanes with carbon preference index close to one represent an indication of the relative stage of microbial degradation.

Changes in the UVF spectra also correlate with stage of degradation. Figure 9.9 shows the UVF emission and synchronous scans of the total lipid fraction from the same core sediments. Note the change in ratio of relative spectral maxima at approximately 370 and 410 nm. This change is further illustrated in Figure 9.10, which shows the emission spectra of the original oil in contrast to that in several mangrove sediment cores. The shift to high wavelengths could be due either to the removal of smaller ringed aromatics due to solution processes or to the relative increase in compounds with fluorescence maxima at higher wavelengths due to oxidation processes. Table 9.9 and Figures 9.9, 9.10, and 9.11 show this is a result of both processes. As weathering progresses, there is a decrease in relative fluorescent intensity in the light aromatics in F1 and F2 (solution) and an increase in F4 and F5 due to the formation of oxygenated reaction products of the aromatic hydrocarbons, with fluorescence intensity maxima at higher wavelengths. Note that the estimates for concentrations of these products listed in Table 9.9 were made on the basis of 310 ex/360 em nm. Thus, the concentrations of the oxidation products were underestimated. With the use of 1-pyrenol as a calibration standard, these estimates will be improved in future analyses.

Based on these observations of compositional changes due to weathering and degradation processes, the list of criteria for the relative stages emerges as in Table 9.11. Only the data for samples separated by the HPLC procedure are tabulated. These criteria will be expanded to include the ratios of parent and alkyl polynuclear aromatic hydrocarbons (PAHs) determined by GC/MS as these analyses are completed.

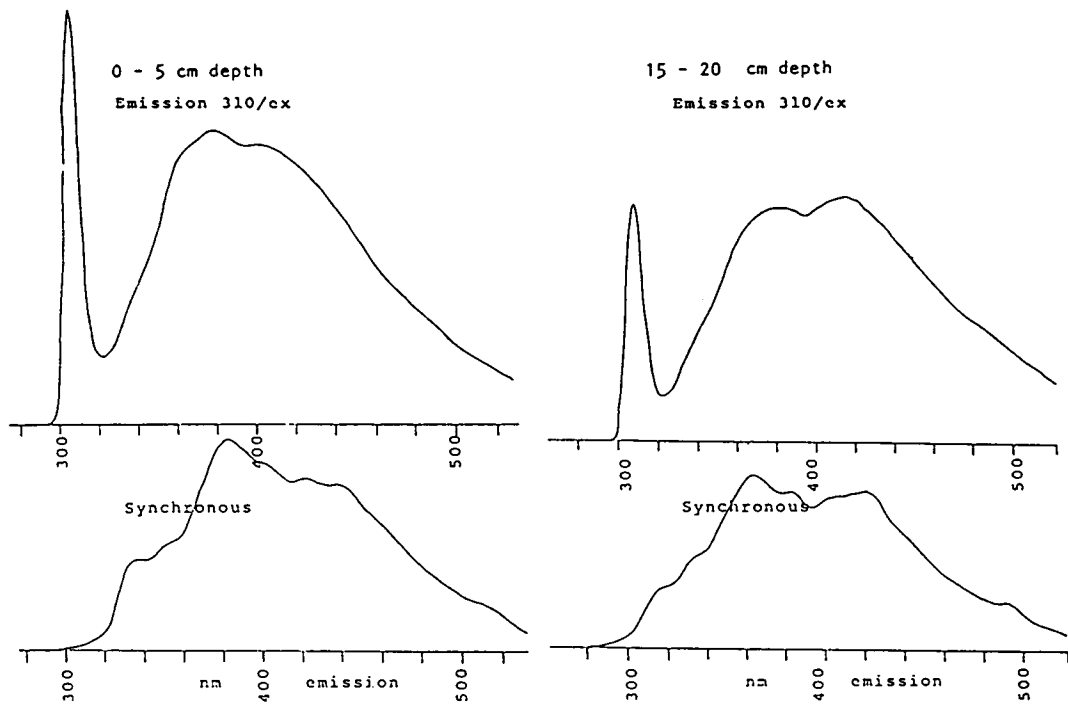


Figure 9.9 Emission and synchronous excitation/emission spectra of aromatic hydrocarbons extracted from mangrove sediment cores. Station LRM, September 1986.

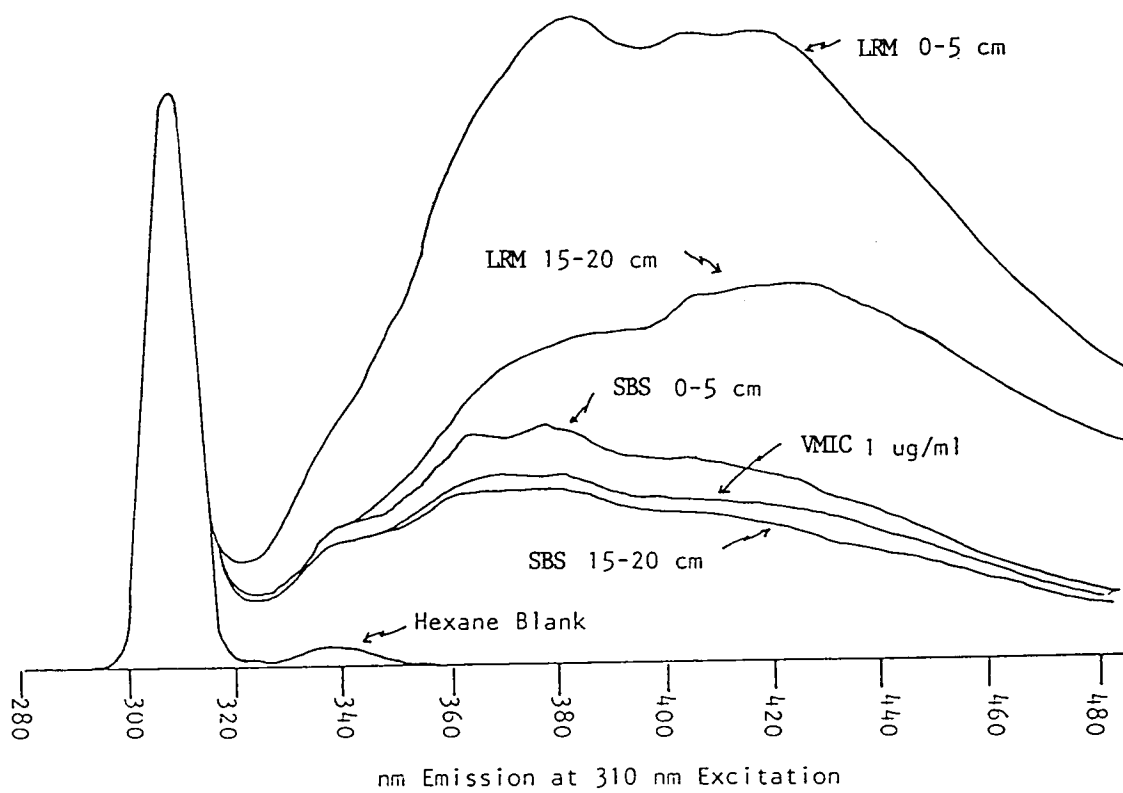


Figure 9.10 Emission fluorescence spectra of solvent extracts from mangrove sediments and the reference crude oil (VMIC).

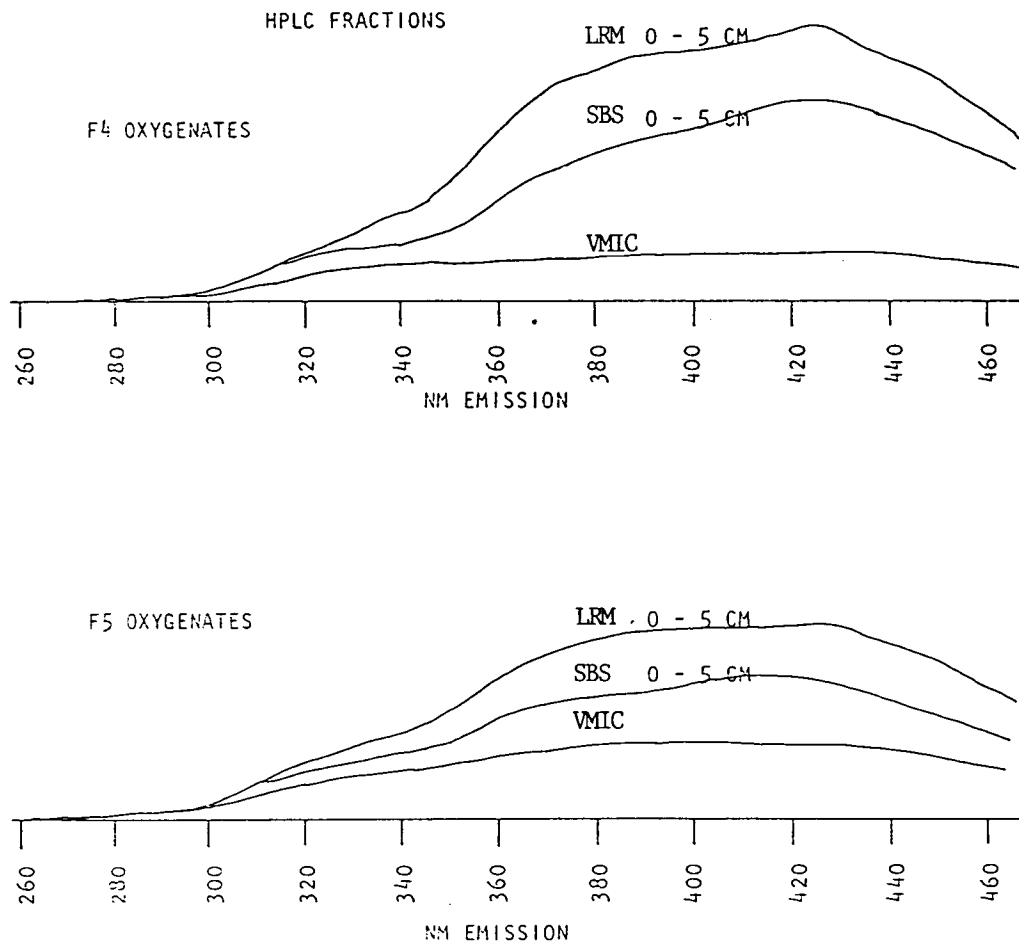


Figure 9.11 Synchronous UVF spectra of VMIC and extracts of mangrove sediments.

Table 9.11 Summary of criteria for estimating stage of weathering of oil residues in sediment samples based on UVF and GC analysis. See Table 9.9 for the sites corresponding to the sample numbers.

Sample	VMIC	P2-77	P2-85	P2-48	P2-46	P2-45
µg URE/g dry	p=0.87	164,557	13,620	80	439	127
alkane peaks ¹	++++	+++	++	+	-	-
aromatic peaks ²	+++	++	++	-	-	-
phyt/URE	3.6	2.8	2.2	0.7	1.0	0.1
URE C ₂₈ /C ₁₉	1.1	1.3	1.5	4.6	2.2	9.7
UVF 410/370 em	0.9	0.9	1.0	1.1	1.1	1.3
410/370 syn	0.7	0.7	0.8	0.9	0.9	1.0
% UVF F4 & F5	1.1	5.8	2.6	4.8	4.4	6.0

¹++++: all oil-derived n-alkanes from C₁₀ through C₃₄ with CPI=1 clearly visible over the URE. Ratios of n-alkanes to corresponding isoprenoids (C₁₇/pristane, C₁₈/phytane) range from 3.0 to 4.0; +++: depletion of volatiles ≤ C₁₄, but peaks still clearly visible above the URE. Ratios are from 1.5 to 3.0; ++: suite of alkanes still visible above the URE, but branched molecules more abundant than n-alkanes. Ratios are ≤ 1.0; +: pattern of alkanes highly modified and only barely visible above the URE; -: all oil-derived n-alkanes degraded and not visible above the URE.

²++++: individual aromatic hydrocarbons such as methyl phenanthrenes clearly visible by FID above the URE. Ratios of m-phenanthrenes/URE range from 5.0 to 7.0; ++: Ratios are approximately 4.0 to 6.0; -: pattern degraded sufficiently that individual PAHs cannot be quantified by FID alone.

Penetration of Oil to Depth in Mangrove Sediments

Despite extremely high levels of oil in surface sediments of mangroves at LRM and SBS, the VMIC did not show rapid transport to deep mangrove sediments except in the seaward front areas subject to rapid tidal flushing. Not only did the cores show a dramatic difference between 0-5 cm and 15-20 cm depths, but also the 0-5 cm cores contrasted with the 0-2 cm surface scrapes show that most of the oil was contained in a discrete layer near the surface layers of sediment (Table 9.4). This consistent lack of high levels of oil in deeper mangrove core sections proves the efficiency of the coring procedure in obtaining accurate sediment profiles.

The areas heavily contaminated with VMIC can be contrasted with sediments at MARM, which were also contaminated with a degraded fuel oil. At this site, more oil was present at depth than at the surface. This may have been the result of a previous contamination incident in the area. The extremely high levels of oil in surface sediments in the oiled mangroves and the relatively less degraded state of the oil in the most heavily contaminated areas indicate that mangrove sediments will be a long-term source of oil contamination to coastal seagrass and coral reef ecosystems as sediments erode and are flushed by tidal action. However, this oil has been highly modified in composition compared to the spilled crude oil. Visual observations of cores collected in 1989 indicate much more transport of oil to depth along channels

created by rotting mangrove roots (Chapter 9.2.3). However, due to the patchy distribution of oil in the mangrove sediments, it will not be possible to construct a mass balance for the residual oil. The chemical data do provide an estimate of potential toxicity due to oil residues in the various habitats that should correlate with the biological studies.

9.2.7 Alternative Sources of Contamination

Assessment of the source of contamination is unquestionable in areas heavily contaminated by VMIC. GC and UVF patterns in environmental samples differ from the original oil in ways explainable by weathering processes. Particular scrutiny was given to samples taken near the site of the old (1968) *Witwater* wreck. Nearby mangroves, seagrass sediments and coral reef sediments were contaminated primarily with D.VMIC, although traces of degraded fuel oil were seen in coral reef sediments. The only area with significant contamination by another oil was MARM. This appeared to be an old spill of fuel oil. Levels were higher at depth than at the surface. The surface assemblage of animals and plants seemed to be similar to the MARS site based on cursory observation. At unoiled sites, the residual contamination by tar balls and fuel residues likely originating from ships is visible only by the highly sensitive UVF method. During the 1989 collection (Chapter 9.3), one site was tagged as having suffered a recent spill of fuel oil. This event will be evident in the oil residues in the sediments by GC and UVF analyses.

Additional information on sources is obtained by means of selected ion searching for stable marker compounds by GC/MS. BBSR has purchased relevant triterpane and sterane standards and initiated scanning of selected extracts for these compounds. Since the residues in the samples were already at an advanced stage of weathering five months after the spill, it is anticipated that this level of analysis may be required for positive identification of VMIC residues in later samples. It is also anticipated that it will be possible to follow stages of degradation by tabulating changes in the ratios of specific marker peaks caused by the selective degradation of some compounds and possible bacterial synthesis of others. Ratios of specific aromatics may also yield useful information. Therefore, both saturated and aromatic fractions of selected samples will be analyzed by GC/MS.

The data from the various analyses will be synthesized to construct a model for the rates of degradation and dispersion processes in this tropical environment compared to similar models derived in temperate regions.

Work has also been initiated to characterize the Sunniland crude oil from Florida in comparison with the Venezuelan/Mexican Isthmian crude in order to make predictions about the relative toxicity of the two crudes.

9.3 1988/89 Sample Collection

Collection

A second major sampling effort was initiated in December 1988. The tides at this time of year prevented the collection of mangrove sediments. Thus, the sampling effort was divided into two phases. In December 1988, the mangrove root organisms, seagrass sediments, reef flat urchins, and most of the coral reef samples were collected. The remaining coral samples were collected in April/May 1989 when the tides were more favorable for the collection of mangrove sediments. Cleaned jars were left at the laboratory for use by collaborating scientists in procuring samples from their special study sites (for example, stomatopods). Detailed field notes, including drawings of the sediment cores, have been archived. Quarterly collections of bivalves are frozen until transported to BBSR.

The inventory of samples collected through June 1989 is given in Appendix Table F.1. The appendix shows the BBSR accession numbers, a descriptive name that includes a code for the station sampled, a brief description of the site and the date collected.

Sample Processing

Bivalves were shucked into preweighed, organically clean glass jars, weighed and frozen.

Mangrove cores were taken at low tide with thin walled 8.5 cm diameter aluminum pipe cut to 45 cm lengths. If the substratum was soft, the corer was manually pushed into the sediment; if firm, a heavy steel cap was placed over the corer and the pipe was forced into the sediment with a large hammer. A machete cut to a straight edge was used to cut away surrounding root matter. The ends were capped with cork plugs and the cores placed in an ice chest to transport to the laboratory. Upon arrival at the laboratory, cores were extruded onto sheets of solvent-cleaned aluminum foil, wrapped, marked with an arrow indicating the top and frozen. For sectioning, the cores were partially defrosted. Sections at 0 - 2, 8 - 10, and 18 - 20 cm were cut and placed flat on a clean piece of foil. The outer edges of the core sections were cut away and only the inner sections placed in sample jars. The same depth sections from three replicate cores at each station were combined. The remaining sections of the core were cut in half length-wise and cross sectional drawings were made. From these drawings, numbers of dead and live roots were compiled (Table 9.12). Observations on the visible appearance of oil layers and the smell of oil were also recorded (Table 9.13).

Table 9.12 Number of live and dead mangrove roots in sediment cores, May 1989.

Site	Live	Dead	% Dead	Site	Live	Dead	% Dead
Oiled open habitat				Unoiled open habitat			
MINM	4,8,3	3,0,0	17	MSM	4,2,5	1,3,0	27
PGM	3,6,4	4,4,10	58	PBM	13,3,6	0,0,0	0
DROM	2,2,8	4,6,6	57	PADM	7,9,8	0,1,1	8
PMM	12,10,5	2,0,1	10	LINM	5,10,5	0,0,0	0
Total	67	40	37		77	6	7
Oiled channel habitat				Unoiled channel habitat			
SBCE	3,3,4	8,4,3	60	MACS	10,14,8	0,0,1	3
SBCS	7,2,10	3,8,3	42	MACN	3,8,8	5,2,0	27
PCE	4,4,5	0,4,2	31	HIDC	5,8,6	3,2,1	24
PCS	4,4,3	2,4,6	52	SBCW	5,5,8	4,0,2	25
LRCW	10,12,7	3,0,0	9	LRCS	13,5,3	1,4,3	28
Total	82	42	34		108	27	20
Oiled stream habitat				Unoiled stream habitat			
LRRS	6,7,4	9,3,4	48	HIDR	3,7,2	0,0,2	14
PAYR	1,5,3	2,2,5	50	UNR	2,2,6	3,6,2	52
PMRE	0,1,2	4,5,10	86	ALER	5,6,6	4,3,1	32
PMRW	13,6,6	6,3,3	32	MERR	3,3,6	5,1,3	43
Total	54	56	51		51	30	37

Note: Drawings of cores have been archived. Observations made by Jennifer MacPherson.

Surface sediments in mangrove areas were taken with a metal cork borer. Nine small cores were cut from a transect across the station. The surface (0 - 2 cm) sections were composited as three cores per jar in a regular pattern of 1-2-3 across the open sites. In river and channel areas, movement was restricted and three surface cores were taken around each large core and composited into each of three jars.

Seagrass sediment cores were taken with 45 cm x 8.5 cm stainless steel pipe, manually pushed into the sediment. Only cores greater than 24 cm in length were drained, plugged with corks, transported on ice to the laboratory, extruded onto foil and frozen. Sectioning was done as per the mangrove sediments. Surface sediments were obtained by using a glass jar as a scoop. Fifteen scoops from across each site were composited into three jars as per the mangrove surface sediments. The same procedure was used for sediments from coral reefs.

Table 9.13 Observations on oil in mangrove sediment cores at oiled sites, May 1989.

Site	Observations
Oiled Open Habitat	
MINM	1. oily smell 2. oily smell 3. no oily smell
PGM	1. no oily smell 2. oily smell, oil visible 3. oily smell, oil draining from core
DROM	1. oily smell 2. oily smell 3. oily smell
PMM	1. no oily smell 2. no oily smell 3. no oily smell
Oiled Channel Habitat	
SBCE	1. oil saturating dead roots 2. oily smell at all levels 3. oil saturation to 12 cm, oil draining from core
SBCS	1. oily smell, oil draining from core 2. oily smell, oil draining from core 3. oily smell
PCE	1. oily smell, oil draining from core 2. oily smell 3. oily smell, oil draining from core
PCS	1. oily smell, oil draining from core 2. oily smell, oil saturated roots to 30 cm 3. oily smell, visible oil to 13 cm
LRCW	1. oily smell 2. faint smell deep in core 3. faint smell deep in core
Oiled Stream Habitat	
LRRS	1. oily smell throughout core 2. faint oily smell 3. no oily smell
PAYR	1. oily smell all depths 2. oil saturated throughout 3. visible oil and oily smell throughout
PMRE	1. oily smell, iridescent sheen all depths 2. oily smell, iridescent sheen all depths 3. oily smell all depths
PMRW	1. oily smell, surface globules of oil 2. oily smell, surface globules of oil 3. no oily smell or visible oil

Note: Drawings of cores have been archived. Observations made by Jennifer MacPherson.

Coral heads contiguous to the biological study areas were dislodged using a chisel. The living portions were not touched as they were brought to the surface, handed into a small boat and allowed to drain on solvent-cleaned foil. No oil slicks were present to contaminate corals passed through the surface waters. Each head was wrapped in foil with a label giving the reef name, replicate number and coral species. The heads were placed in ice chests for transport to the laboratory. Since the coral heads needed to be large enough for meaningful sclerochronology, they may not have cooled completely during the return to the laboratory. Coral heads were then frozen. Removing the tissue from the coral heads was initiated using the compressed air technique as had been done on the 1986 samples. The heads were partially defrosted, the tissue blown off and composited into vials kept on ice. Most of the collection of *Siderastrea siderea* was processed in this manner. Approximately 4 g of tissue were taken from each head to a total of nine vials per station per species. This process was also applied to most of the *Diploria strigosa* collections. For the collections done in April/May 1989, tissue recovery was aided by mechanical scraping with stainless steel knives. For scraped samples, 10 g of the tissue/skeleton mixture were collected per vial. All specimens of *Porites astreoides* and *Acropora palmata* were processed by the scraping method. *Agaricia tenuifolia* is a thin fragile coral; boring sponges were removed and the remaining material was packed directly into glass jars.

Sample Storage

Samples were frozen and air freighted to the BBSR for analysis. At the BBSR, samples are stored in labeled boxes at -20°C in a locked deep freezer.

9.4 Analytical Quality Control and Quality Assurance Practices in the BBSR Organic Chemistry Laboratory

The BBSR organic chemistry laboratory follows quality and assurance procedures tailored to the needs of individual projects. Procedures relevant to petroleum hydrocarbon, PAH and oxidation product analysis in reference to the Bahía Las Minas oil spill study are described here.

9.4.1 Routine Instrument Calibrations

The basis of any chemical analysis is the analytical balance. BBSR has a complete set of calibration weights, and the organic chemistry staff routinely verify the accuracy of the balances. All calibration solutions of hydrocarbons are made up to dilutions based on weights. Solution weights are written on each vial and checked before use as calibration standards. Standards are the best available purity. Calibration mixtures are tailored to meet specific analytical objectives. Stock solutions are made up in isooctane or hexane, and dilutions for instrument calibrations are made fresh as use warrants. In addition to mixtures of pure

compounds made in the lab, some commercial solutions are purchased to act as certified checks on laboratory accuracy. These include a reference mixture of polynuclear aromatic hydrocarbons. The reference alkane mixture was made in 1984 and aliquots of the concentrated stock solution were heat-sealed in glass ampoules. Thus, when new dilutions are made from this stock, they are checked against the commercial PAH mixture and the most recent alkane calibration runs. In this way, the laboratory maintains a continuous record of calibration data for each instrument. Thus, the accuracy of a sample analysis is ensured not only by calculating sample data from response factors generated by standards run the same day, but also with the knowledge that the daily response factors are within the normal and well characterized range. In practice, the technicians operating the instruments recognize when the standard runs deviate more than 20% from the long-term average. When this happens, the standard mixtures are checked against each other and, if found to be satisfactory, then the instrument undergoes trouble-shooting procedures to return it to optimum sensitivity. If this is not possible, the system is completely recalibrated and samples are run only when responses are stable. This applies to the gas chromatographs (GCs), the gas chromatograph/mass spectrometry system (GC/MS), the high performance liquid chromatograph (HPLC) and the fluorometer (UVF).

GC-FID Calibrations

The linearity of the flame ionization detector (FID) response is checked by injection of a series of dilutions of hydrocarbons, some of which are used as internal standards. Calibration mixtures are made up in the range of 10 to 30 ng/ μ l. The FID GCs are linear in this range. Calibration mixtures of saturated or aromatic hydrocarbons (PAHs) or both are run daily when sample extracts are being analyzed. Response factors (RFs) are calculated from the areas generated and these are examined for consistency over time. The commercial PAH mixture run at two dilutions on the same days as the alkane standard showed that the RFs calculated over the same elution ranges are nearly identical. In this manner, laboratory-made mixtures are checked for accuracy. Standards injected at several dilutions also show the practical limit of detection on the FID. At the 2 to 4 ng level, the peaks are too small to be accurately integrated by the electronic integrator although they are clearly visible above the baseline. Samples are concentrated or diluted to yield accurately quantifiable results within the established linear range of the detector.

GC/MS

Internal auto-tune procedures conducted on a regular basis establish the mass accuracy and ion abundance requirements. Calibration mixtures are run in both total ion current and selected ion monitoring (SIM) mode. Response factors of peaks with mass/charge (m/z) ratios of compounds of interest are tabulated as per the GC-FID. When these deviate by more than 20%, evaluation and trouble-shooting procedures are initiated. An example of the reproducibility of retention times is given in Table

9.14. The data were generated by repeated manual injections of 1 μl aliquots of standard solutions into a standard vaporizing injector operating in the splitless mode. An example of a five point calibration curve or linearity test over the 50 pg through 5,000 pg range and the mass ion identifications of individual PAHs is shown in Figure 9.12. Similar calibration plots were produced for 13 PAHs and alkyl-substituted PAHs spanning the naphthalene through dibenzanthracene elution range. All PAH quantifications on the GC/MS through November 1989 were based on external standard procedures using response factors of the parent PAH for quantification of the alkyl PAH. Response factor tables will be upgraded for other alkyl PAH based on pure standards for an additional five alkyl compounds for which standards have been obtained. Response factors for others, for which no pure standards are available, must be interpolated from the relative abundance of specific mass fragments as determined from scan data on the Venezuelan/Mexican Isthmian crude oil. The deuterated surrogate compounds that were ordered for internal standard calculations were: d10-diphenyl, d10-phenanthrene and d12-perylene. For analysis of the 1988/89 Bahía Las Minas samples, the procedure will be changed to use the internal standard procedures as have already been applied to the analysis of the sterane biomarkers.

UVF

The signal-to-noise ratio of the PE 650 instrument is checked routinely using the Raman emission of distilled water as per the instructions in the manual. If it drops below the specified limit, the lamp is replaced. Standard response graphs are made from fresh dilutions of crude oil or other relevant calibration mixtures. A new calibration graph is prepared for use with every batch of samples. The analyst initiates trouble-shooting procedures if standard graphs vary by more than 20% on a daily basis. Because UVF is only a semi-quantitative technique, calibration standards must be tailored to the application. For fractionated samples, the D.VMIC was a quantitative standard for the aromatic fractions of the 1986 sediment samples. The procedure is currently being recalibrated to use 1-pyrenol as the calibration standard for oxygenated PAH in the more polar fractions of samples. A diesel oil will be used as a calibration standard for extracts of bivalve tissue (see Chapter 9.2.5).

9.4.2 Routine Blank and Recovery Experiments

Every batch of six to 10 samples has a blank extraction spiked with a mixture of analytical standards. This blank undergoes complete analysis as a sample, following all cleanup and evaporation stages. The procedures typically recover 80-100% of the C_{14} to C_{34} elution range. Recovery drops to 65 to 70% at C_{10} due to the evaporation steps. Table 9.15 shows an example of a spiked blank recovery experiment.

Table 9.14 Example of retention time reproducibility with SIM acquisition on the GC/MS system. Retention time data from MSD, July-August 1989.

MSD run	1	2	3	4	5	6	7	8	9	10	11	12
P5	17.94		22.62	28.56	31.69	37.51	44.91	46.22	53.78	54.01	62.27	
P10	17.94	22.02	22.66	28.56	31.70	37.50	44.90	46.21	53.75	54.00	62.20	69.48
P11	17.91	21.97	22.58	28.52	31.64	37.47	44.87	46.18	53.74	53.97	62.20	69.48
P13	17.90	21.98	22.60	28.53	31.66	37.49	44.89	46.20	53.75	53.99	62.21	69.48
P14	17.95	22.02	22.63	28.56	31.70	37.52	44.92	46.25	53.79	54.02	62.25	69.54
K29	17.97			28.59		37.53	44.93	46.25	53.80	54.04	62.26	69.55
K31	17.95			28.57		37.53	44.93	46.25	53.80	54.04	62.26	69.55
K26	17.99	22.07	22.71	28.63	31.75	37.57	44.97	46.28	53.82	54.07	62.28	69.54
K21	17.92	22.05	22.66	28.59	31.72	37.54	44.94	46.25	53.80	54.04	62.26	69.57
K27	17.87	21.93	22.56	28.49	31.61	37.43	44.83	46.14	53.69	53.92	62.14	69.42
K24	17.97			28.57		37.54	44.93	46.23	53.81	54.04	62.27	69.65
K32	17.96			28.57		37.53	44.93	46.24	53.80	54.03	62.28	69.63
K28	17.94	22.01	22.64	28.56	31.69	37.52	44.91	46.23	53.77	54.01	62.22	69.50
Avg.	17.94	22.01	22.63	28.56	31.69	37.51	44.91	46.22	53.78	54.01	62.24	69.53
STD	0.3	0.04	0.04	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.04	0.06
%Var.	0.17	0.18	0.18	0.11	0.12	0.09	0.07	0.07	0.06	0.06	0.06	0.09
(n)	13	8	9	13	9	13	13	13	13	13	13	12

1. Naphthalene m/z 128; 2. 2-M-Naphthalene m/z 142; 3. 1-M-Naphthalene m/z 142; 4. Acenaphthene m/z 153; 5. Fluorene m/z 166; 6. Phenanthrene m/z 178; 7. Fluoranthene m/z 202; 8. Pyrene m/z 202; 9. 1,2-Benzanthracene m/z 228; 10. Chrysene m/z 228; 11. Perylene m/z 252; 12. 1,2,5,6-Dibenzanthracene m/z 278.

Ideally, internal standards are added to every sample to compute recovery through the procedures, and the results are corrected for recovery. The organic chemists at BBSR use this level of precise sample handling for samples that yield similar levels of contaminants. In practice, with oil spill sediment samples, concentrations vary over many orders of magnitude making it impossible to estimate how much internal standard to add. Thus, a second recovery standard is added after determination of total lipids. Recovery is also based on that of the standards in the spiked blanks and precision among replicate samples. Tissue samples are more amenable to accurate use of the internal standard method, as the range of concentration of hydrocarbons is much narrower than in sediments. When samples are reduced in volume for GC or GC/MS analysis, a GC quantification surrogate standard is added just before analysis to facilitate accurate determination of dilution factors and for use as internal standards for the quantifications. Examples of standards used are C_{18:1}, C_{22:1}, O-terphenyl and d10-phenanthrene, which resolve from usual peaks in the sample chromatograms. The O-terphenyl is a particularly useful standard for samples undergoing UVF analysis, as it elutes with the bulk of the aromatic hydrocarbons during the isolation procedures, but has negligible fluorescence. As stated above, deuterated surrogate compounds were obtained for the internal standard method of quantification of PAHs by GC/MS.

Naphthalene (1)
Sample amount: 1.003
Multilevel curve fit: Linear
Reference peak window: .5:00 % of Ret Time

Ret Time	Pk #	Ch Description	Amt pg/ul	Lvl	(Area)
17.909	1	Ion Mass 128	50.15	1	1117
			250.8	2	17431
			501.5	3	39318
			1254	4	81788
			5015	5	304182

HP MSD September, 1989

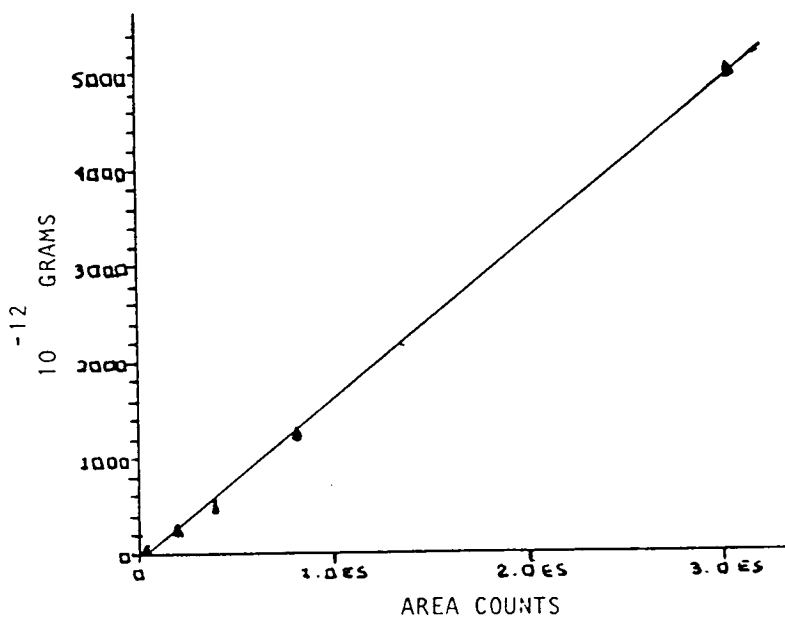


Figure 9.12 Sample calibration graph showing the linear response of the GC/MS in SIM mode over the 50 to 5,000 pg range.

Table 9.15 Example of spike-recovery experiments. Standard mixture eluted from silica/alumina column and fractions rotary evaporated.

Compound	Date:	% recovery									
		F1					F2/3				
		16/1 SP1	23/1 SP2	1/1 SP3	mean	%cv	16/1 SP1	23/1 SP2	1/2 SP3	mean	%cv
C ₁₀ naphthalene		70	69	65	68	3.8	61	75	78	71	1.7
C ₁₂ triethylbenzene		78	82	77	79	3.3	68	78	79	75	8.1
1-methyl naphthalene							68	77	79	75	7.8
1-ethyl naphthalene							70	76	78	75	5.5
2,6-dimethyl naphthalene							72	77	80	76	5.2
C ₁₄ di-methyl phthalate		83	82	79	81	2.5					
acenaphthene							77	80	82	80	3.1
2,3,6-trimethyl naphthalene							76	79	80	78	2.6
fluorene							77	77	80	78	2.2
C ₁₆		86	86	84	85	1.1					
C ₁₇ pristane		89	86	89	88	1.9					
phenanthrene		87	84	91	87	4.0					
n-octadecene							83	82	84	83	1.2
C ₁₈ phytane		86	83	*	85	2.5					
2-methyl phenanthrene		88	88	*	88	0.0					
1-methyl phenanthrene		87	85	*	86	1.6					
3,6-dimethyl phenanthrene							90	83	85	86	4.1
C ₂₀ fluoranthene							101	87	87	92	8.8
pyrene							76	79	79	78	2.2
C ₂₁ di-pentyl phthalate		86	87	89	87	1.7					
C ₂₂ 1-methyl pyrene							87	87	86	87	0.6
C ₂₄ chrysene							81	88	86	85	4.2
C ₂₆ squalane		83	88	89	87	3.7					
di-octyl phthalate							81	87	85	84	3.6
C ₂₈ perylene		81	88	89	86	5.0					
C ₃₀							79	85	83	82	3.7
C ₃₂		81	88	89	86	5.0					
C ₃₄							85	73	80	79	7.6
		85	86	*	86	0.8					
		84	84	*	84	0.0					
		88	83	*	86	4.1					
							109	52	52	71	46.3
		93	78	95	89	10.4					
		99	68	96	88	19.5					
		107	57	88	84	30.0					

9.4.3 Coding of Samples for Analysis

As samples are removed from the freezer for analysis they are assigned a sequential number. The inventory number is listed against the sample analytical number in the laboratory notebook. From this stage on, the analyst does not know the background of the samples. It is not until the data collation stage that the samples are grouped into sites and analytical replicates. In this way, analytical bias is minimized.

9.4.4 Adherence to Internationally Recognized Analytical Procedures and Analysis of Standard Reference Materials

The methods used in the BBSR organic chemistry lab for the analysis of petroleum hydrocarbons in marine samples conform to the IOC/UNEP Manual and Guides Nos. 13 and 11 (UNESCO 1984, 1990). A Quality Control Chart is kept by the BBSR organic chemistry lab using the Ma-K-1 oyster sample reference material for petroleum hydrocarbons. This material from the IAEA, Monaco, is being analyzed yearly during the laboratory's participation in the Bahía Las Minas oil spill study.

9.5 Acknowledgments

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Chapter 10 Physical Environmental Monitoring

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10.1 Introduction

The basic purpose of the Smithsonian Institution's Environmental Sciences Program (ESP) is to monitor long-term physical environmental and biological variation, i.e., to collect "baseline" data. A marine program was initiated at the Galeta Marine Laboratory in 1972. This site had been a Smithsonian Tropical Research Institute (STRI) biological preserve since 1965. After the 1968 oil spill from the tanker *Witwater* (Rützler and Sterrer 1970), there was a three-year survey (1970-1973) of intertidal communities (Birkeland *et al.* 1976). The Galeta ESP started during this survey. Physical environmental data have been collected continuously since then, with some gaps and changes in parameters or instruments (Meyer and Birkeland 1974; Meyer *et al.* 1975; Hendler 1976, 1977a; Cubit *et al.* 1986, 1988b, 1989).

Management of ESP data from Galeta was modified during 1988. Data from January 1986 until September 1989 have been entered into computer files, checked, and verified. Graphs have been prepared and summary statistics have been calculated for some of the more recent data. Below, we describe the kinds of physical environmental data being collected and the instruments used, and present a summary table of monthly checks on each instrument and the necessary adjustments, replacements, or calibrations. We also show environmental data during the 1986 spill.

10.2 Monitoring Methods

The parameters monitored are water level, air temperature, rainfall, solar radiation, wind speed and direction, salinity, and upstream (seaward) and downstream (landward) water temperatures. Most of the monitoring devices are affixed to the reef flat adjacent to the laboratory, often taking advantage of existing concrete structures from earlier military use of the reef flat. Subsurface (3 m depth) water temperature has been monitored since 1988.

Types of equipment used to monitor the above factors (see Cubit *et al.* 1988b) are based on years of experience with several criteria: resolution, cost-effectiveness, ability to withstand the harsh environment on the Caribbean coast of Panama (high temperatures, suspended sediments, air-borne salts, wave-driven debris, high humidity, intense solar radiation, heavy rainfall, strong winds, perching birds, nesting wasps, excavating animals, crevice corrosion, electrolysis, unstable electrical service, and heavy lightning), and serviceability of the equipment in this remote area. Instruments are usually inspected five days per week for proper function and need for maintenance. Table 10.1 shows the number of checks carried out on the recorder and sensor, and the number of calibrations, adjustments, or replacements made. Such changes were not frequent but may require that certain data be corrected or discarded, depending on the nature of the change.

Table 10.1 Summary of monthly inspections of environmental monitoring equipment at Punta Galeta, January 1987 - June 1989. The first number of each pair is the number of inspections; the number after the "/" is frequency of adjustments, calibrations, or replacements.

Year	Parameter	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Total
1987	Sol. Rad.	4/0	4/0	6/1	4/0	4/0	5/0	5/0	7/0	4/0	6/0	5/0	4/0	58/1
	Rainfall	16/0	15/0	15/0	8/0	15/0	11/0	18/0	19/0	12/0	13/0	14/0	11/0	177/0
	Tide	15/0	13/0	16/1	15/0	15/0	16/1	16/0	20/0	17/0	12/0	12/0	14/0	181/2
	Wind	5/1	5/0	4/0	4/0	3/0	2/0	3/0	6/2	4/0	5/0	4/0	5/0	50/3
	Sea Temp.	5/0	6/0	5/0	4/0	5/0	5/0	4/0	7/0	4/0	4/0	6/0	4/0	59/0
	Air Temp.	5/0	6/0	5/0	4/0	5/0	5/0	4/0	7/0	4/0	4/0	6/0	4/0	59/0
	Salinity	16/0	14/0	14/0	19/0	15/0	17/0	17/0	19/0	19/0	18/0	17/0	15/0	200/0
1988	Sol. Rad.	4/0	12/0	22/0	19/1	18/1	15/1	12/0	18/0	16/0	18/0	15/0	11/0	180/3
	Rainfall	7/0	10/0	15/0	20/0	18/0	16/0	12/0	8/0	11/0	10/0	11/0	8/0	146/0
	Tide	13/0	16/3	22/1	20/3	19/0	18/0	17/0	20/0	18/0	19/1	19/0	14/0	215/8
	Wind	5/0	10/1	22/0	19/0	19/0	15/0	12/0	19/0	15/0	18/1	17/0	11/0	182/2
	Sea Temp.	5/0	12/0	22/0	19/0	18/0	16/0	14/0	20/0	17/0	16/0	17/0	12/0	188/0
	Air Temp.	5/0	12/0	22/0	19/0	18/1	16/1	14/0	20/0	17/0	16/0	17/0	12/0	188/2
	Salinity	17/0	19/0	22/0	20/0	20/0	20/0	20/0	21/0	21/0	19/0	19/0	13/0	231/0
1989	Sol. Rad.	11/1	14/0	20/0	8/1	11/0	14/1	17/0	15/0	14/0	17/0	10/0	7/0	152/3
	Rainfall	8/0	8/0	15/0	9/0	14/0	10/0	9/0	14/0	8/0	11/0	12/1	5/0	123/1
	Tide	14/0	17/3	26/0	16/0	14/0	19/0	17/1	15/1	15/0	14/0	13/0	10/0	190/5
	Wind	12/0	16/0	20/0	10/1	13/0	15/0	18/0	17/0	16/1	14/0	11/0	8/0	170/2
	Sea Temp.	12/0	17/0	19/0	10/0	14/0	17/0	17/0	16/1	15/0	14/0	12/0	9/0	172/1
	Air Temp.	12/0	17/0	19/0	10/0	14/0	17/0	17/0	16/0	13/0	14/0	12/1	9/0	170/1
	Salinity	15/0	16/0	22/0	17/0	20/0	21/0	20/0	22/0	19/0	19/0	19/0	11/0	221/0

10.2.1 Water Level

Water level is measured with a Stevens "Type A" water level recorder (Leopold E. Stevens Inc., Beaverton, Oregon). This instrument contains a mechanical chart recorder whose pen is directly driven through a gear connected by a pulley to a float and counterweight. It is a still-well type recorder; rates of chart movement and accuracy of measurement of water level are manually checked about five times per week by comparison with direct measurements using a meter stick. The same apparatus has been kept in use because its accuracy, precision (± 0.25 mm), resolution, and reliability were better than any electronic method that could be found.

The data recorded are continuous water levels expressed relative to an arbitrary datum at the base of the tide gauge. The datum is approximately 30 cm below extreme low water (Cubit *et al.* 1986). The mean diurnal range of water level is 25 cm; the range between extreme high and low waters averages 59 cm (Cubit *et al.* 1986). Fluctuations in water level tend not to be simply tidal and depend largely on wave action. Rough sea conditions appear to push water onto the reef flat and raise water level on it relative to the open sea.

Efforts are still being made to find reliable electronic equipment to supplement the existing mechanical equipment. The disadvantage of the chart recorder is the laborious effort of calculating sea levels from charts and then entering the data into computer files.

10.2.2 Air Temperature

Hourly mean air temperatures are monitored with a Datapod 212 portable digital recorder (Omnicdata International Inc.) connected to a sensor mounted about 1.5 m above mean low water level. The sensor is shielded from rainfall, salt spray, static charges, and direct sun light. Temperatures are checked every two to three days with a precision ($\pm 0.5^\circ\text{C}$) mercury thermometer. The accuracy of the monitored temperatures is 0.5°C .

10.2.3 Rainfall

Rainfall is measured with a Stevens "Type A" recorder (the same type as for water level). Its float is in a cistern connected to a 30.5 cm diameter collector on the roof of the laboratory. The data recorded are continuous, cumulative rainfall converted to total hourly accumulations. The precision of the measurements is ± 0.25 mm. The data recorded are checked against data from simple collector gauges. Also, a known volume of water is periodically added to the cistern to ensure proper functioning of the recorder.

10.2.4 Solar Radiation

Wide-spectrum solar radiation is monitored by a Datapod 211 digital recorder connected to a LI200 Licor Pyranometer Sensor with a range of 400-1100 nm (Li-cor Inc.). Data are recorded as the average millivolt output for one-hour intervals and converted to watts/m² for analysis. The precision of this instrument is ± 32 watts/m². Solar radiation is checked against an identical sensor kept in the laboratory as a standard. The standard is returned to the manufacturer every two years for recalibration.

10.2.5 Wind Speed and Direction

An anemometer at the seaward edge of the reef flat measures wind speed and direction 3 m above low water level. A Datapod 214 records hourly mean wind speed, hourly mean wind direction, hourly peak wind speed, and the direction of peak speed. Wind speed is checked against a hand-held anemometer several times a week. An identical anemometer is operated 3 m away from the primary one as a backup.

10.2.6 Salinity

Salinity is measured with a temperature-compensated, hand-held refractometer (American Optical Co.) about five times per week for water samples taken from the upper 15 cm of water flowing across the reef flat and past the end of the laboratory dock. The precision of the refractometer is ± 1 o/oo. The zero point of the refractometer is checked using distilled water before each daily measurement. A standard saline solution will be prepared (or obtained) for routine calibrations.

10.2.7 Upstream and Downstream Water Temperatures

Water temperatures are recorded hourly with a Datapod 212 (upstream) and 211 (downstream). The upstream sensor is located approximately 36 m downstream from the seaward edge of the reef flat. During the lowest water levels, it is about 20 cm below the surface of a 1.5-m diameter pool. The downstream sensor was installed in 1984 approximately 91 m downstream from the upstream sensor in very similar circumstances (1.5-m diameter pool, 25 cm deep at lowest low water). Temperature readings generally are checked five times per week (at least once) with a precision mercury thermometer. The tolerance limits for monitored temperatures are $\pm 0.5^{\circ}\text{C}$ of the thermometer reading (Cubit *et al.* 1988b).

10.2.8 Subtidal Temperature

Daily minimum, maximum, and mean temperatures are monitored at a depth of 3 m with a Datapod 212 at three different sites with two sensors for each recorder.

The sites are Punta Galeta, Isla Payardi, and Isla Grande (Figure 1.1). The recorders are protected by an Ikelite housing. One sensor is inside the housing and the other is attached to the sea floor. Data from both sensors are compared to check for proper functioning.

10.3 Environmental Conditions during the 1986 Oil Spill

Weather and sea conditions affecting movements of the spilled oil were described briefly in Jackson *et al.* (1989). Here we show environmental data for April-June 1986 (Figures 10.1 - 10.3); the spill occurred on 27 April (Chapter 1). These data show the rainfall and shifting winds that flushed oil out of Bahía Cativá toward the open sea in early May (Figure 10.2). Also apparent are the subsequent low water levels that caused the spilled oil to accumulate on the seaward margin of reef flats (Chapters 1 and 2).

10.4 Discussion

The ESP data for populations and communities on the reef flat at Punta Galeta provided extensive sets of pre-spill data for comparisons with data after the spill (Chapter 2). This long-term information shows the extreme variation through time in abundances of organisms on the reef flat and continues to be collected. The physical environmental data enabled comparison of sea level during the spill against average conditions (Jackson *et al.* 1989). The data also show general features of the environment on the reef flat, such as the influence of water level on both water and air temperatures.

Daytime exposures of reef flats were unusually extreme during June - September 1988. Such continuing natural variability, which in this case caused considerable reductions in abundances of organisms on reef flats for this time of year (Chapter 2), is one sort of "noise" that complicates interpretations of ecological recovery in this habitat following the spill.

Legend for Figures 10.1 - 10.3

"Rain" = rainfall (mm).

"Wind" = average hourly wind speed (vertical line between "W" and "S" = 20 km/h).
Wind direction is indicated by the abbreviated compass headings, "N", "W", "S", and "E". The open circles show when the maximum was greater than 20 km/h more than the average for that hour, e.g., storm-associated gusts.

"AT" = air temperature (°C).

"WT" = upstream water temperature (°C).

"SR" = solar radiation (watts/m²).

"Delta WT" = upstream minus downstream water temperature (°C).

"WL" = water level relative to Galeta datum (cm) (Cubit *et al.* 1986); extensive emersion of the reef flat occurs when water level is below 30 cm.

"Sal" = salinity (o/oo).

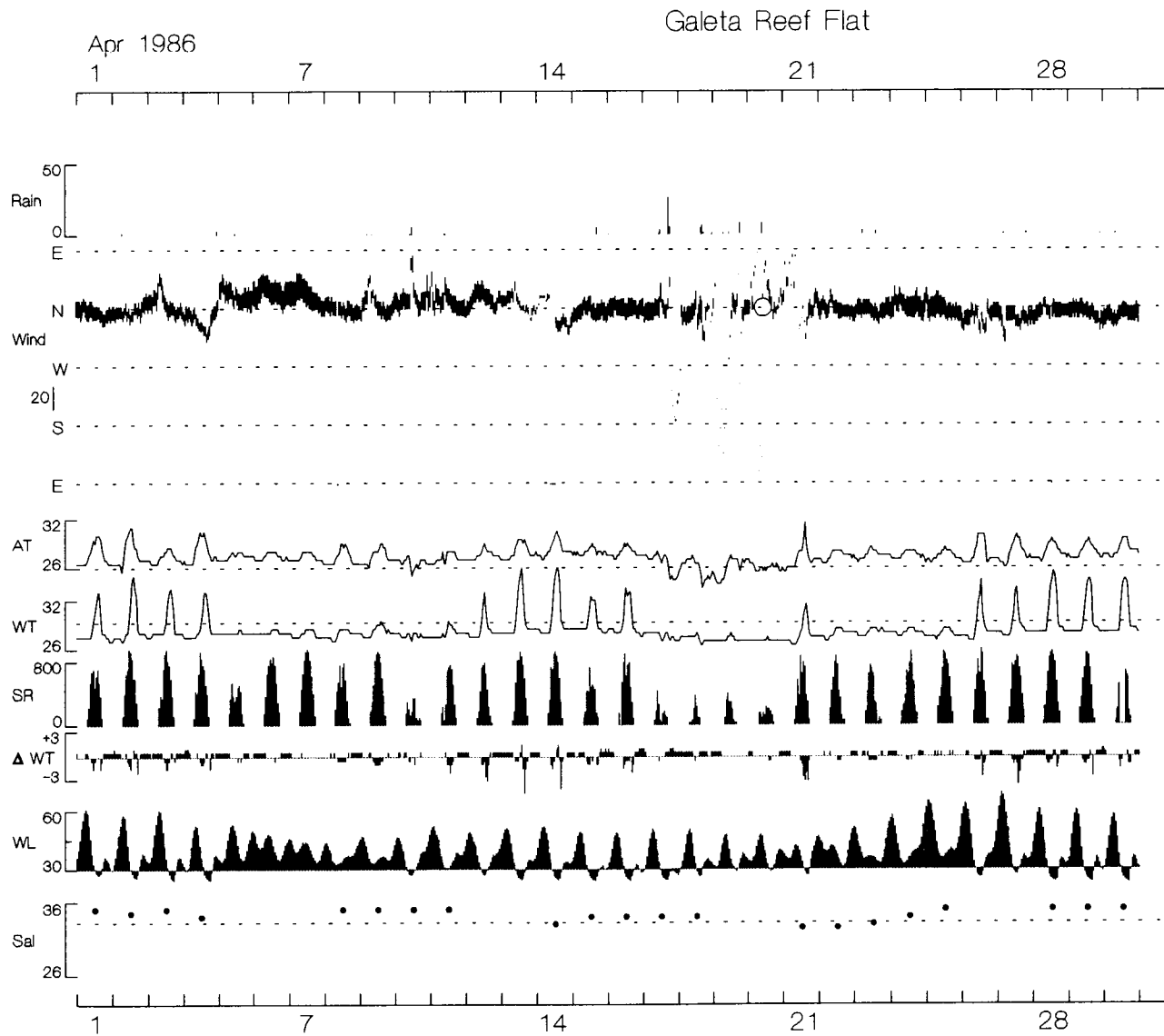


Figure 10.1 Physical environmental data for April 1986; the oil spill occurred on 27 April.

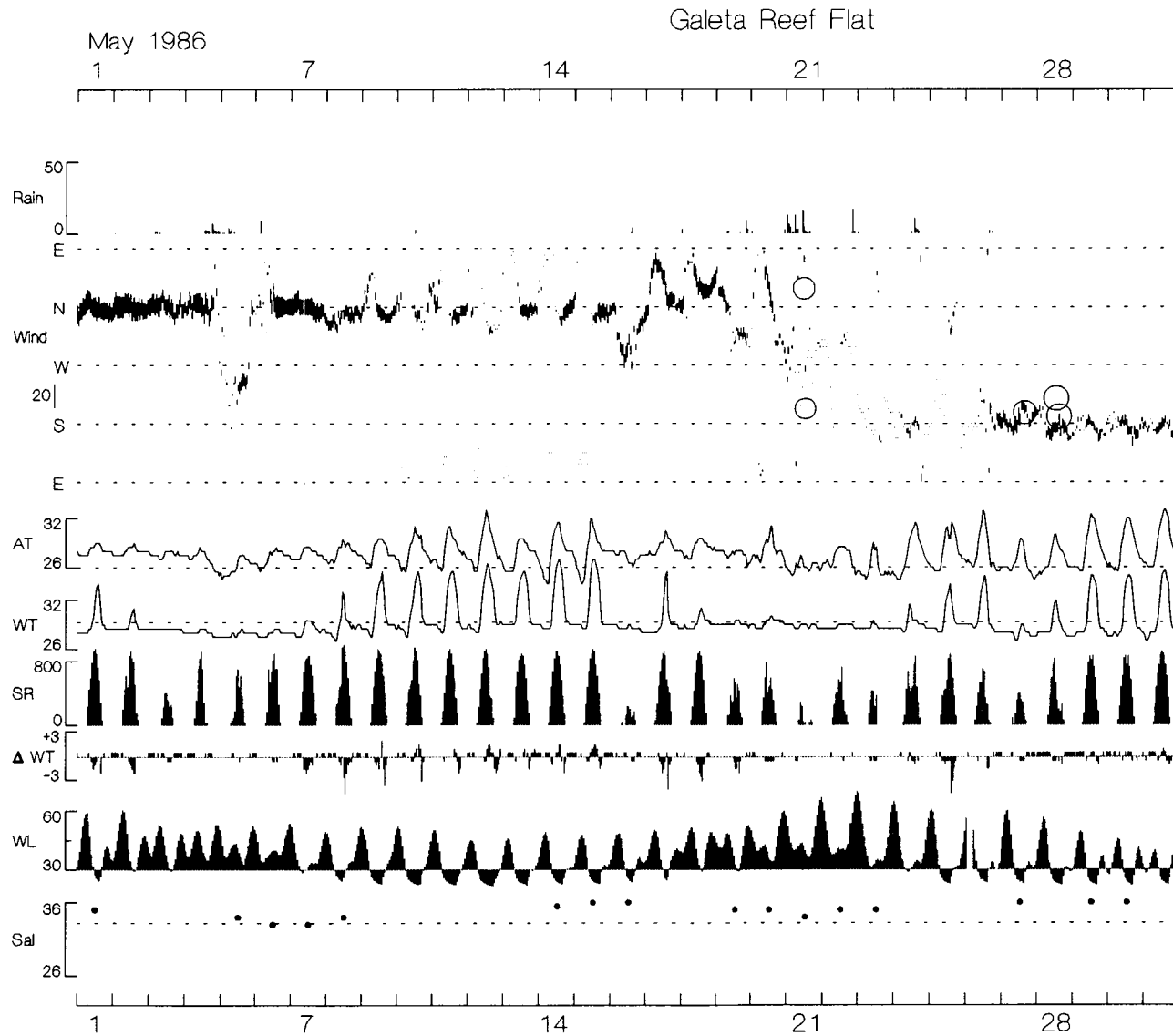


Figure 10.2 Physical environmental data for May 1986, the month after the oil spill.

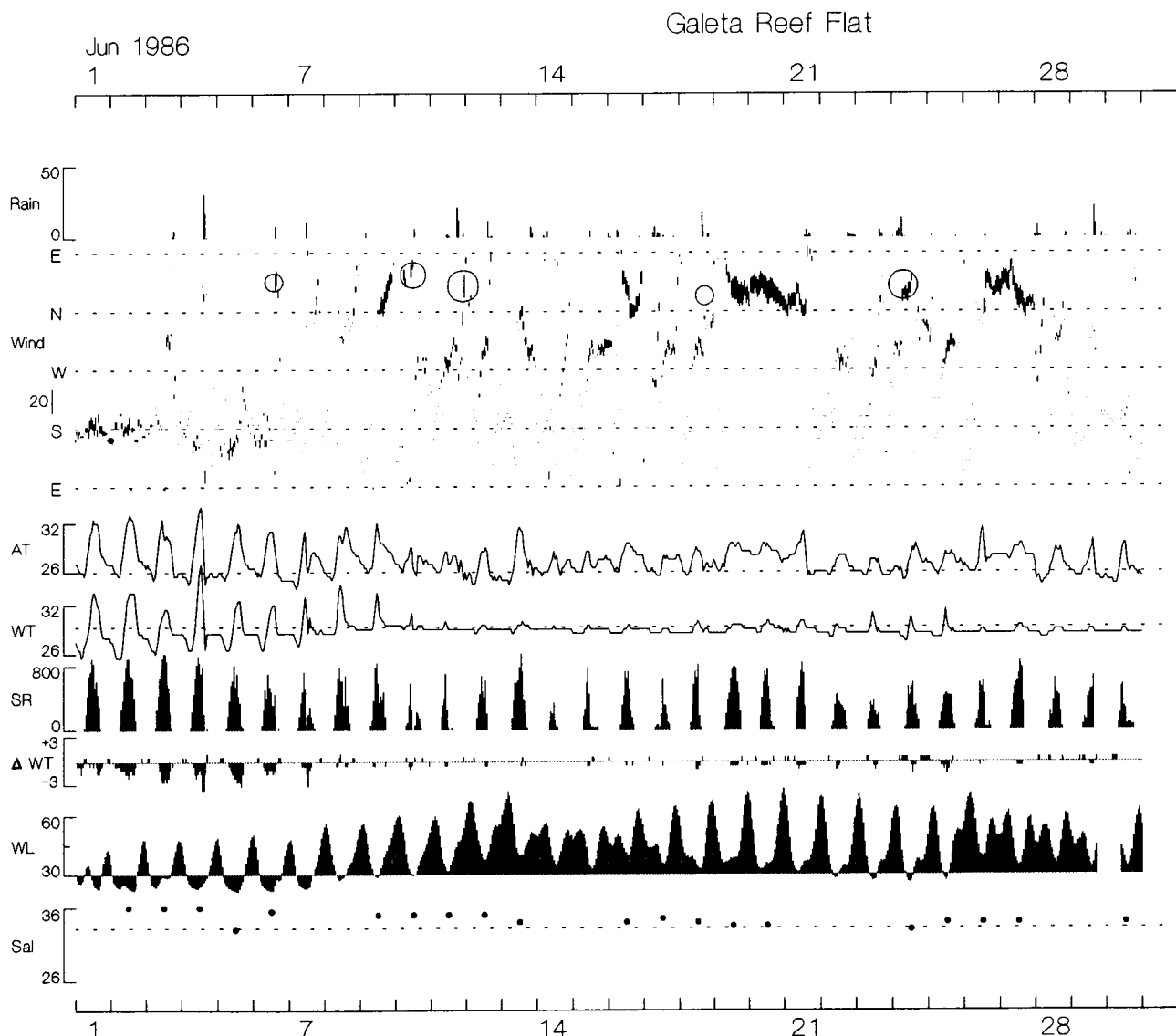


Figure 10.3 Physical environmental data for June 1986, two months after the oil spill.

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Appendix A

Data Management

Data Management

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Personnel: M. Helena Fortunato and Gabriel Jácome

A.1 Responsibilities of the Data Management Section

Managing the data begins with insuring that they are collected accurately, efficiently, and completely in the field. This is done through the preparation of data sheets of a uniform style for each subproject. It includes entering and checking the data and providing analysis files to the investigators along with simple summary reports of counts, means, and variances. Finally, it includes keeping complete and accurate archives for both the data sheets and the entered data.

A.2 Magnitude of the Data Management Task

There are seven biological projects within the oil spill study, and each has several subprojects requiring a different data file format (Table A.1). There are a total of 35 such subprojects. In addition, data from the Environmental Sciences Program (ESP) are managed.

At the time of this report, 231 separate collections of data on 5,921 data sheets have been entered and checked (Table A.2). The data sets from the subprojects now range in size from 48 to 70,845 lines with a total 332,804 lines. There are 541 entries in all of the species lists. The raw data files alone occupy over 17 megabytes of storage space and the total space occupied by all of the project computer files is 76 megabytes, not counting backup copies. In addition, there are 60 megabytes of biological data and 12 megabytes of physical data from the ESP. This large amount of data requires different techniques for management than do the usual small data sets most ecologists work with.

Table A.1 List of projects, subprojects and their codes.

MGR	- Mangrove Roots
	C Community development
	D Vertical distribution of organisms
	L Long term study of cover
	M Mobile organisms
	R Recruitment
	W Weights of organisms on dowels (biomass)
SRE	- Subtidal Reef Corals
	C Sclerochronology
	F Percent partial mortality
	P Percent cover - coral reef transects
	R Resuspended sediments
	S Suspended sediments
RFL	- Reef Flat Communities - Sea urchins, sessile macrobiota, and infauna of <i>Laurencia</i> algal beds.
	B Urchins census (Bird exclosures)
	C Percent Cover
	I Infaunal study (algae core census)
	U Urchins census
GRS	- Subtidal Seagrass Communities
	C Coralline/non-coralline algae weights - pushnet samples
	E Environmental data
	G Major group counts - core samples
	L Coralline/non-coralline algae weights - core samples
	N Counts of major groups - pushnet samples
	O Sediments (granulometrics, organics, CaCO ₃)
	P Plant weights - core samples
	R Reproductive condition of animals
	S Species counts - pushnet samples

Table A.1 List of projects, subprojects and their codes (continued).

SNA	- Reef Flat Gastropods
	D Densities
	S Size frequencies
STO	- Reef Flat Stomatopods
MGF	- Mangrove Forests
	D Seedlings
	L Litter fall
	S Shoot measurement
	T Temperature/salinity
MGx	- Mangrove Forests (Dr. Getter's data)
	F Foliage - obsolete
	L Leaf litter - obsolete
	P Plot - obsolete
	S Station - obsolete
	T Transect - obsolete

Table A.2 List of data in the Oil Spill Project archives (to October 1989).

Project Code ¹	CID ²	Pages ³	First Day of Collection	Project code	CID	Pages	First Day of Collection
Mangrove Roots							
MGR B	9	4	Nov 1, 1987	MGR L	9	76	Nov 14, 1987
MGR B	10	4	Feb 17, 1988	MGR L	10	97	Feb 17, 1988
MGR B	11	6	Feb 24, 1988	MGR L	12	107	Aug 8, 1988
MGR B	12	2	Aug 8, 1988	MGR L	11	95	May 18, 1988
MGR B	13	6	Nov 24, 1988	MGR L	13	65	Nov 24, 1988
MGR B	14	7	Mar 1, 1989	MGR L	14	196	Feb 12, 1989
MGR B	15	7	May 13, 1989	MGR L	15	192	May 3, 1989
				MGR L	16	194	Aug 15, 1989
MGR C	7	12	May 15, 1987				
MGR C	8	0	Aug 1, 1987	MGR M	7	8	Jul 16, 1987
MGR C	10	30	Feb 18, 1988	MGR M	9	6	Dec 15, 1987
MGR C	11	41	May 19, 1988	MGR M	10	6	Feb 17, 1988
MGR C	12	43	Aug 8, 1988	MGR M	14	12	Mar 21, 1989
MGR C	13	35	Nov 24, 1988	MGR M	12	12	Sep 19, 1989
MGR C	14	42	Feb 12, 1989				
MGR C	15	44	May 3, 1989	MGR R	8	8	Aug 1, 1987
MGR C	16	43	Aug 15, 1989	MGR R	10	19	Feb 22, 1988
				MGR R	11	24	May 23, 1988
MGR D	12	130	Aug 8, 1988	MGR R	12	24	Aug 9, 1988
MGR D	13	130	Nov 24, 1988	MGR R	13	25	Nov 29, 1988
MGR D	14	8	Mar 1, 1989	MGR R	14	25	Feb 27, 1989
				MGR R	15	26	May 13, 1989
MGR E	14	4	Mar 2, 1989	MGR R	16	26	Aug 19, 1989
MGR E	15	7	May 24, 1989				
MGR L	1	0	Sep 1, 1981	MGR S	13	2	Nov 24, 1988
MGR L	2	0	Jan 1, 1982	MGR S	14	1	Feb 12, 1989
MGR L	3	0	Jun 1, 1982	MGR S	15	2	May 8, 1989
MGR L	4	33	Jul 1, 1986	MGR S	16	2	Aug 16, 1989
MGR L	5	26	Oct 1, 1986	MGR T	14	1	Feb 12, 1989
MGR L	6	127	Feb 13, 1987				
MGR L	7	83	May 1, 1987	MGR W	16	6	Aug 31, 1989
MGR L	8	56	Aug 13, 1987				

¹ The first three letters are the project code in the name of the data file; the fourth letter is the subproject code in the name.

² The collection identification number.

³ Number of data sheets.

Table A.2 List of data in the Oil Spill Project archives (continued).

Project Code	CID	Pages	First Day of Collection	Project code	CID	Pages	First Day of Collection
Subtidal Seagrass Communities				Mangrove Forests			
GRS D	8	20	Jan 16, 1988	MGF L	2	26	Jul 11, 1989
GRS G	3	35	Nov 14, 1986	MGF L	3	26	Aug 8, 1989
GRS G	3	49	Nov 14, 1986	MGF L	4	26	Sep 12, 1989
GRS G	4	42	Jan 19, 1987	MGF S	1	30 ⁴	Jun 8, 1989
GRS G	5	3	Apr 18, 1987	MGF S	2	x ⁴	Jul 11, 1989
GRS G	6	3	Jul 23, 1987	MGF S	3	x ⁴	Aug 8, 1989
GRS G	7	30	Oct 8, 1987	MGF T	2	1	Jul 11, 1989
GRS G	8	39	Jan 16, 1988	MGF T	3	1	Aug 8, 1989
GRS H	3	22	Nov 14, 1986	MGF T	4	1	Sep 12, 1989
GRS H	8	14	Jan 16, 1988	MGF T	5	1	Oct 10, 1989
GRS L	1	0	Sep 4, 1986	MGx ⁵ F	1	12	Sep 2, 1986
GRS L	3	0	Nov 14, 1986	MGx L	2	19	Oct 1, 1986
GRS L	4	4	Jan 19, 1987	MGx L	3	19	Oct 29, 1986
				MGx L	4	15	Nov 27, 1986
GRS N	3	22	Nov 14, 1986	MGx P	1	27	Sep 2, 1986
GRS N	4	20	Jan 19, 1987	MGx P	2	33	Sep 29, 1986
GRS N	5	23	Apr 8, 1987	MGx P	3	33	Oct 28, 1986
GRS N	6	25	Jul 23, 1987	MGx P	4	27	Nov 27, 1986
GRS N	7	46	Oct 8, 1987	MGx S	1	27	Sep 4, 1986
GRS N	8	30	Jan 16, 1988	MGx S	2	27	Sep 30, 1986
GRS N	9	42	Apr 13, 1988	MGx S	3	27	Oct 29, 1986
				MGx S	4	27	Nov 27, 1986
GRS O	7	10	Oct 8, 1987	MGx T	1	9	Sep 3, 1986
				MGx T	2	9	Sep 30, 1986
GRS P	1	0	Sep 4, 1986	MGx T	3	9	Oct 30, 1986
GRS P	3	0	Nov 14, 1986	MGx T	4	9	Nov 27, 1986
GRS P	4	77	Jan 19, 1987				
GRS P	8	80	Jan 16, 1988				
GRS R	3	67	Nov 21, 1986				
GRS S	1	149	Sep 4, 1986				
GRS W	2	8	Oct 8, 1987				

⁴ Collections 1, 2, 3 are on same data sheet.⁵ Inactive files collected by Dr. Getter.

Table A.2 List of data in the Oil Spill Project archives (continued).

Project Code	CID	Pages	First Day of Collection	Project code	CID	Pages	First Day of Collection
Reef Flat Communities - Sea urchins, sessile macrobiota, and infauna of <i>Laurencia</i> beds.							
RFL B	196	4	Jun 17, 1988	RFL U	196	1	Jun 17, 1988
RFL B	199	4	Sep 28, 1988	RFL U	197	1	Jul 17, 1989
RFL B	203	4	Jan 31, 1989	RFL U	198	1	Aug 17, 1989
RFL B	200	4	Oct 28, 1988	RFL U	199	16	Sep 20, 1988
RFL B	202	5	Dec 27, 1988	RFL U	200	1	Oct 28, 1988
RFL B	204	4	Feb 28, 1989	RFL U	202	4	Dec 27, 1988
RFL B	205	4	Mar 29, 1989	RFL U	203	1	Jan 30, 1989
RFL B	205	4	Mar 8, 1989	RFL U	204	1	Feb 28, 1989
RFL B	206	4	Apr 24, 1989	RFL U	205	4	Mar 29, 1989
RFL B	207	4	May 26, 1989	RFL U	206	1	Apr 24, 1989
RFL B	208	3	Jun 28, 1989	RFL U	207	1	May 25, 1989
RFL B	197	4	Jul 17, 1989	RFL U	208	4	Jun 23, 1989
RFL B	198	4	Aug 17, 1989	RFL U	209	1	Jul 25, 1989
RFL B	209	4	Jul 26, 1989	RFL U	210	1	Aug 28, 1989
RFL B	210	4	Aug 28, 1989	RFL U	211	1	Sep 25, 1989
RFL B	211	4	Sep 26, 1989	RFL U	212	3	Oct 11, 1989
RFL B	212	4	Oct 27, 1989				
RFL C	203	25	May 23, 1988				
RFL C	204	25	Sep 20, 1988				
RFL C	205	25	Mar 27, 1989				
RFL C	206	26	Jun 22, 1989				
RFL C	207	20	Oct 10, 1989				
RFL I	1	107	Sep 1, 1988				
RFL I	1	70	Sep 1, 1987				
RFL I	2	79	Apr 1, 1988				
RFL I	2	80	Apr 1, 1988				
RFL I	3	85	Oct 1, 1988				
RFL I	4	83	Apr 1, 1989				

Table A.2 List of data in the Oil Spill Project archives (continued).

Project Code	CID	Pages	First Day of Collection	Project code	CID	Pages	First Day of Collection
Subtidal Reef Corals							
SRE C	1	76	Aug 17, 1987	SRE R	10	1	Jun 22, 1989
				SRE R	11	1	Jul 10, 1989
SRE E	4	23	Feb 18, 1988	SRE R	12	1	Aug 1, 1989
				SRE R	13	1	Sep 5, 1989
SRE F	1	39	Jul 31, 1986	SRE R	14	1	Oct 6, 1989
SRE F	2	17	Oct 6, 1986				
SRE F	3	76	Nov 14, 1986	SRE S	1	1	Oct 1, 1987
SRE F	4	24	Apr 30, 1987	SRE S	3	1	Dec 13, 1987
SRE F	5	24	Aug 7, 1987	SRE S	4	1	Feb 18, 1988
SRE F	6	24	Nov 24, 1987	SRE S	5	1	Apr 21, 1988
SRE F	7	24	May 3, 1988	SRE S	6	1	May 18, 1988
SRE F	8	20	Jun 28, 1988	SRE S	7	1	Jun 22, 1988
SRE F	9	24	Nov 2, 1988	SRE S	8	1	Jul 21, 1988
SRE F	10	24	Mar 28, 1989	SRE S	9	1	Sep 8, 1988
SRE F	11	24	Jun 30, 1989	SRE S	10	2	Oct 3, 1988
SRE F	12	24	Sep 28, 1989	SRE S	11	1	Nov 6, 1988
				SRE S	12	1	Dec 1, 1988
SRE L	14	80	Aug 16, 1989	SRE S	13	1	Apr 1, 1989
				SRE S	14	1	May 1, 1989
SRE P	1	308	Jul 17, 1985	SRE S	15	1	Jun 13, 1989
SRE P	2	95	Jul 24, 1986	SRE S	16	1	Jul 5, 1989
SRE P	3	47	Jul 8, 1987	SRE S	17	1	Aug 1, 1989
SRE P	4	68	May 19, 1988	SRE S	18	1	Sep 6, 1989
SRE P	5	123	Jun 21, 1989	SRE S	19	1	Oct 6, 1989
SRE P	14	80	Aug 16, 1989				
SRE R	3	1	May 1, 1988				
SRE R	4	1	Jun 1, 1988				
SRE R	5	1	Jul 1, 1988				
SRE R	6	2	Sep 1, 1988				
SRE R	7	2	Oct 1, 1988				
SRE R	8	1	Nov 8, 1988				
SRE R	9	1	May 1, 1989				

Table A.2 List of data in the Oil Spill Project archives (continued).

Project Code	CID	Pages	First Day of Collection
Reef Flat Communities - Gastropods			
SNA D	2	17	Nov 1, 1982
SNA D	3	17	Aug 31, 1986
SNA D	4	4	Aug 17, 1986
SNA D	5	13	Oct 1, 1986
SNA D	6	17	Feb 17, 1987
SNA D	7	12	May 17, 1987
SNA D	8	12	Aug 10, 1987
SNA D	10	17	Jan 29, 1988
SNA D	11	13	May 11, 1988
SNA D	12	13	Jul 27, 1988
SNA D	13	12	Nov 24, 1988
SNA D	14	13	Feb 8, 1989
SNA D	15	13	May 16, 1989
SNA S	2	17	Nov 1, 1982
SNA S	3	22	Aug 1, 1986
SNA S	4	2	Sep 1, 1986
SNA S	5	8	Nov 1, 1986
SNA S	6	26	Feb 17, 1987
SNA S	7	14	May 17, 1987
SNA S	8	14	Aug 18, 1987
SNA S	10	18	Jan 29, 1988
SNA S	11	11	May 11, 1988
SNA S	12	18	Jul 27, 1988
SNA S	13	16	Nov 24, 1988
SNA S	14	14	Feb 8, 1989
SNA S	15	20	May 16, 1989
Total pages			5,921

A.3 Description of Hardware and Software Used

A.3.1 Hardware

Microcomputers have provided an efficient and cost-effective way to provide computing facilities for the project. The project uses 13 microcomputers, ranging in capability from those with an 80286 microprocessor and a 20 megabyte hard disk to one with an 80386 microprocessor, a 300 megabyte hard disk, and a dual 44 megabyte Bernoulli box. The total cost of these computers, including software and peripherals, was less than a third that of a minicomputer of comparable abilities (i.e., a VAX 8350, at a price quoted to STRI in 1987).

The smaller computers are used primarily for data entry and word processing, while the larger computers are used for graphics, extracting and rearranging data, and maintaining the archives. Statistics are done about equally on the larger and smaller computers.

The speed of these computers is adequate for most of our needs. Foxplus, the main database program used for this project, will index a 15 megabyte file with 250,000 records in just under six minutes. Most of the graphics figures take from a few seconds to a minute to come on the screen; the more complicated figures may take two minutes to plot. There are some bottlenecks, however. The crosstabulation program may take up to 45 minutes to process a two megabyte file. The reef flat subproject (Chapter 3) routinely works with files as large as 12 megabytes, and some complex queries with Rbase can take up to an hour. Some repeated measures MANOVA's with missing values need more memory than the statistics programs can use at present in a microcomputer. A version of BMDP that can take advantage of more memory in a microcomputer is scheduled to be ready next year.

A.3.2 Software

Foxplus is used as the primary database program because it uses files that can be easily imported into other programs, has a flexible programming language, and provides quick and easy access to the data. Foxplus is an almost exact clone of Dbase, the database used by the majority of the business users of microcomputers. Its major advantage over Dbase is that it is substantially faster and cheaper. The lack of suitable data management capabilities in the major scientific statistics packages (SAS, SPSS, BMDP) makes the use of a business-oriented database such as Foxplus necessary.

Because of Dbase's widespread use, nearly all major statistics and database programs, and many graphics programs, can import Dbase files directly without having to first translate them to ASCII files. This saves a substantial amount of time because files are constantly being moved from one program to another for analysis.

The programming language allows command files to be written to enter and manage the data, greatly simplifying the many repetitive tasks needed for this type

of work. More important, the use of command files insures that the same procedures are followed each time so that the reliability of the data is maintained. When data are manipulated during analysis, the use of the programming language rather than interactive manipulation through menus provides documentation of what has been done to the data. Foxplus also provides an easy way to look at the data directly on the monitor and quickly select particular lines and variables to use. Most major statistics programs do not provide this ability.

Foxplus suffers several minor disadvantages. It was designed primarily for business use where the requirements for database management are somewhat different than those for environmental monitoring projects. Data entry for scientific use is often episodic and intensive rather than occurring at random during the day as various financial transactions occur. As a result, Foxplus is excellent for entering data where each line has a large amount of changing information, such as names and addresses, but is poor for entering data where each line contains much redundant identifying data, such as site and transect names, and has only one or two entries that change on each line. This deficiency appears to be common to all microcomputer database programs but can be remedied with a macro program such as Superkey.

Other software used in the project includes a variety of statistics, graphics, database, word processing and utility programs. This wide selection of programs is necessary for two reasons. First, it is rare that any one statistics or graphics program does everything that is needed. SPSS, for example, has excellent summary statistics, but is not as strong with multivariate statistics as BMDP. Rbase provides an interface with some SQL (Structured Query Language) commands that Foxplus does not provide and that are very useful for some kinds of analysis. Second, scientists who are already familiar with one program are reluctant to learn another. Indeed, it usually saves money to buy a program they are already familiar with rather than retrain them to use another.

A.4 Structure of the Data Management Section

Data are initially written on data sheets, entered into a data entry file, transferred to storage in a raw data file, and transformed into analysis files. They are then returned to the project scientist to work with or to request analysis using various statistics and graphics programs (Figure A.1).

A.4.1 Data Sheets

Data sheets are used for all the projects (see Figure A.2). Data that are similar for all projects, such as site name and collection date, are in the same place and have the same name on all data sheets. The arrangement of the remainder of the data sheet depends on the requirements of the individual project. An effective data sheet is arranged so that data recording in the field is quick, simple and error free. The way that the data are entered into the computer can often be adjusted to accommodate ease of data collection. By standardizing the data sheets where

MGF-L (Litter fall)

GALETA OIL SPILL
MANGROVE FOREST STUDY

CID _____

CDate _____

SDate _____
Weighed by _____

Site _____ N. _____

Tag_N	Sp	Leaves		Stipules		Wood	Debris	Trap	Trap
		Wt gm	N	Wt gm	N	Wt gm			
A		<input type="checkbox"/>	<input type="checkbox"/>
B		<input type="checkbox"/>	<input type="checkbox"/>
C		<input type="checkbox"/>	<input type="checkbox"/>

Tag_N	Sp	Repro.Parts		Buds		Flowers		Fruit		F-expend		Hypocotyls		
		Wt gm	N	N	Rem	N	Rem	N	Rem	N	Rem	N	Rem	Wt gm
A		.												.
B		.												.
C		.												.

Tag_N	Sp	Leaves		Stipules		Wood	Debris	Trap	Trap
		Wt gm	N	Wt gm	N	Wt gm			
A		<input type="checkbox"/>	<input type="checkbox"/>
B		<input type="checkbox"/>	<input type="checkbox"/>
C		<input type="checkbox"/>	<input type="checkbox"/>

Tag_N	Sp	Repro.Parts		Buds		Flowers		Fruit		F-expend		Hypocotyls		
		Wt gm	N	N	Rem	N	Rem	N	Rem	N	Rem	N	Rem	Wt gm
A		.												.
B		.												.
C		.												.

COMMENTS:

Figure A.2 A sample data sheet from the Mangrove Forests Project.

this length is that they can usually be easily deciphered by others familiar with the taxa under study when they are used in the printout of statistical analyses.

It is not uncommon for synonyms to be assigned to a single taxon. Rather than update the original entry in the data file (it would then not match what is written on the data sheet) a second code, called the present abbreviation (presabbr), is used. Then it is only necessary to update the species file. Both the data abbreviation and the present abbreviation are kept in the analysis files, and it is very easy to use the former to look up the proper code for the latter whenever taxa are combined. If a mistake is made in combining two taxa, it is easily corrected. Further abbreviations are used to classify the species into higher taxa. A program has been written to insert these codes into the analysis file when desired.

A.4.3 The Data Entry Program

A single data entry program was written in Dbase with Superkey macros and can be quickly adapted to any of the data formats required by the project. The program consists of a shell of 650 lines of code to which a very limited number of lines are added to accommodate each different data file. This kind of program was possible because the database fields, or variables, with a common function in each study, such as site and species name and collection date, have identical formats. The remaining fields in the database are usually some measure of abundance or size of an organism or taxonomic group. To adapt the program to a new data set takes a few hours to a day for more complicated files.

The data entry program is designed to be used after only a few minutes of instruction to a person otherwise inexperienced in computer use. Once the data entry routine is learned for one substudy, it is easy to enter data for any of the others. The main menu for the program can be obtained by pushing only two keys after turning on the computer. From the main menu, a single key will find the end of the file, list several of the previous lines of data, if any, and position the cursor for the next entry. Other keys will allow comments to be added to a text file, a backup to be made to a floppy disk, or new species to be added to the species list.

The data are entered one line at a time, with previously entered data scrolling off the top of the screen. Data that do not change frequently from one line to the next, such as site name, are copied automatically to each new line, and the cursor is automatically positioned for entering the new data for each line. When a species name is to be entered, a short one- to three-letter data entry code that is later substituted for a longer name is used. If a new species is encountered, a single keystroke allows the data enterer to add it to the species list, along with the data entry code. The program will check that the name and code are not already in use before proceeding.

A.4.4 Verifying Entered Data

After the data are entered, another keystroke runs a data screening program that will check for range and spelling errors and do other checking particular to each project. For example, if percent cover in a quadrat is being recorded, it will check that each quadrat adds up to 100. The program will also print lists of each different entry for fields with a limited number of values, such as site names. If a name is entered wrong, it will be easily recognized. The species or taxon abbreviation, called the data abbreviation, that corresponds to the data entry code is looked up on the species list and inserted into the file. When the screening program is finished, the version number in the file is incremented by one. Any errors found by the screening program that are other than data entry errors are referred back to the scientist for correction.

The screened file is then checked by the data enterer. First, the errors found by the computer are corrected, then another person reads the data to the data enterer who checks each entry. At this time, the species abbreviation, which had been entered by the computer to match the data entry code, appears on the screen and should match closely what is written on the data sheet. When the visual checking is done, the screening program is run again to ensure that no more errors have been entered. The version number is incremented each time the data screening program is run, and when the data enterer is satisfied that the data are entered accurately, the latest version number is noted in the verification file with his or her initials.

A.4.5 Speed of Data Entry

The Mangrove Roots Project provides a good example of the speed of data entry. Every three months, three sets of biological data totaling about 260 pages, and an additional 20 to 40 pages of physical data, including salinity, water temperature, water depth, and water movement, are received. Each line of the biological data includes an entry for species and percent cover that changes with each line, as well as several other fields, such as root condition and site, that change less frequently. The species list has 192 names currently in use. The physical data consist of two to four smaller files. In all there are approximately 20,000 lines of data to enter. This task takes one person 40 hours for the initial entry and then two persons working together 20 hours to check the data, update the species list, and run the programs for the reports.

A.4.6 Error Rate for Data Entry

All of the data for one of the projects were checked a third time after going through the above process of computer and data enterer checking. Out of 36,885 records entered after the data entry program was initiated, 33 had errors, for a 0.089 % error rate. Each record had a short character field and a five digit numerical

field that changed with each record, in addition to several identification fields. Of these errors, 24 could have been avoided by more careful data entry, and a small change in the data checking process has been initiated to further reduce this error rate. The remaining nine errors resulted from poorly written numbers, miscoding by the data enterer, loss of digits by photocopying of the data sheets, and an error in computer processing. None of these errors was of the type that could have seriously affected the results because the computer checking had already found major errors and omissions. There were, however, a number of corrections to be made to the files because of changes in site names, assigning the same name to two different types of cover in different localities, correcting a coding system that was in the process of development while the data were being collected, and filling in data that were not originally recorded on the data sheets. Some of these were unavoidable, but these types of changes, which can affect many records and which require separate command files to correct in a systematic manner, took much time to complete.

By far the most time was spent in assimilating and correcting older files that had been collected before the oil spill or before use of the data entry program started. Many errors occurred here because the data sheets were not organized properly to facilitate comparison with the entered data and because they were too cramped for easy legibility.

Much time can be saved by a careful and thorough attention to setting up the data sheets and data entry format before any data are collected.

A.4.7 Documentation of the Data Correction Process

The last printout of the data screening program containing summary lists of numbers of records of different types is kept with the data, and a copy is given to the Scientist-in-Charge. If available, simple summaries of the data are also sent to the Scientist-in-Charge. These verify that all of the data have been entered and that they have been carefully checked. A version number is included in each data file and kept, where appropriate, in subsequent files made from it. This number is incremented whenever a change is made to the file. A log is kept in the raw data directory that records the change, including what was done, who did it, and when, along with the new version number. This verification file increases one's confidence that the data have been checked and that the data have not been corrupted by improper corrections. The version numbers also help avoid losing track of whether a given file is the most recently corrected and determine whether an analysis file was made before or after a critical error was found and corrected.

A.4.8 Storing the Data

For each data file, programs have been written that transform it into a format suitable for analysis. Usually all that is required is the removal of a few fields needed to assist data entry and the addition of the present species abbreviation. Other changes involve translating measurements taken in feet to meters and

subtracting a tare from weights measurements. The project computer files are kept in a form easily accessible to anyone wishing to work with the data. The data files are written as Dbase files and all of the text files are written as ASCII files. Both types of files can be read and used by a wide variety of other programs, thus facilitating easy access to the files by other researchers, now and in the future. Each project has its own directory with the same name as the three letter study identification used to label the project in all other phases of the project. Within each of these directories are subdirectories. The subdirectories have uniform names, each prefaced with the one-letter code identifying each substudy. Fields within each file that are the same for all of the different studies have the same name. Again, this makes finding and using the files simple for other users.

Because of the volatility of magnetic media, a regular system of backing up the data is required. Two backup cartridges are kept for each Bernoulli cartridge, and one of these is kept in a separate location. Since the beginning of the project, approximately eight Bernoulli cartridges and one hard disk have lost all or part of their data, yet no large data files and only a few smaller files have been lost.

A.4.9 Reports to the Investigator

For some of the projects, in particular the Mangrove Roots and the Reef Flat Gastropods studies, a series of reports have been designed that are sent to the investigator after each collecting trip. These include means, standard deviations and counts of major groups of organisms as well as histograms of size frequency distributions. They serve both to inform the investigator of the state of the study area and to provide checks on the accuracy of the data entry. These reports are produced by running several Foxplus and SPSS programs after the data are entered and take only a few minutes of the data enterer's time.

A.4.10 Data Analysis

Statistical or graphical analysis is often a two-step process. The first step is transforming the data into a form usable by the graphics or statistics package, and the second step is actually producing the graph or running the statistics program. Two programs are available to assist with the first step. The first one, a crosstabulation program written in Dbase by the Data Manager, transposes a data file where species names are found in one column to one where the names appear at the head of separate columns. The same program can be used in a similar way to create separate columns for collection number, year, or any field with a discrete number of values. The second is the "Aggregate" program in SPSS. This takes a group of records, or lines, in a data file, calculates summary statistics, such as mean or variance, for some variable in the data, and produces a new file that includes these newly calculated values. Because SPSS can read and write Dbase files, it is very easy to convert a Dbase file into another Dbase file using the SPSS "Aggregate" procedure.

A.5 Conclusion

All data that have been collected have been entered and checked and are in good order. The system for managing new data and for integrating new projects into the study has been set up and is working smoothly. The two assistants have been entering the data, producing graphs, maintaining the management files, and have learned some of the programming skills necessary to help with setting up new data files. They have also been working under the direction of the scientists to manipulate files and produce simple summary reports.

Appendix B

Project Management

Project Management

Brian D. Keller

Smithsonian Tropical Research Institute

B.1 Facilities

Upon accepting this contract, the Smithsonian Tropical Research Institute (STRI) needed to provide offices and laboratories for a relatively large group of scientists and technicians. The Galeta Marine Laboratory was far too small to house the project. The possibility of expanding this facility was rejected because of important logistical and administrative problems associated with operating a relatively isolated laboratory. For example, the driving time between Galeta and STRI's administrative offices at Tivoli is over 1.5 hours (see Figure B.1). Also, the nearby city of Colón lacks most of the supplies available in Panama City. Existing facilities at the Naos Marine Laboratory on the Pacific side of Panama were fully occupied and could not accommodate the project. However, STRI was able to renovate a nearby unoccupied building at Naos obtained through an interagency transfer from the U.S. military, the "Surfside Theater" (Figure B.2).

Renovation of a section of this building was a major undertaking for STRI. It included constructing interior walls, installing flooring and false ceilings, replacing windows, and rewiring the building, including a separate circuit exclusively for computers. Work started in April 1987 and was completed enough by the end of August for staff to start to set-up and use laboratories and offices.

This new facility covers 520 square meters. There are two large laboratories, an x-ray room and photographic dark room, an instruments room, a specimens room, a computer room, a conference room, six offices, and a reception area. Support for equipping the building came from both STRI and the Minerals Management Service (MMS).

The laboratory for corals and the x-ray and photographic facilities are described in the chapter on subtidal reef corals (Chapter 5).

The other laboratory is shared by the reef flat and subtidal seagrass projects (Figures B.3 and B.4). It is equipped with a fume hood, magnifying lenses, dissecting stereomicroscopes, and a compound microscope. It is used mainly to pick and identify organisms from samples of reef-flat algal turf, subtidal seagrass sediments, and seagrass epifauna.

The instruments room (Figure B.5) contains drying ovens, two top-loading balances, and an analytical balance. It is used mainly to dry and weigh samples or specimens.

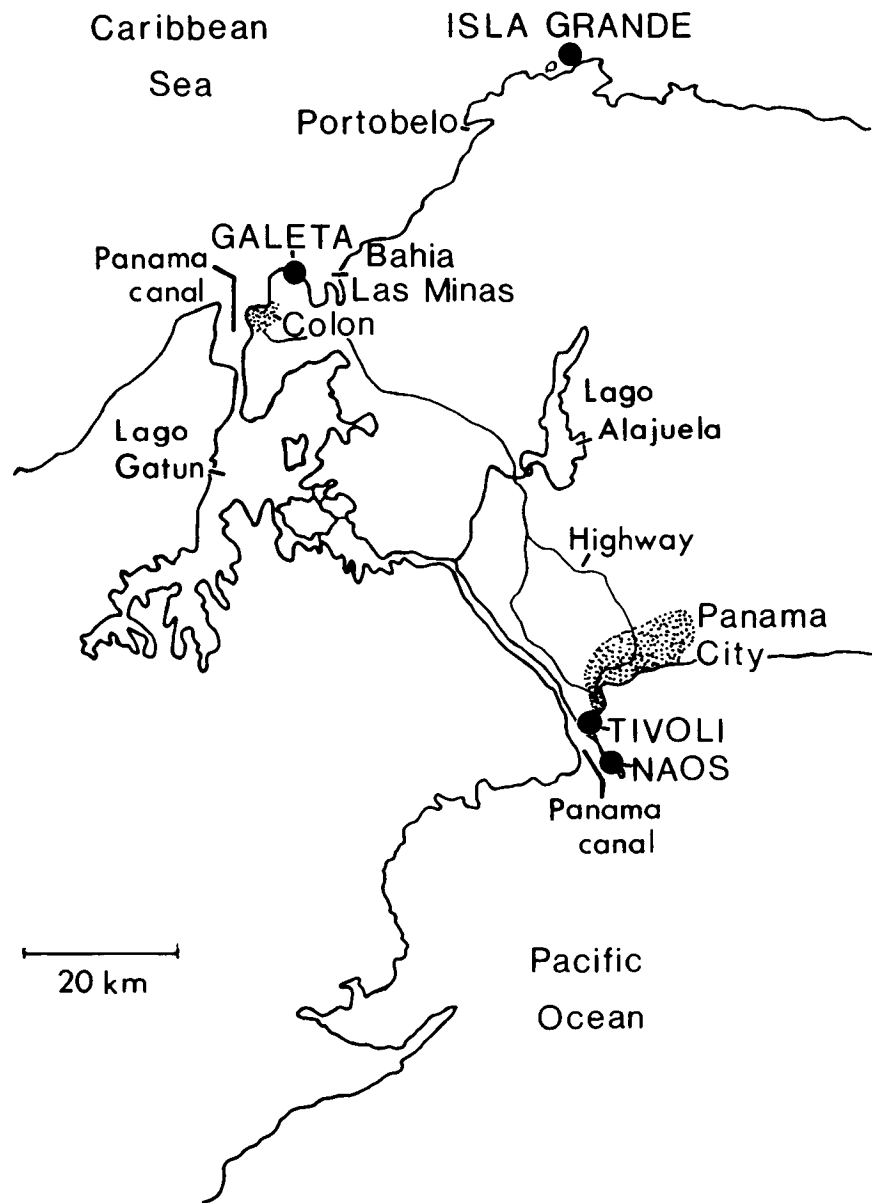


Figure B.1 Map of central Panama, showing the locations of the STRI marine laboratories at Naos and Galeta and the administrative offices at Tivoli.



Figure B.2

The Surfside Theater, Naos Marine Laboratory. The section of the building renovated for the Oil Spill Project is on the ground floor, proximal end, for more than half the length of the building (as far as the air conditioners extend).



Figure B.3

Section of the laboratory used mainly for studies of reef-flat and subtidal seagrass communities. A technician (Esther Jaén) is picking organisms from a sample of reef-flat *Laurencia* turf, using a stereomicroscope. The fume hood is in the left background.



Figure B.4

The other side of the laboratory shown in Figure B.3. Three technicians (Victoria Batista, Digna Matías, and Marco Díaz) are picking organisms from samples of seagrass communities using magnifying lenses.

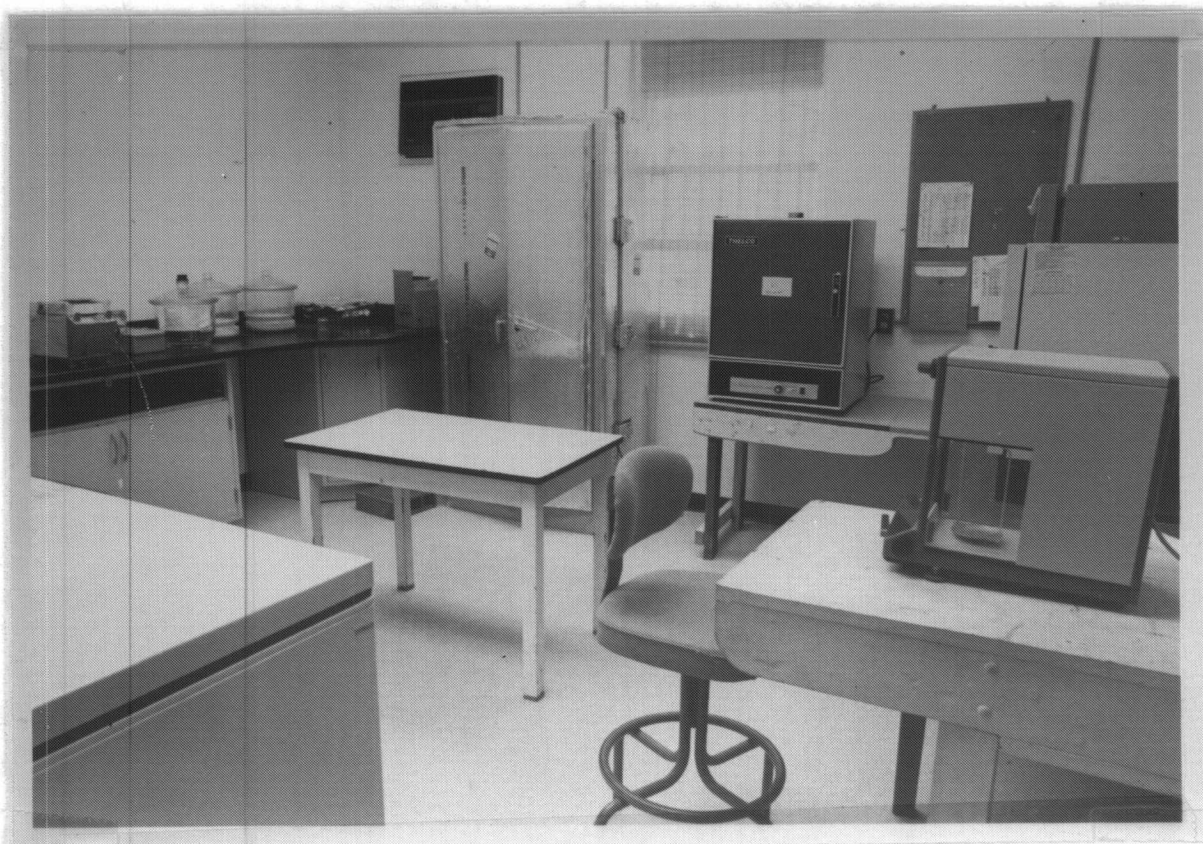


Figure B.5 The Instruments Room, showing the analytical balance (foreground), drying ovens and cabinet (background), and a top-loading balance (left).

The computer room (Figure B.6) contains a "386" microcomputer with a 70-Mb fixed disk, a "286" microcomputer with a 20-Mb fixed disk, a "286" microcomputer with a 70-Mb fixed disk, a laser printer, and a pen-and-ink computer-driven plotter. The equipment in this room is used mainly to enter and analyze data and to prepare computerized graphics for reports and manuscripts. A second room is equipped with a "386" microcomputer with a 300-Mb fixed disk and a "286" microcomputer with a 40-Mb fixed disk. These computers are used primarily for analysis of data (Appendix A).

The conference room (Figure B.7) accommodates various-sized meetings, and serves as a small lecture hall for groups of visitors interested in findings of the project, for example, an ecology class from the University of Panama.

The renovated section of the Surfside Theater has provided the project with needed laboratories and offices. It functions as the main center of operations, including coordination of field activities, interfacing with STRI support services and administration, and all aspects of communication.

The Galeta Marine Laboratory is the focal point of field work. There are three 15-foot Boston Whaler boats, which are used extensively. A 22-foot Mako boat was purchased because the small boats are not safe in rough sea conditions and cannot carry large loads.

Diving is used extensively for field work on subtidal reef corals, and the Galeta Marine Laboratory is equipped with an air compressor for SCUBA. The laboratory has a flowing seawater system and a variety of research equipment, including a dissecting microscope, a compound microscope, a top-loading balance, a drying oven, and a microcomputer. Housing is located a few kilometers away from the laboratory.

A Smithsonian Institution Environmental Sciences Program has operated at the Galeta Marine Laboratory since 1972, with various instruments and recorders as described in the chapter on physical environmental monitoring (Chapter 10).

There is daily van transportation between Tivoli and Galeta. There are two field vehicles, a 4x4 truck and a 4x4 jeep. The project also has a small van. Nevertheless, with so many simultaneously active field studies, vehicles are nearly always fully scheduled.

B.2 Operations

The scientists, technicians, and other staff comprising the project are listed in the preface. Each study is conducted largely independently, using the facilities described above. Findings are shared at staff meetings and at annual meetings with a Scientific Review Board (four or five members; see Figure B.7).

Since June 1987, a few months after the project began, Panama has been in a state of political, social, and economic turmoil. This study has suffered some brief interruptions because of these circumstances, particularly during the U.S. invasion of Panama in December 1989. However, for the most part work has proceeded remarkably smoothly.

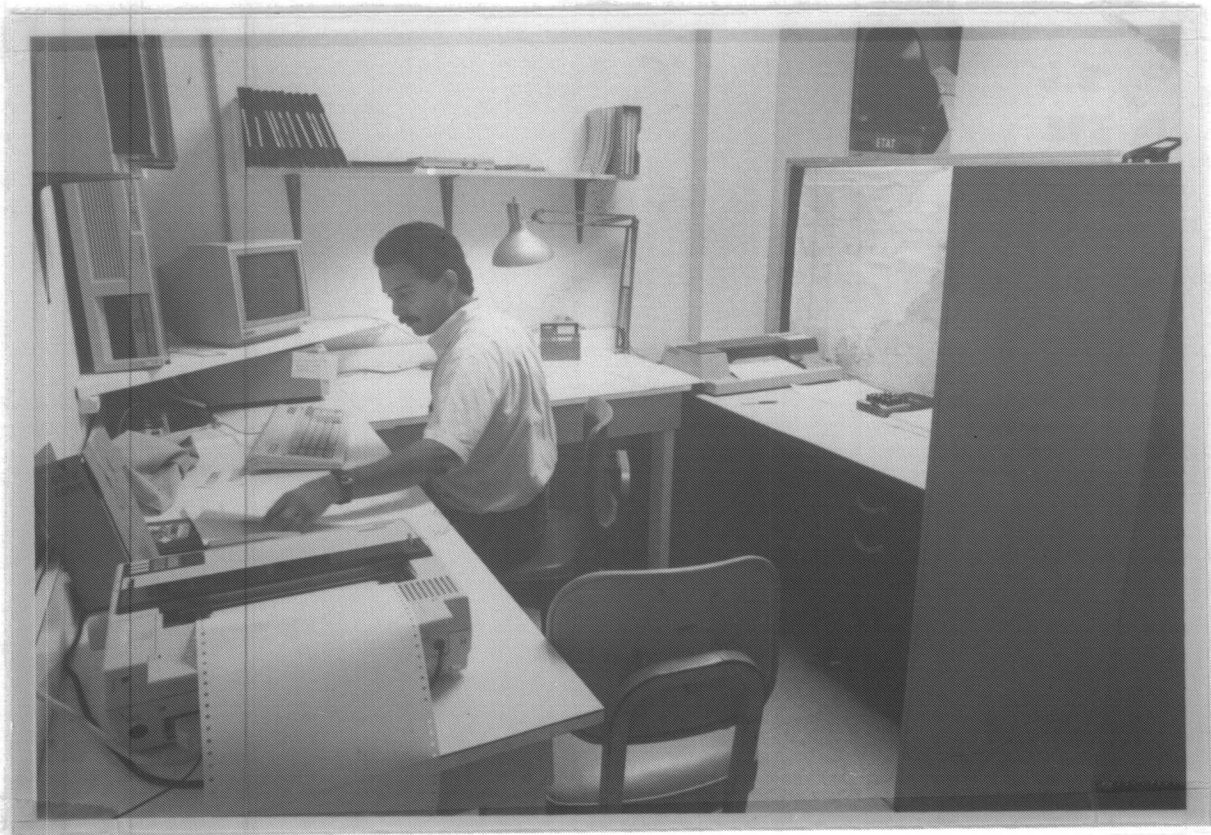


Figure B.6

The Computer Room, showing a technician (Gabriel Jácome) at a "386" microcomputer. The pen-and-ink plotter can be seen in the left background. The "286" microcomputers are in a different section of the room.

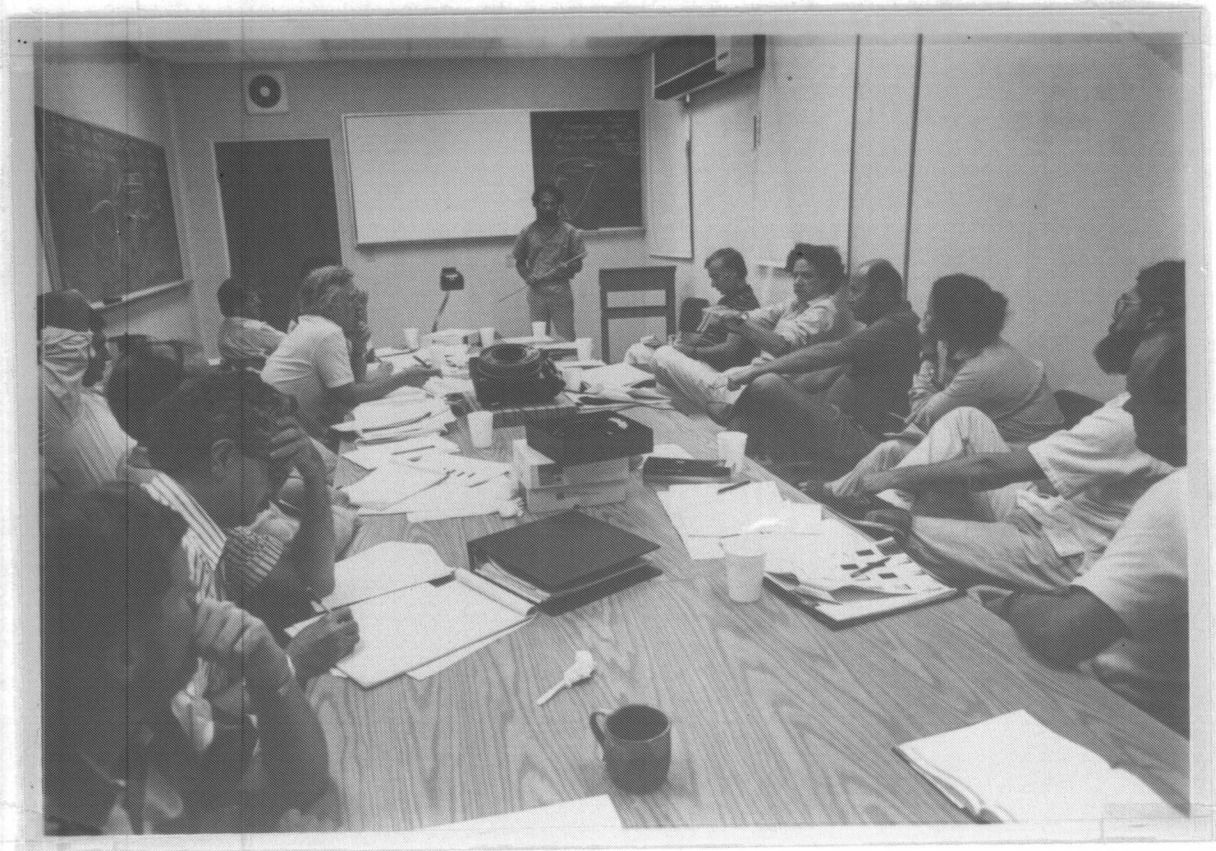


Figure B.7

The Conference Room, which can accommodate more than 20 people. This photograph was taken during the meeting of the Scientific Review Board in November 1987. Going around the table from left to right are: Karl Kaufmann, Robert Carney (Scientific Review Board), Richard Dodge (SRB), Anthony Knap (Bermuda Biological Station for Research), Roger Green (SRB), John Cubit, Brian Keller, Héctor Guzmán (standing), Yossi Loya (SRB), Jeremy Jackson, Murray Brown (MMS), Sally Levings, Stephen Garrity, and Michael Marshall [Gail Irvine (MMS) cannot be seen in this view].

Appendix C

**The Reef-Flat Sub-Project: Sessile Biota, Infauna,
and Sea Urchins on Intertidal Flats**

Appendix C Contents:

- Table C.1 Abbreviations for major type categories.
- Table C.2 Abbreviations for specific type categories with cross-references to major type categories.
- Table C.3 Mean percent primary cover for major type categories in transects at Punta Galeta, 1983 to 1988.
- Table C.4 Mean percent primary cover for specific type categories in transects at Punta Galeta, 1983 to 1988.
- Table C.5 Mean percent secondary cover for specific type categories in transects at Punta Galeta, 1983 to 1988.

Table C.1 Abbreviations for major type categories.

cni	All soft-bodied cnidarians
cnical	Calcareous cnidarians
corb	Branching (articulated) coralline algae
corc	Crustose coralline algae
fleshyc	Fleshy (non-calcareous) crustose algae
fleshy	All erect fleshy algae except <i>Laurencia papillosa</i>
lfleshy	<i>Laurencia papillosa</i>
grass	Seagrasses (mainly <i>Thalassia testudinum</i>)
grnc	All calcareous green algae
mic	Microalgae
spo	Sponges
ssinv	Sessile invertebrates other than sponges and cnidarians
sub	Substrata: loose coral rubble, sand, bare rock.

Table C.2 Abbreviations for specific type categories with cross-references to major type categories.

Primary/ Secondary Cover	Type	Genus	Species	Description
ac	fleshy	<i>Acanthophora</i>	<i>spicifera</i>	
amp	corb	<i>Amphiroa</i>	sp.	
ana	grnc	<i>Anadyomene</i>	<i>stellata</i>	
anem	cni			Anemone
brcu	fleshyc			Brown Crust
brspo	spo			Brown Sponge
caulr	fleshy	<i>Caulerpa</i>	<i>racemosa</i>	
cauls	fleshy	<i>Caulerpa</i>	<i>sertularioides</i>	
cent	fleshy	<i>Centroceras</i>	sp.	
cor	corc			Crustose coralline algae
cw	corc	<i>Neogoniolithon</i>	cf. <i>westindianum</i>	
cx	mic	<i>Calothrix</i>	sp.	
dasya	fleshy	<i>Dasya</i>	sp.	
ds	grnc	<i>Dictyosphaeria</i>	sp.	
dx	fleshy	<i>Dichothrix</i>	sp.	
dy	fleshy	<i>Dictyota</i>	sp.	
er	cni	<i>Erythropodium</i>	sp.	
galax	corb	<i>Galaxaura</i>	sp.	
gel	fleshy	<i>Gelidiella</i>	sp.	
grfuz	mic			Green Fuzz
h	grnc	<i>Halimeda</i>	<i>opuntia</i>	
hp	fleshy	<i>Hypnea</i>	sp.	
ht	grnc	<i>Halimeda</i>	<i>tuna</i>	
is	cni	<i>Isaurus</i>	sp.	
jan	lfleshy	<i>Jania</i>	sp.	
jlm	lfleshy	<i>Jania/Laurencia</i>	(<i>L. papillosa</i>)	Two species matrix
l	fleshy	<i>Laurencia</i>	<i>papillosa</i>	
lint	fleshy	<i>Laurencia</i>	<i>intricata</i>	
lob	fleshy	<i>Laurencia</i>	<i>obtusa</i>	
mi	cnical	<i>Millepora</i>	sp.	
penici	grnc	<i>Penicillus</i>	sp.	

Table C.2 Abbreviations for specific type categories with cross-references to major type categories (continued).

peys	fleshyc			<i>Peyssonellia</i> -like species
por	cnical	<i>Porites</i>	sp.	
pt	cni	<i>Palythoa</i>	sp.	
r	sub			rock
rfuz	mic			Red Fuzz
rub	sub			rubble
s	sub			sand
sarg	fleshy	<i>Sargassum</i>	sp.	
scz	mic			scuz = microalgae
spo	spo			Sponge
spy	fleshy	<i>Spyridia</i>	sp.	
struv	grnc	<i>Struvea</i>	<i>anastomosans</i>	
sx	mic	<i>Schizothrix</i>	sp.	
th	grass	<i>Thalassia</i>	<i>testudinum</i>	
tri		<i>Trididemnum</i>	sp.	
un				unknown
val	fleshy	<i>Valonia</i>	sp.	
zs	cni	<i>Zoanthus</i>	<i>sociatus</i>	
dicho	cnical	<i>Dichocoenia</i>	sp.	
avr	fleshy	<i>Avrainvillea</i>	sp.	
ag	cnical	<i>Agaricia</i>	spp.	
caulc	fleshy	<i>Caulerpa</i>	<i>cupressoides</i>	
grac	fleshy	<i>Gracilaria</i>	spp.	
bc				bleached coralline
gf		<i>Cladophora?</i>		
foram	invsess	<i>Homotrema</i>	spp.	

Table C.3 Mean percent cover for major type categories (continued).

Type	Year and Month (as yrmo)			
	8806	8809	8904	8906
cni	3.78	3.46	4.55	5.00
cnical	0.00	0.00	0.00	0.13
corb	0.83	0.13	0.83	0.32
corc	10.64	8.08	9.10	15.83
fleshy	0.13	0.00	0.71	1.28
fleshyc	0.13	0.00	0.00	0.32
grass	1.15	1.09	2.37	1.54
grnc	5.26	1.67	2.95	4.74
lfleshy	3.65	5.64	58.27	45.06
mic	62.69	73.59	16.67	18.59
spo	0.13	0.13	0.00	0.06
ssinv	0.00	0.00	0.06	0.00
sub	11.60	6.22	4.49	7.12
totals	100.00	100.00	100.00	100.00

Table C.4 Mean percent primary cover for specific type categories (continued).

Primary cover	Year and Month (as yrmo)					
	8612	8703	8706	8709	8801	8804
ac	3.27	4.36	2.24	0.58	2.43	2.76
amp	0.00	0.06	0.00	0.00	0.00	0.13
anem	0.51	0.00	0.38	0.38	0.64	0.13
bcor	0.06	0.00	0.00	0.00	0.00	0.00
brcu	0.00	0.00	0.06	0.00	0.00	0.00
brspo	0.00	0.19	0.00	0.00	0.00	0.13
bscz	0.00	0.00	0.00	0.13	0.00	0.00
caulr	0.00	0.58	0.00	0.00	0.13	0.00
cauls	0.00	0.19	0.06	0.19	0.00	0.06
cor	8.01	12.37	13.53	11.03	7.04	12.37
cw	1.22	1.86	3.53	1.09	0.83	2.24
ds	0.19	0.00	0.13	0.00	0.00	0.00
dy	0.06	0.19	0.00	0.06	0.00	0.00
er	0.00	0.00	0.13	0.00	0.00	0.00
foram	0.00	0.00	0.00	0.00	0.06	0.06
galax	0.13	0.00	0.00	0.00	0.00	0.00
gel	0.19	0.06	0.19	0.00	0.00	0.45
grac	0.00	0.00	0.00	0.00	0.00	0.06
h	3.33	5.64	5.26	6.60	10.05	10.71
hp	0.00	0.26	0.00	0.00	0.06	0.19
ht	0.00	0.00	0.00	0.00	0.00	0.32
is	0.00	0.06	0.00	0.00	0.00	0.06
jan	1.28	5.06	2.50	1.35	2.05	5.06
jlm	0.00	0.00	0.00	1.86	1.02	5.26
l	53.21	42.31	30.13	58.59	50.75	25.19
lint	0.38	0.00	0.00	0.00	0.13	0.06
lob	0.06	0.00	0.00	0.00	0.13	0.00
mi	0.00	0.06	0.19	0.71	0.77	0.45
penici	0.00	0.00	0.00	0.00	0.06	0.06
peys	0.13	0.51	0.38	0.26	0.13	0.00
pt	0.32	0.38	0.58	0.26	0.77	0.77
r	0.45	1.60	1.99	0.58	0.38	0.19
rub	1.73	2.18	1.67	0.58	0.70	3.01
s	6.28	6.09	6.41	1.15	5.57	8.21
scz	11.92	8.85	23.59	6.41	7.30	13.27
spy	0.00	0.00	0.00	0.00	0.00	0.06
sx	0.00	0.00	0.00	0.06	0.00	0.00
th	1.92	1.99	1.09	1.92	2.11	1.79

Table C.4 Mean percent primary cover for specific type categories (continued).

tri	0.00	0.06	0.00	0.00	0.00	0.00
val	0.00	0.00	0.00	0.00	0.00	0.06
zs	5.32	5.06	5.96	6.22	6.72	6.86
Totals	100.00	100.00	100.00	100.00	99.84	100.00
Primary cover	Year and Month (as yrmo)					
	8806	8809	8904	8906		
ac	0.00	0.00	0.26	1.28		
anem	0.32	0.00	0.26	0.45		
brclu	0.06	0.00	0.00	0.19		
brspo	0.13	0.13	0.00	0.06		
caulr	0.00	0.00	0.19	0.00		
cor	2.18	7.76	8.14	13.78		
corbl	7.69	0.00	0.00	0.00		
cw	0.77	0.32	0.96	2.05		
er	0.00	0.00	0.06	0.00		
foram	0.00	0.00	0.06	0.00		
gel	0.13	0.00	0.06	0.00		
gf	6.99	0.00	0.00	0.32		
h	5.19	1.47	2.82	4.49		
hp	0.00	0.00	0.13	0.00		
ht	0.06	0.19	0.13	0.26		
is	0.00	0.00	0.00	0.13		
jan	0.83	0.13	0.83	0.32		
jlm	0.06	0.00	0.13	0.45		
l	3.59	5.64	58.14	44.62		
lint	0.00	0.00	0.06	0.00		
ly	0.00	0.00	0.06	0.00		
mi	0.00	0.00	0.00	0.06		
peys	0.06	0.00	0.00	0.13		
porf	0.00	0.00	0.00	0.06		
pt	0.38	0.26	0.38	0.64		
r	0.00	0.77	1.15	2.69		
rfuz	0.00	0.06	0.13	0.51		
rub	2.63	0.90	0.19	1.47		
s	8.97	4.55	3.14	2.95		
scz	55.71	73.53	16.47	17.76		
th	1.15	1.09	2.37	1.54		
zs	3.08	3.21	3.85	3.78		
Totals	100.00	100.00	100.00	100.00		

Table C.5 Mean percent secondary cover for specific type categories in transects at Punta Galeta, 1983 to 1988.

Secondary cover	Year and Month (as yrmo)					
	8303	8304	8305	8306	8309	8310
ac	2.22	0.33	0.72	0.07	3.05	3.91
amp	1.63	1.57	0.52	0.26	0.33	0.38
cr	0.00	0.00	0.00	0.00	0.00	0.13
dy	1.05	0.65	0.07	0.07	0.00	0.00
gel	0.07	0.13	0.20	0.26	0.00	0.00
h	0.26	0.13	0.00	0.07	0.13	0.19
hp	0.26	0.39	0.20	0.00	0.07	0.13
l	1.31	1.37	1.44	1.18	1.85	0.32
th	0.07	0.26	0.33	0.33	0.13	0.26
Totals	6.99	4.84	3.46	2.22	5.56	5.32

Secondary cover	Year and Month (as yrmo)					
	8402	8403	8404	8405	8406	8407
ac	1.73	1.09	0.00	0.51	0.13	0.26
amp	0.06	0.13	0.00	0.32	0.13	0.64
dict	0.00	0.06	0.00	0.00	0.00	0.00
dy	0.13	0.26	0.06	0.06	0.00	0.19
gel	0.00	0.00	0.00	0.26	0.00	0.06
hp	0.58	0.32	0.00	0.00	0.00	0.00
jan	0.00	0.00	0.00	0.19	0.06	0.13
l	0.13	0.32	0.00	0.06	0.00	0.00
lyngbia	0.00	0.00	0.06	0.00	0.00	0.00
th	0.13	0.00	0.00	0.00	0.00	0.00
Totals	2.76	2.18	0.13	1.41	0.32	1.28

Table C.5 Mean secondary percent cover for specific type categories (continued).

Secondary cover	Year and Month (as yrmo)					
	8409	8410	8411	8412	8606	8609
ac	0.45	0.96	2.50	2.31	0.13	1.73
amp	0.96	1.79	0.64	0.13	0.00	0.00
dy	0.13	0.06	0.13	0.00	0.00	0.00
gel	0.13	0.00	0.00	0.13	0.00	0.00
h	0.00	0.00	0.00	0.06	0.00	0.00
hp	0.00	0.06	0.00	0.13	0.00	0.06
jan	0.06	0.26	0.13	0.00	0.06	0.38
l	0.19	0.00	0.00	0.00	0.13	0.00
scz	0.00	0.00	0.00	0.00	5.00	0.06
th	0.00	0.06	0.00	0.00	0.00	0.00
Totals	1.92	3.21	3.40	2.76	5.32	2.24

Secondary cover	Year and Month (as yrmo)					
	8612	8703	8706	8709	8801	8804
ac	0.00	1.15	0.00	6.73	2.88	1.47
cs	0.00	0.06	0.00	0.00	0.00	0.00
h	0.00	0.06	0.00	0.00	0.00	0.00
hp	0.00	0.06	0.00	0.00	0.06	0.00
jan	0.00	0.00	0.19	0.00	0.00	0.00
l	0.00	0.38	0.00	0.00	0.00	0.00
scz	0.83	0.00	0.51	0.00	0.06	2.76
Totals	0.83	1.73	0.71	6.73	3.01	4.23

Secondary cover	Year and Month (as yrmo)			
	8806	8809	8904	8906
ac	0.00	0.00	0.00	0.90
cor	0.00	0.00	0.06	0.00
gf	0.13	0.00	0.00	0.00
l	0.00	0.00	0.00	0.06
scz	0.26	0.06	2.44	1.79
Totals	0.38	0.06	2.50	2.76

Appendix D

Subtidal Reef Corals

Table D.1 Yearly mean growth rates (mm) from 1977 to 1986 for *Siderastrea siderea* at 11 reefs grouped by level of oiling in 1986 (none, moderate, and heavy). Standard errors in parentheses. Sample size for all means is five.

Yr	None				Moderate		Heavy				
	9	10	11	12	1	8	2	4	5	6	7
86	4.50 (0.35)	4.30 (0.56)	3.20 (0.41)	4.50 (0.96)	5.30 (0.41)	3.90 (0.46)	4.40 (0.48)	3.20 (0.34)	4.00 (0.35)	3.40 (0.46)	3.90 (0.78)
85	5.60 (0.29)	4.70 (0.64)	4.50 (0.69)	4.70 (0.94)	6.60 (0.48)	5.00 (0.69)	6.00 (0.45)	3.40 (0.90)	4.84 (1.39)	3.50 (0.55)	5.2 (0.51)
84	5.80 (0.20)	4.40 (0.66)	4.10 (0.37)	5.30 (1.20)	5.70 (0.54)	5.1 (0.87)	5.30 (1.01)	3.7 (0.89)	5.50 (1.24)	3.60 (0.46)	5.60 (0.95)
83	6.10 (0.29)	4.60 (1.04)	4.90 (0.64)	5.94 (1.24)	4.40 (0.48)	4.40 (0.29)	5.10 (1.01)	4.30 (0.37)	4.20 (0.75)	3.90 (0.64)	5.80 (0.64)
82	6.10 (0.19)	4.00 (0.81)	4.50 (0.50)	5.40 (0.48)	5.90 (0.83)	4.20 (0.46)	5.50 (0.84)	3.40 (0.60)	4.00 (0.69)	3.70 (0.25)	5.50 (0.45)
81	5.10 (0.37)	4.20 (0.80)	4.90 (0.40)	4.30 (0.44)	5.10 (0.62)	5.10 (0.33)	4.8 (0.66)	3.50 (0.67)	4.60 (0.70)	4.70 (1.36)	5.50 (1.00)
80	4.7 (0.30)	4.70 (0.98)	4.70 (0.62)	3.50 (0.35)	5.40 (0.62)	4.80 (0.56)	6.30 (1.24)	3.70 (0.62)	3.70 (0.51)	4.20 (0.82)	5.40 (1.03)
79	4.60 (0.37)	5.30 (1.01)	4.30 (0.46)	3.8 (0.58)	5.20 (0.60)	4.70 (0.49)	4.20 (0.75)	2.80 (0.20)	4.70 (0.82)	5.70 (1.11)	5.80 (0.66)
78	4.90 (0.58)	4.50 (0.79)	4.10 (0.58)	4.70 (0.78)	5.30 (0.37)	5.20 (0.60)	4.70 (0.93)	2.80 (0.46)	3.50 (1.04)	3.60 (0.29)	5.50 (0.92)
77	4.20 (0.20)	4.60 (0.76)	4.90 (0.24)	4.90 (0.43)	5.20 (0.60)	5.00 (0.65)	5.70 (0.78)	3.40 (0.43)	4.00 (0.32)	4.90 (0.62)	5.10 (0.97)

Table D.2 Yearly mean growth rates (mm) for *Montastrea annularis* at 8 reefs grouped by level of oiling in 1986 (none, moderate, and heavy). Standard errors in parentheses. Where not specified, sample size is five.

Yr	None				Moderate		Heavy	
	9	10	11	12	8	4	6	7
86	8.80 (2.13)	6.70 (1.27)	9.00 (2.08)	12.13 (1.31) N=4	7.60 (0.40)	5.38 (0.56) N=4	6.67 (0.88) N=3	8.25 (1.25) N=2
85	7.50 (1.55)	6.60 (1.35)	8.70 (2.35)	10.38 (1.28) N=4	8.40 (0.70)	6.88 (0.56) N=4	9.50 (1.15) N=3	7.75 (0.25) N=2
84	9.40 (1.48)	7.10 (1.41)	10.30 (2.01)	12.25 (1.90) N=4	9.50 (1.04)	9.13 (1.00) N=4	6.67 (1.74) N=3	6.00 (1.00) N=2
83	11.00 (2.08)	6.20 (1.11)	9.00 (1.79)	10.38 (1.41) N=4	10.20 (0.72)	9.38 (1.52) N=4	8.17 (0.93) N=3	8.50 (4.00) N=2
82	7.90 (1.62)	6.40 (0.83)	7.40 (1.43)	12.38 (1.77) N=4	8.30 (1.49)	8.13 (1.47) N=4	8.17 (0.93) N=3	8.50 (1.50) N=2
81	7.90 (1.70)	4.90 (0.76)	7.30 (1.33)	10.88 (1.70) N=4	8.10 (1.09)	8.38 (1.58) N=4	8.33 (2.32) N=3	8.50 (2.00) N=2
80	.20 (1.14)	6.80 (1.25)	9.00 (1.46)	11.75 (1.61) N=4	8.70 (1.14)	9.38 (1.25) N=4	9.00 (2.00) N=3	5.75 (0.75) N=2
79	6.30 (1.66)	7.00 (1.14)	10.10 (1.94)	12.50 (2.52) N=4	7.70 (1.22)	9.63 (2.26) N=4	8.67 (0.73) N=3	6.25 (0.25) N=2
78	8.20 (1.10)	6.60 (0.89)	9.70 (1.01)	12.75 (1.88) N=4	10.00 (1.39)	8.00 (1.46) N=4	7.50 (0.29) N=3	7.00 (1.00) N=2
77	8.90 (1.01)	6.80 (0.88)	10.40 (1.56)	14.25 (1.45) N=4	10.30 (0.93)	11.63 (2.53) N=4	7.67 (0.33) N=3	5.50 (0.00) N=1

Table D.3 Yearly mean growth rates (mm) for *Porites astreoides* at 10 reefs grouped by level of oiling in 1986 (none, moderate, and heavy). Standard errors in parentheses. Where not specified, sample size is five.

Yr	None				Moderate		Heavy			
	9	10	11	12	1	8	2	4	6	7
86	5.70 (0.51) N=4	5.00 (0.74) N=4	5.38 (0.24) N=4	4.90 (0.48)	4.80 (0.66)	5.00 (1.06)	3.80 (0.34)	3.40 (0.37)	4.90 (0.83)	3.90 (0.33)
85	5.80 (0.73) N=4	3.63 (0.43) N=4	4.63 (0.66) N=4	5.30 (0.75)	4.30 (0.41)	4.13 (0.24)	4.10 (0.37)	3.40 (0.37)	5.20 (0.64)	5.40 (0.58)
84	6.30 (1.50) N=4	4.00 (0.50) N=4	3.75 (0.25) N=4	5.90 (0.40)	3.9 (0.43)	5.25 (0.32)	5.40 (0.93)	3.60 (0.48)	5.50 (0.96)	3.80 (0.41)
83	5.10 (0.33) N=4	4.75 (0.52) N=4	5.38 (0.55) N=4	6.90 (0.60) N=4	4.60 (0.81)	4.63 (0.66)	4.50 (0.87)	4.00 (0.76)	6.30 (1.59)	4.30 (0.70)
82	4.40 (0.62) N=4	4.88 (0.24) N=4	5.88 (1.48) N=4	7.00 (0.79) N=4	4.70 (0.72) N=4	4.50 (0.87)	4.25 (1.11)	4.30 (0.49)	5.75 (0.43)	3.90 (0.29)
81	5.00 (0.42) N=4	5.00 (0.20) N=4	6.00 (0.74) N=4	6.30 (0.20) N=4	5.40 (0.90) N=4	5.63 (0.69) N=4	5.13 (1.33)	3.13 (0.31)	4.25 (0.32)	3.80 (0.54)
80	6.70 (0.56) N=4	5.13 (1.25) N=4	6.88 (1.25) N=4	6.00 (0.35) N=4	4.60 (0.70) N=4	4.75 (0.97) N=2	5.75 (0.66)	3.25 (0.60)	5.75 (1.75)	3.60 (0.24)
79	5.70 (0.54) N=4	3.88 (0.24) N=4	5.63 (1.64) N=4	5.20 (0.58) N=4	5.10 (0.29) N=4	5.25 (1.27) N=2	5.25 (0.66)	3.38 (0.43)	6.00 (2.00)	4.80 (0.64)
78	5.20 (0.58) N=4	4.63 (0.38) N=4	6.38 (1.16) N=4	6.13 (0.94) N=3	4.20 (0.46) N=4	4.17 (0.93) N=2	5.38 (0.77) N=3	3.17 (0.17)	5.00 (0.00) N=1	3.60 (0.24)
77	5.90 (0.46)	6.38 (0.90) N=4	5.38 (0.63) N=4	5.38 (0.97) N=3	4.13 (0.80) N=4	4.25 (0.25) N=2	4.38 (1.36) N=3	2.50 (0.00) N=1	---- ---- ----	3.67 (0.60) N=3

Table D.4 Yearly mean growth rates (mm) for *Diploria strigosa* at 8 reefs grouped by level of oiling in 1986 (none, moderate, and heavy). Standard errors in parentheses. Where not specified, sample size is five.

Yr	None				Moderate	Heavy		
	9	10	11	12	8	4	6	7
86	5.70 (0.20)	4.70 (0.56)	6.80 (0.58)	4.60 (0.53)	4.40 (0.87)	4.50 (0.82)	4.80 (0.82)	3.60 (0.58)
85	5.20 (0.82)	4.40 (0.75)	8.10 (1.09)	4.90 (0.62)	4.80 (0.87)	6.00 (1.30)	5.60 (1.37)	3.70 (0.46)
84	5.40 (0.73)	4.50 (0.42)	8.60 (0.86)	5.00 (0.57)	4.30 (0.64)	5.90 (1.25)	6.50 (2.02)	3.50 (0.45)
83	5.20 (0.64)	4.70 (0.34)	7.90 (0.94)	5.20 (0.56)	4.80 (1.27)	5.40 (1.02)	5.80 (1.25)	3.70 (0.30)
82	4.30 (0.75)	4.60 (0.40)	7.40 (0.91)	5.40 (1.18)	5.70 (0.93)	5.30 (0.34)	5.90 (1.81)	4.80 (0.98)
81	4.70 (0.49)	4.90 (0.46)	8.60 (0.93)	5.40 (0.89)	5.60 (1.02)	5.60 (0.43)	6.20 (1.75)	3.90 (0.43)
80	5.40 (0.91)	5.50 (0.65)	7.60 (0.81)	4.60 (0.43)	5.00 (0.89)	4.80 (0.60)	5.60 (1.04)	4.20 (0.75)
79	4.80 (0.54)	5.20 (0.56)	6.90 (0.62)	5.20 (0.73)	4.20 (0.34)	6.10 (0.89)	7.40 (1.98)	3.90 (0.70)
78	4.80 (0.86)	4.50 (0.42)	6.60 (0.71)	4.80 (0.34)	5.20 (0.41)	5.70 (0.80)	6.40 (1.43)	4.20 (0.80)
77	4.10 (0.78)	4.00 (0.42)	6.88 (1.26)	4.20 (0.46)	4.60 (0.78)	5.40 (0.73)	6.70 (1.49)	4.50 (0.71)
			N=4					

Appendix E

**Effects of the April 1986 Oil Spill at Isla Payardi
on the Epibiota of Mangrove (*Rhizophora mangle* L.) Roots**

Table E.1 Salinity (o/oo) and water temperature (°C).

Site	Date	Salinity		Temperature	
		Surface	1 m	Surface	1 m
A. Open Coast, Oiled					
Isla Droque Mangrove	15Dec1988	33.25	33.25	.	.
	16Feb1989	36.00	36.00	26.75	26.75
	08May1989	31.00	31.00	30.00	29.75
Mina Mangrove	15Dec1988	32.50	33.00	.	.
	12Feb1989	36.00	36.00	27.75	27.75
	08May1989	32.50	34.00	31.25	31.25
Peña Guapa Mangrove	15Dec1988	33.00	33.00	.	.
	13Feb1989	36.00	36.00	28.75	28.00
	08May1989	31.50	28.00	30.00	28.75
Punta Muerto Mangrove	15Dec1988	34.00	34.00	.	.
	17Feb1989	36.00	36.00	28.25	27.75
	08May1989	34.00	32.00	30.00	29.75
B. Open Coast, Unoiled					
Isla Lintón Mangrove	14Dec1988	34.00	34.0	.	.
	18Feb1989	36.00	36.0	28.25	28.00
	08May1989	30.00	32.0	34.50	31.50
María Soto Mangrove	14Dec1988	34.00	34.0	.	.
	20Feb1989	36.00	36.0	29.75	29.25
	08May1989	31.00	33.0	38.25	34.25
Isla del Padre Mangrove	14Dec1988	35.00	35.0	.	.
	18Feb1989	36.00	36.0	29.25	29.00
	08May1989	31.00	32.5	29.25	29.00
Portobelo Mangrove	14Dec1988	34.00	34.0	.	.
	20Feb1989	36.00	36.0	28.25	28.25
	08May1989	32.00	33.0	28.75	28.75

Table E.1 Salinity and water temperature (continued).**C. Channels and Lagoons, Oiled**

Largo Remo Channel West

29Dec1988	36.0	36.0	.	.
15Feb1989	36.0	36.0	.	.
08May1989	33.5	29.0	33.00	32.75

Payardi Channel East

28Dec1988	34.5	35.0	.	.
21Feb1989	35.0	35.5	30.75	30.25
08May1989	32.0	35.0	30.00	30.25

Payardi Channel South

28Dec1988	33.0	34.0	.	.
28Feb1989	34.5	35.0	29.25	29.00
08May1989	32.0	33.0	31.75	30.50

Samba Bonita Channel East

27Dec1988	34.0	34.5	.	.
17Feb1989	35.0	35.0	28.50	28.50
08May1989	28.5	32.0	29.50	29.50

Samba Bonita Channel South

27Dec1988	33.5	34.5	.	.
16Feb1989	35.0	34.0	28.75	28.75
08May1989	31.5	33.0	30.25	29.75

D. Channels and Lagoons, Newly Oiled

Hidden Channel

29Dec1988	28.00	31.00	.	.
16Feb1989	36.00	36.00	29.0	29.25
08May1989	29.00	33.00	30.5	30.75

E. Channels and Lagoons, Unoiled

Largo Remo Channel South

15Feb1989	36.0	36.0	28.25	28.25
08May1989	35.0	33.0	30.75	30.50

Margarita Channel North

29Dec1988	31.5	32.0	.	.
27Feb1989	34.0	34.5	27.75	27.75
08May1989	32.0	33.5	29.2	29.50

Margarita Channel South

29Dec1988	30.0	34.0	.	.
27Feb1989	34.0	34.0	27.00	27.00
08May1989	32.0	34.0	29.50	29.50

Table E.1 Salinity and water temperature (continued).

Samba Bonita Channel West					
	29Dec1988	32.0	34.0	.	.
	12Feb1989	36.0	36.0	29.00	28.75
	08May1989	37.0	33.0	30.75	31.00
F. Drainage Streams, Oiled					
Largo Remo River South					
	27Dec1988	34.0	34.0	.	.
	03Mar1989	37.5	38.0	29.00	29.50
	08May1989	31.0	31.5	32.25	31.25
Payardi River					
	28Dec1988	33.0	33.0	.	.
	25Feb1989	35.0	34.0	27.00	27.00
	08May1989	32.0	31.5	32.00	32.00
Punta Muerto River East					
	28Dec1988	34.0	34.0	.	.
	24Feb1989	35.0	35.0	28.50	28.50
	08May1989	31.5	31.0	32.00	30.75
Punta Muerto River West					
	28Dec1988	34.0	35.0	.	.
	21Feb1989	35.0	35.0	29.50	29.00
	08May1989	33.0	33.0	32.25	32.00
G. Drainage Streams, Unoiled					
Río Alejandro					
	28Dec1988	6.5	18.5	.	.
	24Feb1989	21.5	31.5	28.00	29.00
	08May1989	25.0	30.0	29.00	31.00
Hidden River					
	27Dec1988	6.5	22.0	.	.
	13Feb1989	27.0	32.5	26.50	26.75
	08May1989	21.5	28.0	29.75	31.00
Quebrada Las Mercedes					
	27Dec1988	8.5	27.5	.	.
	17Feb1989	19.0	33.0	28.00	28.00
	08May1989	27.0	33.5	30.00	31.00
Unnamed River					
	28Dec1988	7.0	24.0	.	.
	23Feb1989	9.0	32.0	28.25	29.50
	08May1989	14.0	33.5	29.25	31.25

. = no data

Table E.2 Mean depth in centimeters.

	Date	Coll.*	Depth (cm)	Stderr**	N***
A. Oiled, Open Coast					
Isla Droque Mangrove	02Mar1989	sg	55.64	2.16	55
Isla Mina Mangrove	02Mar1989	sg	64.69	1.51	51
Peña Guapa Mangrove	02Mar1989	sg	55.18	2.41	51
Punta Muerto Mangrove	02Mar1989	sg	55.09	3.22	54
B. Unoiled, Open Coast					
Isla Lintón Mangrove	09Sep1989	sg	55.39	1.91	51
María Soto Mangrove	05Mar1989	sg	61.40	1.82	57
Isla del Padre Mangrove	01Mar1989	sg	65.88	3.83	51
Portobelo Mangrove	01Mar1989	sg	62.30	2.19	50
C. Oiled Channels and Lagoons					
Largo Remo Channel West	31May1989	cg	23.36	2.47	50
Payardi Channel East	30May1989	cg	34.16	1.58	50
Payardi Channel South	30May1989	cg	52.40	2.02	50
Samba Bonita Channel East	30May1989	cg	22.50	2.15	50
Samba Bonita Channel South	30May1989	cg	38.07	1.16	28
D. Newly Oiled Channels and Lagoons					
Hidden Channel	24May1989	cg	37.06	1.45	50
E. Unoiled Channels and Lagoons					
Largo Remo Channel South	31May1989	cg	45.36	2.17	50
Margarita Channel North	01Jun1989	cg	31.76	1.51	50
Margarita Channel South	02Jun1989	cg	18.00	0.85	48
Samba Bonita Channel West	24May1989	cg	63.54	2.88	50
F. Unoiled Drainage Streams					
Río Alejandro	26May1989	cg	172.54	10.72	50
Hidden River	03Mar1989	sg	130.04	13.20	50
Quebrada Las Mercedes	02Mar1989	sg	93.88	6.84	50
Unnamed River	26May1989	cg	127.96	4.97	50

* Coll. = Data collector, cg = C. Gonzales, sg = S.D. Garrity. Data collected by cg need to be checked as of date of report submission.

** Stderr. = Standard error.

*** N = Number of measurements.

Table E.3 Substratum characteristics at each site; qualitative description.

Habitat	Site	Description
Open, oiled	Isla Mina Mangrove	sand, shell fragments, coral rubble, calcareous algal fragments
	Peña Guapa Mangrove	sand, shell fragments, peat, coral rubble, inorganic mud
	Isla Droque Mangrove	sand, shell fragments, coral rubble, peat, calcareous algal fragments
	Punta Muerto Mangrove	sand, rock, organic mud, coral rubble, shell fragments
Open, unoiled	María Soto Mangrove	sand, coral rubble
	Portobelo Mangrove	sand, coral rubble, inorganic mud, rock, peat, shell fragments
	Isla Padre Mangrove	sand, peat, shell fragments, rock
	Isla Lintón Mangrove	sand, shell fragments, coral rubble, peat, calcareous algal fragments, inorganic mud
Channel, oiled	Samba Bonita East	peat, inorganic mud
	Samba Bonita South	organic and inorganic mud, peat
	Payardi Channel East	peat, calcareous algal fragments, inorganic mud
	Payardi Channel South	inorganic mud
Channel, unoiled	Largo Remo West	peat, organic mud
	Margarita Channel South	peat, coral fragments, inorganic mud
	Margarita Channel North	inorganic mud, peat
	Hidden Channel *	inorganic mud, peat
Streams, oiled	Samba Bonita West	peat, organic mud
	Largo Remo South	peat, organic mud
	Largo Remo River South	inorganic mud, peat
	Payardi River	thick inorganic mud
Streams, unoiled	Punta Muerto River East	peat, inorganic mud
	Punta Muerto River West	inorganic mud
	Hidden River	fine sediment
	Unnamed River	fine sediment
	Río Alejandro	fine sediments, inorganic mud, peat
	Quebrada Las Mercedes	peat, inorganic mud

Note: These descriptions are from 3 cores sampled at each site for sediment hydrocarbon analyses in May 1989. Inorganic mud = grey soil, low in dark organic matter; organic mud = dark brown soil, highly organic.

* Hidden Channel secondarily oiled between May and August 1988.

Table E.4 Root condition, March-August 1987.

Site	Date	Percent		N
		Damaged /dead	Live	
A. Unoiled Open Sites				
María Soto Mangrove (MSM)	5-87	2	98	50
	8-87	-	100	20
Portobelo Mangrove (PBM)	5-87	-	100	50
	8-87	-	100	20
Isla del Padre Mangrove (PADM)	8-87	-	100	20
Isla Lintón Mangrove (LINM)	8-87	5	95	20
B. Oiled Open Sites				
Punta Galeta Mangrove (GALM)	3-87	35	65	88
	5-87	32	68	25
	8-87	65	35	20
Isla Mina Mangrove (MINM)	3-87	26	74	415
	5-87	44	68	75
	8-87	55	45	20
Peña Guapa Mangrove (PGM)	8-87	45	55	20
Isla Droque Mangrove (DROM)	8-87	55	45	20
Punta Muerto Mangrove (PMM)	8-87	60	40	20
C. Unoiled Channel Sites				
Hidden Channel (HIDC)	3-87	11	89	191
	5-87	4	96	50
	8-87	5	95	20
Samba Bonita Channel West (SBCW)	3-87	3	97	225
	5-87	2	98	50
	8-87	-	100	20
Margarita Channel North (MACN)	8-87	-	100	20
Margarita Channel South (MACS)	8-87	-	100	20

Table E.4 Root condition, March-August 1987 (continued).

D. Oiled Channel Sites				
Peña Guapa Channel (PGC)	3-87	41	59	249
	5-87	28	72	50
Largo Remo Channel West (LRCW)	3-87	34	66	162
	5-87	32	68	25
Largo Remo Channel CO3 (CO3)	3-87	36	64	110
Payardi Channel East (PCE)	8-87	50	50	20
Payardi Channel South (PCS)	8-87	55	45	20
Samba Bonita Channel East (SBCE)	8-87	60	40	20
Samba Bonita Channel South (SBCS)	8-87	60	40	20
E. Unoiled River Sites				
Hidden River (HIDR)	5-87	-	100	50
	8-87	10	90	20
Río Coco Solo (CSR)	5-87	-	100	50
Unnamed River (UNR)	8-87	-	100	20
Río Alejandro (ALER)	8-87	-	100	20
Quebrada Las Mercedes (MERR)	8-87	-	100	20
G. Oiled River Sites				
Largo Remo River South (LRRS)	3-87	97	3	238
	5-87	54	46	50
	8-87	90	10	20
Largo Remo River North (LRRN)	3-87	98	2	176
	5-87	58	42	50
Payardi River (PAYR)	8-87	35	65	20
Punta Muerto River East (PMRE)	8-87	65	35	20
Punta Muerto River West (PMRW)	8-87	75	25	20

Note: Data taken before standardization of methods.

3-87 - random sample of roots at study site;

5, 8-87 - roots sampled in longterm study, recorded separately from percent cover data.

Table E.5 Bivalve species in oiled and unoiled channels: mean percent cover \pm one standard error. Long-term census.

Date	<i>Brachidontes ?exustus</i>	<i>Crassostrea rhizophorae</i>	<i>Crassostrea sp. 2</i>	<i>Isognomon ?alatus</i>	<i>Mytilopsis sallei</i>
A. Unoiled Channels					
Hidden Channel					
11-81 *	3.19 \pm 0.69	49.81 \pm 2.48	0	7.06 \pm 0.96	13.00 \pm 1.05
01-82 *	2.42 \pm 0.67	54.21 \pm 2.29	0	5.69 \pm 0.78	12.32 \pm 1.13
06-82 *	4.50 \pm 1.09	53.75 \pm 5.06	0	4.25 \pm 1.12	10.50 \pm 1.96
08-86	9.46 \pm 1.92	35.71 \pm 3.81	0	2.86 \pm 0.64	5.86 \pm 1.44
11-86	2.40 \pm 0.43	27.22 \pm 3.06	0	3.78 \pm 0.92	8.29 \pm 2.12
02-87	2.60 \pm 0.65	30.15 \pm 2.73	0	3.15 \pm 0.53	4.38 \pm 0.84
05-87	2.26 \pm 0.52	27.60 \pm 1.75	0.36 \pm 0.21	5.00 \pm 0.62	7.02 \pm 1.34
08-87	1.30 \pm 0.60	32.65 \pm 2.79	0.95 \pm 0.41	4.15 \pm 1.02	15.85 \pm 3.55
11-87	1.96 \pm 0.62	27.80 \pm 2.17	1.24 \pm 0.39	4.04 \pm 0.94	14.72 \pm 2.28
02-88	0.40 \pm 0.18	21.75 \pm 2.42	3.70 \pm 0.98	1.65 \pm 0.50	13.15 \pm 2.59
05-88	0.75 \pm 0.36	22.95 \pm 2.50	1.00 \pm 0.40	2.00 \pm 0.49	10.80 \pm 2.00
08-88**	0.35 \pm 0.25	22.15 \pm 3.11	1.60 \pm 0.51	1.35 \pm 0.42	10.15 \pm 1.61
11-88**	0.65 \pm 0.28	21.65 \pm 2.19	1.15 \pm 0.56	3.50 \pm 0.78	5.75 \pm 1.23
02-89**	0.60 \pm 0.36	20.10 \pm 2.47	1.15 \pm 0.47	2.05 \pm 0.64	5.20 \pm 1.54
05-89**	0.45 \pm 0.21	18.85 \pm 2.27	0.75 \pm 0.35	2.20 \pm 0.69	6.30 \pm 1.58
Samba Bonita Channel West					
08-86	7.92 \pm 1.40	32.78 \pm 3.82	0	2.17 \pm 0.45	8.92 \pm 1.48
11-86	4.30 \pm 1.16	35.28 \pm 2.79	0	1.92 \pm 0.43	3.04 \pm 0.95
02-87	1.80 \pm 0.57	27.12 \pm 2.45	0	1.66 \pm 0.35	4.22 \pm 1.18
05-87	5.74 \pm 0.90	25.50 \pm 2.14	0.44 \pm 0.18	1.70 \pm 0.49	0.28 \pm 0.13
08-87	2.85 \pm 0.71	30.90 \pm 3.41	2.15 \pm 0.56	1.50 \pm 0.47	24.65 \pm 5.36
11-87	3.92 \pm 1.25	29.24 \pm 2.46	1.48 \pm 0.62	2.04 \pm 0.58	12.04 \pm 1.97
02-88	2.50 \pm 0.99	22.80 \pm 2.31	3.40 \pm 0.81	1.60 \pm 0.69	8.15 \pm 2.11
05-88	6.75 \pm 1.41	20.20 \pm 2.63	1.45 \pm 0.40	0.75 \pm 0.26	6.95 \pm 1.57
08-88	2.25 \pm 0.65	20.35 \pm 1.93	1.60 \pm 0.56	0.55 \pm 0.29	1.55 \pm 0.55
11-88	2.00 \pm 0.58	19.05 \pm 1.90	2.35 \pm 0.71	0.55 \pm 0.20	2.40 \pm 0.78
02-89	2.90 \pm 0.85	20.95 \pm 1.65	2.90 \pm 0.80	1.55 \pm 0.56	4.45 \pm 1.79
05-89	6.25 \pm 1.24	22.90 \pm 3.18	2.20 \pm 0.72	2.20 \pm 0.69	1.40 \pm 0.47

Table E.5 Bivalve species in oiled and unoiled channels (continued).

Margarita Channel North					
08-87	0.35 ± 0.24	17.75 ± 2.03	0.25 ± 0.25	13.55 ± 2.69	4.15 ± 1.24
11-87	0.08 ± 0.08	18.36 ± 1.75	0.64 ± 0.31	4.52 ± 0.92	4.12 ± 0.95
02-88	0.20 ± 0.12	12.40 ± 1.91	0.30 ± 0.25	5.45 ± 1.63	2.65 ± 0.98
05-88	0	12.00 ± 2.55	0	5.00 ± 1.75	3.40 ± 1.30
08-88	0.20 ± 0.20	10.10 ± 1.67	0.35 ± 0.35	5.20 ± 1.73	1.65 ± 0.96
11-88	0.10 ± 0.07	8.40 ± 1.54	0.60 ± 0.35	3.15 ± 1.01	0.55 ± 0.29
02-89	0	9.90 ± 1.80	0.35 ± 0.35	4.60 ± 1.55	0.90 ± 0.70
05-89	0.10 ± 0.10	7.05 ± 1.48	0.35 ± 0.24	7.45 ± 2.00	0.80 ± 0.66
Margarita Channel South					
08-87	1.50 ± 0.74	24.10 ± 2.22	0	4.75 ± 1.10	3.50 ± 0.88
11-87	0.40 ± 0.24	17.00 ± 2.01	0.48 ± 0.34	6.08 ± 1.43	2.20 ± 0.81
02-88	0	8.95 ± 1.76	0	1.95 ± 0.79	0.55 ± 0.29
05-88	0	12.85 ± 2.19	0	2.60 ± 1.02	0.95 ± 0.57
08-88	0	6.00 ± 1.87	0	1.05 ± 0.55	2.40 ± 1.46
11-88	0	6.50 ± 1.08	0.25 ± 0.25	1.00 ± 0.42	1.85 ± 0.73
02-89	0	3.60 ± 0.96	0	1.45 ± 0.82	1.10 ± 0.50
05-89	0	8.95 ± 1.35	2.00 ± 0.74	1.65 ± 0.62	0.25 ± 0.18
Largo Remo Channel South					
08-88	4.50 ± 1.06	18.40 ± 2.15	1.15 ± 0.49	0.75 ± 0.25	0.45 ± 0.18
11-88	3.15 ± 0.86	20.10 ± 1.61	0.70 ± 0.34	1.55 ± 0.65	1.40 ± 0.55
02-89	4.05 ± 0.98	23.40 ± 2.02	2.50 ± 0.77	3.00 ± 0.76	2.55 ± 0.62
05-89	2.20 ± 0.52	18.60 ± 1.88	1.15 ± 0.48	3.60 ± 0.81	3.50 ± 1.27
B. Oiled Channels					
Largo Remo Channel West					
08-86	1.39 ± 0.35	25.56 ± 5.96	0	3.00 ± 0.94	4.22 ± 1.80
11-86	1.83 ± 0.70	3.58 ± 1.19	0	1.92 ± 0.77	2.83 ± 1.07
02-87	2.00 ± 0.81	5.13 ± 1.04	0	3.21 ± 0.54	1.63 ± 0.52
05-87	4.00 ± 1.03	5.96 ± 1.16	0	2.12 ± 0.50	1.80 ± 0.52
11-87	0.55 ± 0.46	2.05 ± 0.77	0	0.50 ± 0.50	4.75 ± 1.54
02-88	1.15 ± 0.51	4.65 ± 0.94	0	0.50 ± 0.29	10.20 ± 2.70
05-88	1.40 ± 0.58	4.80 ± 1.40	0	0.50 ± 0.28	3.95 ± 1.15
08-88	1.35 ± 0.66	3.30 ± 0.89	0.20 ± 0.20	0.75 ± 0.35	6.85 ± 2.01
11-88	0.95 ± 0.45	5.40 ± 1.40	0.20 ± 0.20	0.75 ± 0.35	5.05 ± 2.06
02-89	0.95 ± 0.55	5.45 ± 1.61	0.25 ± 0.18	0.85 ± 0.37	7.55 ± 1.93
05-89	0.70 ± 0.40	4.15 ± 1.10	0	2.45 ± 0.78	10.75 ± 3.33

Table E.5 Bivalve species in oiled and unoiled channels (continued).

Payardi Channel East					
08-87	0.20 ± 0.20	6.35 ± 1.75	0.15 ± 0.15	1.15 ± 0.70	4.45 ± 3.08
11-87	0.12 ± 0.09	13.20 ± 2.09	0	1.28 ± 0.51	2.32 ± 1.27
02-88	0.05 ± 0.05	15.45 ± 2.49	1.35 ± 0.45	2.20 ± 0.81	0.25 ± 0.16
05-88	0.05 ± 0.05	11.45 ± 2.00	0.20 ± 0.12	1.80 ± 0.88	0.30 ± 0.25
08-88	0.05 ± 0.05	15.35 ± 2.11	0.25 ± 0.18	3.80 ± 1.51	0.05 ± 0.05
11-88	0.05 ± 0.05	10.30 ± 1.72	0	1.45 ± 0.71	0.05 ± 0.05
02-89	0	14.20 ± 1.97	0.50 ± 0.37	3.10 ± 1.30	0
05-89	0.15 ± 0.15	12.90 ± 2.16	0	1.95 ± 0.96	0
Payardi Channel South					
08-87	0.05 ± 0.05	6.05 ± 1.48	0.25 ± 0.18	0.95 ± 0.37	2.95 ± 1.93
11-87	0.60 ± 0.22	13.04 ± 2.12	0.96 ± 0.39	2.40 ± 0.82	9.16 ± 3.06
02-88	0.05 ± 0.05	24.45 ± 2.23	0.25 ± 0.18	1.75 ± 0.49	3.50 ± 1.95
05-88	0.15 ± 0.11	19.40 ± 3.08	1.00 ± 0.55	1.25 ± 0.49	1.45 ± 1.15
08-88	0.05 ± 0.05	11.05 ± 2.10	0.20 ± 0.16	0.90 ± 0.46	2.60 ± 0.88
11-88	0.45 ± 0.20	12.65 ± 2.07	0.45 ± 0.31	1.10 ± 0.38	0.70 ± 0.40
02-89	0.25 ± 0.25	18.05 ± 2.36	0	3.15 ± 0.74	0.15 ± 0.11
05-89	0.25 ± 0.18	14.75 ± 2.28	0.15 ± 0.15	2.00 ± 0.60	0.25 ± 0.18
Samba Bonita Channel East					
08-87	0	7.10 ± 1.70	0	0.50 ± 0.31	3.50 ± 1.89
11-87	0.56 ± 0.26	7.60 ± 1.62	0	2.36 ± 1.10	0.48 ± 0.22
02-88	0	19.55 ± 3.81	0.25 ± 0.25	0.90 ± 0.66	0
05-88	0.15 ± 0.15	13.80 ± 3.08	0	0.80 ± 0.48	0.05 ± 0.05
08-88	0.35 ± 0.24	10.70 ± 1.98	0.40 ± 0.28	1.40 ± 0.56	0
11-88	0.10 ± 0.10	8.75 ± 1.90	0.15 ± 0.15	3.20 ± 1.67	0
02-89	0	14.60 ± 2.24	0.15 ± 0.15	4.25 ± 1.43	0
05-89	0	6.95 ± 1.98	0	1.55 ± 0.69	0
Samba Bonita Channel South					
08-87	0	3.50 ± 0.97	0	1.40 ± 0.61	2.25 ± 1.26
11-87	0	3.84 ± 1.69	0	0.88 ± 0.73	0.16 ± 0.11
02-88	0.25 ± 0.16	12.90 ± 2.65	0.90 ± 0.68	0.25 ± 0.18	0.20 ± 0.14
05-88	0.25 ± 0.14	17.65 ± 3.35	0.65 ± 0.39	0.20 ± 0.14	0.15 ± 0.15
08-88	0.90 ± 0.37	10.15 ± 1.92	0.70 ± 0.38	0.30 ± 0.25	0
11-88	0.15 ± 0.11	11.55 ± 2.20	0.55 ± 0.46	1.45 ± 0.80	0
02-89	0.40 ± 0.28	15.60 ± 3.15	1.15 ± 0.73	0.20 ± 0.14	0
05-89	0.20 ± 0.20	12.95 ± 2.80	0.30 ± 0.21	1.20 ± 0.48	0

Table E.5 Bivalve species in oiled and unoiled channels (continued).

Peña Guapa Channel						
08-86	0.62 ± 0.20	12.87 ± 1.95	0	8.98 ± 1.98	4.62 ± 1.24	
11-86	1.49 ± 0.37	7.21 ± 1.15	0	4.36 ± 0.86	1.04 ± 0.28	
02-87	1.58 ± 0.53	3.94 ± 0.67	0	2.25 ± 0.45	1.46 ± 0.43	
05-87	1.76 ± 0.40	7.48 ± 0.78	0.02 ± 0.02	3.30 ± 0.50	0.42 ± 0.21	
Largo Remo Channel Site CO3						
08-86	0.20 ± 0.14	43.87 ± 7.26	0	6.13 ± 1.50	3.13 ± 1.51	
11-86	0.58 ± 0.28	1.79 ± 0.81	0	2.21 ± 0.57	0.09 ± 0.06	
02-87	1.68 ± 0.50	1.44 ± 0.41	0	1.12 ± 0.45	1.24 ± 0.44	
05-87	1.52 ± 0.67	13.96 ± 2.53	0.04 ± 0.04	4.96 ± 0.96	0.12 ± 0.09	

* Monitorings before the oil spill.

** Monitorings of Hidden Channel done after the area was secondarily oiled between May and August 1988.

Appendix F

Hydrocarbon Analyses

Table F.1 Inventory of samples collected in 1988/89 in support of the long-term assessment of the Bahía Las Minas oil spill.

P3 number	Sample name	STRI site name	Collection date
Mangrove Organism Samples			
1	CO1-oyster/old	SBCE	07/12/88
7	CO1-oyster/new	SBCE	07/12/88
5	CO2-oyster/old	SBCS	07/12/88
8	CO2-oyster/new	SBCS	07/12/88
9	CO3-oyster/old	PCE	07/12/88
4	CO3-oyster/new	PCE	07/12/88
23	CO4-oyster/old	PCS	07/12/88
19	CO4-oyster/new	PCS	07/12/88
28	RU2-mussel/old	UNR	07/12/88
34	RU2-mussel/new	UNR	07/12/88
32	RU3-mussel/old	ALER	07/12/88
25	RU3-mussel/new	ALER	07/12/88
30	RO3-oyster/old	PMRE	07/12/88
35	RO3-oyster/new	PMRE	07/12/88
33	RU4-mussel/old	MERR	07/12/88
29	RU4-mussel/new	MERR	07/12/88
27	RO4-oyster/mixed	PMRW	07/12/88
6	CU1-oyster/mixed	MACS	08/12/88
12	CU1-oyster/mixed	MACS	08/12/88
2	CU2-oyster/old	MACN	08/12/88
10	CU2-oyster/new	MACN	08/12/88
3	CN3-oyster/old	HIDC	08/12/88
11	CN3-oyster/new	HIDC	08/12/88
20	CU4-oyster/old	SBCW	08/12/88
13	CU4-oyster/new	SBCW	08/12/88
21	CU5-oyster/old	LRCS	08/12/88
15	CU5-oyster/new	LRCS	08/12/88
17	CO5-oyster/mixed	LRCW	08/12/88
18	RU1-mussel/old	HIDR	08/12/88
14	RU1-mussel/new	HIDR	08/12/88
16	RO1-mussel/new	LRR	08/12/88
26	RO2-mussel/mixed	PAYR	09/12/88
36	RO2-oyster/mixed	PAYR	09/12/88

Table F.1 Inventory of samples collected in 1988/89 (continued).

P3 number	Sample name	STRI site name	Collection date
Mangrove Organism Samples			
31	RO4-mussel/mixed	PMRW	09/12/88
37	CN3-recently oiled mangrove root	HIDC	12/12/88
223	OU1-barnacle	MSM	14/12/88
219	OU2-barnacle	PBM	14/12/88
220	OU3-barnacle	PADM	14/12/88
225	OU4-barnacle	LINM	14/12/88
221	OO1-barnacle	MINM	15/12/88
224	OO2-barnacle	PGM	15/12/88
222	OO3-barnacle	DROM	15/12/88
226	OO4-barnacle	PMM	15/12/88
39	OO1-barnacle	MINM	05/03/89
40	OU1-barnacle	MSM	17/03/89
41	OO4-barnacle	PMM	05/03/89
42	OU2-barnacle	PBM	05/03/89
43	CO1-oyster	SBCE	28/02/89
44	CU2-oyster	MACN	27/02/89
45	CO4-oyster	PCS	28/02/89
46	CU4-oyster	SBCW	27/02/89
47	RO1-mussel	LRRS	03/03/89
48	RU2-mussel	UNR	03/03/89
49	RO4-mussel	PMRW	28/02/89
50	RU4-mussel	MERR	02/03/89
51	OU3-barnacle	PADM	10/05/89
52	OU1-barnacle	MSM	10/05/89
53	OO2-barnacle	PGM	11/05/89
54	OO3-barnacle	DROM	11/05/89
55	CU4-oyster	SBCW	11/05/89
56	CO5-oyster	LRCW	11/05/89
57	CO4-oyster	PCS	12/05/89
58	CU2-oyster	MACN	12/05/89
59	RU4-mussel	MERR	11/05/89
60	RU1-mussel	HIDR	12/05/89
61	RO1-mussel	LRRS	13/05/89

Table F.1 Inventory of samples collected in 1988/89 (continued).

P3 number	Sample name	STRI site name	Collection date
Mangrove Organism and Sediment Samples			
62	RO2-mussel	PAYR	13/05/89
134	Core(0-2cm)	MINM	28/04/89
136	Core(8-10cm)	MINM	28/04/89
139	Core(18-20cm)	MINM	28/04/89
141	Surface scrape	MINM	28/04/89
158	Surface scrape	MINM	28/04/89
160	Surface scrape	MINM	28/04/89
130	Core(0-2cm)	PGN	28/04/89
133	Core(8-10cm)	PGN	28/04/89
137	Core(18-20cm)	PGN	28/04/89
143	Surface scrape	PGN	28/04/89
145	Surface scrape	PGN	28/04/89
149	Surface scrape	PGN	28/04/89
129	Core(0-2cm)	DROM	28/04/89
132	Core(8-10cm)	DROM	28/04/89
138	Core(18-20cm)	DROM	28/04/89
161	Surface scrape	DROM	28/04/89
163	Surface scrape	DROM	28/04/89
165	Surface scrape	DROM	28/04/89
67	Core(0-2cm)	PMM	28/04/89
71	Core(8-10cm)	PMM	28/04/89
82	Core(18-20cm)	PMM	28/04/89
152	Surface scrape	PMM	28/04/89
154	Surface scrape	PMM	28/04/89
156	Surface scrape	PMM	28/04/89
64	Core(0-2cm)	SBCW	29/04/89
76	Core (8-10cm)	SBCW	29/04/89
85	Core (18-20cm)	SBCW	29/04/89
142	Surface scrape	SBCW	29/04/89
148	Surface scrape	SBCW	29/04/89
159	Surface scrape	SBCW	29/04/89
69	Core (0-2cm)	LRCS	29/04/89
78	Core (8-10cm)	LRCS	29/04/89
84	Core (18-20cm)	LRCS	29/04/89

Table F.1 Inventory of samples collected in 1988/89 (continued).

P3 number	Sample name	STRI site name	Collection date
Mangrove Sediment Samples			
144	Surface scrape	L RCS	29/04/89
155	Surface scrape	L RCS	29/04/89
166	Surface scrape	L RCS	29/04/89
95	Core (0-2cm)	L RCW	29/04/89
106	Core (8-10cm)	L RCW	29/04/89
110	Core (18-20cm)	L RCW	29/04/89
147	Surface scrape	L RCW	29/04/89
153	Surface scrape	L RCW	29/04/89
164	Surface scrape	L RCW	29/04/89
87	Core (0-2cm)	H IDC	29/04/89
98	Core (8-10cm)	H IDC	29/04/89
101	Core (18-20cm)	H IDC	29/04/89
151	Surface scrape	H IDC	29/04/89
157	Surface scrape	H IDC	29/04/89
162	Surface scrape	H IDC	29/04/89
68	Core (0-2cm)	H IDR	30/04/89
74	Core (8-10cm)	H IDR	30/04/89
83	Core (18-20cm)	H IDR	30/04/89
146	Surface scrape	H IDR	30/04/89
150	Surface scrape	H IDR	30/04/89
179	Surface scrape	H IDR	30/04/89
131	Core (0-2cm)	U NR	30/04/89
135	Core (8-10cm)	U NR	30/04/89
140	Core (18-20cm)	U NR	30/04/89
169	Surface scrape	U NR	30/04/89
177	Surface scrape	U NR	30/04/89
185	Surface scrape	U NR	30/04/89
113	Core (0-2cm)	L RRS	30/04/89
120	Core (8-10cm)	L RRS	30/04/89
128	Core (18-20cm)	L RRS	30/04/89
168	Surface scrape	L RRS	30/04/89
175	Surface scrape	L RRS	30/04/89
181	Surface scrape	L RRS	30/04/89
115	Core (0-2cm)	A LER	30/04/89

Table F.1 Inventory of samples collected in 1988/89 (continued).

P3 number	Sample name	STRI site name	Collection date
Mangrove Sediment Samples			
119	Core (8-10cm)	ALER	30/04/89
123	Core (18-20cm)	ALER	30/04/89
170	Surface scrape	ALER	30/04/89
174	Surface scrape	ALER	30/04/89
182	Surface scrape	ALER	30/04/89
112	Core (0-2cm)	SBCE	01/05/89
121	Core (8-10cm)	SBCE	01/05/89
126	Core (18-20cm)	SBCE	01/05/89
167	Surface scrape	SBCE	01/05/89
178	Surface scrape	SBCE	01/05/89
191	Surface scrape	SBCE	01/05/89
114	Core (0-2cm)	SBCS	01/05/89
117	Core (8-10cm)	SBCS	01/05/89
124	Core (18-20cm)	SBCS	01/05/89
173	Surface scrape	SBCS	01/05/89
184	Surface scrape	SBCS	01/05/89
195	Surface scrape	SBCS	01/05/89
111	Core (0-2cm)	PCS	01/05/89
116	Core (8-10cm)	PCS	01/05/89
125	Core (18-20cm)	PCS	01/05/89
141	Core (10-12cm)	PCS	01/05/89
142	Core (12-14cm)	PCS	01/05/89
143	Core (16-18cm)	PCS	01/05/89
171	Surface scrape	PCS	01/05/89
183	Surface scrape	PCS	01/05/89
194	Surface scrape	PCS	01/05/89
118	Core (0-2cm)	PCE	01/05/89
122	Core (8-10cm)	PCE	01/05/89
127	Core (18-20cm)	PCE	01/05/89
176	Surface scrape	PCE	01/05/89
188	Surface scrape	PCE	01/05/89
197	Surface scrape	PCE	01/05/89
63	Core (0-2cm)	LINM	02/05/89
73	Core (8-10cm)	LINM	02/05/89

Table F.1 Inventory of samples collected in 1988/89 (continued).

P3 number	Sample name	STRI site name	Collection date
Mangrove Sediment Samples			
79	Core (18-20cm)	LINM	02/05/89
180	Surface scrape	LINM	02/05/89
187	Surface scrape	LINM	02/05/89
196	Surface scrape	LINM	02/05/89
66	Core (0-2cm)	PADM	02/05/89
70	Core (8-10cm)	PADM	02/05/89
75	Core (18-20cm)	PADM	02/05/89
172	Surface scrape	PADM	02/05/89
189	Surface scrape	PADM	02/05/89
193	Surface scrape	PADM	02/05/89
90	Core (0-2cm)	MACS	03/05/89
97	Core (8-10cm)	MACS	03/05/89
105	Core (18-20cm)	MACS	03/05/89
190	Surface scrape	MACS	04/05/89
202	Surface scrape	MACS	04/05/89
217	Surface scrape	MACS	04/05/89
65	Core (0-2cm)	MACN	03/05/89
72	Core (8-10cm)	MACN	03/05/89
81	Core (18-20cm)	MACN	03/05/89
198	Surface scrape	MACN	04/05/89
209	Surface scrape	MACN	04/05/89
214	Surface scrape	MACN	04/05/89
92	Core (0-2cm)	PMRW	05/05/89
99	Core (8-10cm)	PMRW	05/05/89
10	Core(18-20cm)	PMRW	05/05/89
186	Surface scrape	PMRW	05/05/89
201	Surface scrape	PMRW	05/05/89
210	Surface scrape	PMRW	05/05/89
88	Core (0-2cm)	MERR	05/05/89
96	Core (8-10cm)	MERR	05/05/89
109	Core (18-20cm)	MERR	05/05/89
192	Surface scrape	MERR	05/05/89
208	Surface scrape	MERR	05/05/89
213	Surface scrape	MERR	05/05/89

Table F.1 Inventory of samples collected in 1988/89 (continued).

P3 number	Sample name	STRI site name	Collection date
Mangrove Sediment Samples			
91	Core (0-2cm)	PAYR	05/05/89
100	Core(8-10cm)	PAYR	05/05/89
108	Core (18-20cm)	PAYR	05/05/89
200	Surface scrape	PAYR	05/05/89
205	Surface scrape	PAYR	05/05/89
212	Surface scrape	PAYR	05/05/89
93	Core (0-2cm)	PMRE	05/05/89
102	Core (8-10cm)	PMRE	05/05/89
104	Core (18-20cm)	PMRE	05/05/89
199	Surface scrape	PMRE	05/05/89
203	Surface scrape	PMRE	05/05/89
216	Surface scrape	PMRE	05/05/89
89	Core (0-2cm)	MSM	06/05/89
94	Core (8-10cm)	MSM	06/05/89
103	Core (18-20cm)	MSM	06/05/89
206	Surface scrape	MSM	06/05/89
211	Surface scrape	MSM	06/05/89
218	Surface scrape	MSM	06/05/89
77	Core (0-2cm)	PBM	06/05/89
80	Core (8-10cm)	PBM	06/05/89
86	Core (18-20cm)	PBM	06/05/89
204	Surface scrape	PBM	06/05/89
207	Surface scrape	PBM	06/05/89
214	Surface scrape	PBM	06/05/89
Reef Flat Urchin Samples			
U21	MSU-urchin	M.S.Arriba	14/12/88
U22	MSU-urchin	M.S.Arriba	14/12/88
U23/24	MSU-urchin	M.S.Arriba	14/12/88
U17	MSD-urchin	M.S.Abajo	14/12/88
U18	MSD-urchin	M.S.Abajo	14/12/88
U19	MSD-urchin	M.S.Abajo	14/12/88

Table F.1 Inventory of samples collected in 1988/89 (continued).

P3 number	Sample name	STRI site name	Collection date
Reef Flat Urchin Samples			
U14	GAL-urchin	GAL	14/12/88
U15	GAL-urchin	GAL	14/12/88
U16	GAL-urchin	GAL	14/12/88
U13*	GAL-urchin	GAL	14/12/88
U1	MSU-urchin	MSU	24/04/89
U2	MSU-urchin	MSU	24/04/89
U3	MSU-urchin	MSU	24/04/89
U4	MSD-urchin	MSD	24/04/89
U5	MSD-urchin	MSD	24/04/89
U6	MSD-urchin	MSD	24/04/89
U7	GAL-urchin	GAL	24/04/89
U8	GAL-urchin	GAL	24/04/89
U9	GAL-urchin	GAL	24/04/89
U10	LAR-urchin	LAR	11/05/89
U11	LAR-urchin	LAR	11/05/89
U12	LAR-urchin	LAR	11/05/89
Seagrass Sediment Samples			
A	BNV1-surface	BVN	08/12/88
I	BNV2-surface	BVN	08/12/88
E	BNV3-surface	BVN	08/12/88
D	PALN1-surface	PALN	08/12/88
K	PALN2-surface	PALN	08/12/88
B	PALN3-surface	PALN	08/12/88
C	LINE1-surface	LINE	08/12/88
L	LINE2-surface	LINE	08/12/88
H	LINE3-surface	LINE	08/12/88
G	DONT1-surface	DONT	08/12/88
F	DONT2-surface	DONT	08/12/88
J	DONT3-surface	DONT	08/12/88
BB	BNV core 0-2	BVN	08/12/88
CC	BNV core 8-10	BVN	08/12/88

Table F.1 Inventory of samples collected in 1988/89 (continued).

P3 number	Sample name	STRI site name	Collection date
Seagrass Sediment Samples			
DD	BNV core 18-20	BVN	08/12/88
Q	LRS1-surface	LRS	09/12/88
U	LRS2-surface	LRS	09/12/88
M	LRS3-surface	LRS	09/12/88
T	MINN1-surface	MINN	09/12/88
V	MINN2-surface	MINN	09/12/88
N	MINN3-surface	MINN	09/12/88
S	PGN1-surface	PGN	09/12/88
W	PGN2-surface	PGN	09/12/88
O	PGN3-surface	PGN	09/12/88
P	LREN1-surface	LREN	09/12/88
R	LREN2-surface	LREN	09/12/88
X	LREN3-surface	LREN	09/12/88
EE	LREN core 0-2	LREN	09/12/88
FF	LREN core 8-10	LREN	09/12/88
GG	LREN core 18-20	LREN	09/12/88
Y	NARC1-surface	NARC	09/12/88
Z	NARC2-surface	NARC	09/12/88
AA	NARC3-surface	NARC	09/12/88
Coral Reef Samples			
XXVI	JUG coral 1	JUG surface seds	10/12/88
XXVIII	JUG coral 2	JUG surface seds	10/12/88
XXIX	JUG coral 3	JUG surface seds	10/12/88
LVIII	JUG Ss 1	JUG <i>S.siderea</i>	10/12/88
XLVII	JUG Ss 2	JUG <i>S.siderea</i>	10/12/88
XLVIII	JUG Ss 3	JUG <i>S.siderea</i>	10/12/88
LVII	JUG Ss 4	JUG <i>S.siderea</i>	10/12/88
L	JUG Ss 5	JUG <i>S.siderea</i>	10/12/88
LX	JUG Ss 6	JUG <i>S.siderea</i>	10/12/88
XLIII	JUG Ss 7	JUG <i>S.siderea</i>	10/12/88
XLIX	JUG Ss 8	JUG <i>S.siderea</i>	10/12/88
XLVI	JUG Ss 9	JUG <i>S.siderea</i>	10/12/88

Table F.1 Inventory of samples collected in 1988/89 (continued).

P3 number	Sample name	STRI site name	Collection date
Coral Reef Samples			
CCXXXI	JUG <i>Pa</i> 1	JUG <i>P.astreoides</i>	10/12/88
CLXLVIII	JUG <i>Pa</i> 2	JUG <i>P.astreoides</i>	10/12/88
CLXLVI	JUG <i>Pa</i> 3	JUG <i>P.astreoides</i>	10/12/88
CCXXXIII	JUG <i>Pa</i> 4	JUG <i>P.astreoides</i>	10/12/88
CCXXXII	JUG <i>Pa</i> 5	JUG <i>P.astreoides</i>	10/12/88
CCVI	JUG <i>Pa</i> 6	JUG <i>P.astreoides</i>	10/12/88
CLXLIV	JUG <i>Pa</i> 7	JUG <i>P.astreoides</i>	10/12/88
CLXX	JUG <i>Pa</i> 8	JUG <i>P.astreoides</i>	10/12/88
CLXXXII	JUG <i>Pa</i> 9	JUG <i>P.astreoides</i>	10/12/88
XXXVI	MAR3 coral 1	MAR3, surface seds	11/12/88
XXXII	MAR3 coral 2	MAR3, surface seds	11/12/88
XXXIV	MAR3 coral 3	MAR3, surface seds	11/12/88
XXXIX	MAR3 <i>Ss</i> 1	MAR3 <i>S.siderea</i>	11/12/88
XLV	MAR3 <i>Ss</i> 2	MAR3 <i>S.siderea</i>	11/12/88
XXXVIII	MAR3 <i>Ss</i> 3	MAR3 <i>S.siderea</i>	11/12/88
XXXVIII	MAR3 <i>Ss</i> 4	MAR3 <i>S.siderea</i>	11/12/88
XXXVIII	MAR3 <i>Ss</i> 5	MAR3 <i>S.siderea</i>	11/12/88
XLI	MAR3 <i>Ss</i> 6	MAR3 <i>S.siderea</i>	11/12/88
XLIV	MAR3 <i>Ss</i> 7	MAR3 <i>S.siderea</i>	11/12/88
XL	MAR3 <i>Ss</i> 8	MAR3 <i>S.siderea</i>	11/12/88
XLII	MAR3 <i>Ss</i> 9	MAR3 <i>S.siderea</i>	11/12/88
CCXXVI	MAR3 <i>Pa</i> 1	MAR3 <i>P.astreoides</i>	11/12/88
CCXXVIII	MAR3 <i>Pa</i> 2	MAR3 <i>P.astreoides</i>	11/12/88
CCXXX	MAR3 <i>Pa</i> 3	MAR3 <i>P.astreoides</i>	11/12/88
CCIII	MAR3 <i>Pa</i> 4	MAR3 <i>P.astreoides</i>	11/12/88
CXXXI	MAR3 <i>Pa</i> 5	MAR3 <i>P.astreoides</i>	11/12/88
CCXVI	MAR3 <i>Pa</i> 6	MAR3 <i>P.astreoides</i>	11/12/88
CCXI	MAR3 <i>Pa</i> 7	MAR3 <i>P.astreoides</i>	11/12/88
XXXVII	MAR3 <i>Pa</i> 8	MAR3 <i>P.astreoides</i>	11/12/88
CCXVIII	MAR3 <i>Pa</i> 9	MAR3 <i>P.astreoides</i>	11/12/88
XI	LRE2 coral 1	LRE2 surface seds	11/12/88
I	LRE2 coral 2	LRE2 surface seds	11/12/88
VII	LRE2 coral 3	LRE2 surface seds	11/12/88
XII	LRE1 coral 1	LRE1 surface seds	11/12/88

Table F.1 Inventory of samples collected in 1988/89 (continued).

P3 number	Sample name	STRI site name	Collection date
Coral Reef Samples			
V	LRE1 coral 2	LRE1 surface seds	11/12/88
X	LRE1 coral 3	LRE1 surface seds	11/12/88
III	PAYW coral 1	PAYW surface seds	11/12/88
IV	PAYW coral 2	PAYW surface seds	11/12/88
VIII	PAYW coral 3	PAYW surface seds	11/12/88
XXXI	PAYN coral 1	PAYN surface seds	11/12/88
XXXIII	PAYN coral 2	PAYN surface seds	11/12/88
XXXV	PAYN coral 3	PAYN surface seds	11/12/88
XV	PM coral 1	PM surface seds	11/12/88
XXII	PM coral 2	PM surface seds	11/12/88
XVII	PM coral 3	PM surface seds	11/12/88
XXIV	NARS coral 1	NARS surface seds	11/12/88
XIX	NARS coral 2	NARS surface seds	11/12/88
XIV	NARS coral 3	NARS surface seds	11/12/88
IX	GALC coral 1	GALC surface seds	12/12/88
VI	GALC coral 2	GALC surface seds	12/12/88
II	GALC coral 3	GALC surface seds	12/12/88
XXI	DONR coral 1	DONR surface seds	12/12/88
XVI	DONR coral 2	DONR surface seds	12/12/88
XXIII	DONR coral 3	DONR surface seds	12/12/88
XVIII	DMA coral 1	DMA surface seds	12/12/88
XIII	DMA coral 2	DMA surface seds	12/12/88
XX	DMA coral 3	DMA surface seds	12/12/88
XXX	PALW coral 1	PALW surface seds	12/12/88
XXV	PALW coral 2	PALW surface seds	12/12/88
XXVII	PALW coral 3	PALW surface seds	12/12/88
LXLVIII	GALC <i>Ss</i> 1	GALC <i>S.siderea</i>	13/12/88
LXXVII	GALC <i>Ss</i> 2	GALC <i>S.siderea</i>	13/12/88
LXXIV	GALC <i>Ss</i> 3	GALC <i>S.siderea</i>	13/12/88
LXLVII	GALC <i>Ss</i> 4	GALC <i>S.siderea</i>	13/12/88
LXXVIII	GALC <i>Ss</i> 5	GALC <i>S.siderea</i>	13/12/88
LXLV	GALC <i>Ss</i> 6	GALC <i>S.siderea</i>	13/12/88
LXXXII	GALC <i>Ss</i> 7	GALC <i>S.siderea</i>	13/12/88
LII	GALC <i>Ss</i> 8	GALC <i>S.siderea</i>	13/12/88

Table F.1 Inventory of samples collected in 1988/89 (continued).

P3 number	Sample name	STRI site name	Collection date
Coral Reef Samples			
CXIII	GALC <i>Ss</i> 9	GALC <i>S.siderea</i>	13/12/88
CLXXX	GALC <i>Pa</i> 1	GALC <i>P.astreoides</i>	13/12/88
CCXXXVIII	GALC <i>Pa</i> 2	GALC <i>P.astreoides</i>	13/12/88
CLXXVI	GALC <i>Pa</i> 3	GALC <i>P.astreoides</i>	13/12/88
CLXLIX	GALC <i>Pa</i> 4	GALC <i>P.astreoides</i>	13/12/88
CLXXXI	GALC <i>Pa</i> 5	GALC <i>P.astreoides</i>	13/12/88
CXLVII	GALC <i>Pa</i> 6	GALC <i>P.astreoides</i>	13/12/88
CLXXXVIII	GALC <i>Pa</i> 7	GALC <i>P.astreoides</i>	13/12/88
CLXXII	GALC <i>Pa</i> 8	GALC <i>P.astreoides</i>	13/12/88
CLXLV	GALC <i>Pa</i> 9	GALC <i>P.astreoides</i>	13/12/88
LXVI	LRE1 <i>Ss</i> 1	LRE1 <i>S.siderea</i>	13/12/88
LXIV	LRE1 <i>Ss</i> 2	LRE1 <i>S.siderea</i>	13/12/88
LXV	LRE1 <i>Ss</i> 3	LRE1 <i>S.siderea</i>	13/12/88
LXXII	LRE1 <i>Ss</i> 4	LRE1 <i>S.siderea</i>	13/12/88
LXXI	LRE1 <i>Ss</i> 5	LRE1 <i>S.siderea</i>	13/12/88
LXVII	LRE1 <i>Ss</i> 6	LRE1 <i>S.siderea</i>	13/12/88
LXII	LRE1 <i>Ss</i> 7	LRE1 <i>S.siderea</i>	13/12/88
LXXIII	LRE1 <i>Ss</i> 8	LRE1 <i>S.siderea</i>	13/12/88
LXIII	LRE1 <i>Ss</i> 9	LRE1 <i>S.siderea</i>	13/12/88
CLXXVIII	LRE1 <i>Pa</i> 1	LRE1 <i>P.astreoides</i>	13/12/88
CCXLVI	LRE1 <i>Pa</i> 2	LRE1 <i>P.astreoides</i>	13/12/88
CCVIII	LRE1 <i>Pa</i> 3	LRE1 <i>P.astreoides</i>	13/12/88
CCXIII	LRE1 <i>Pa</i> 4	LRE1 <i>P.astreoides</i>	13/12/88
CCXX	LRE1 <i>Pa</i> 5	LRE1 <i>P.astreoides</i>	13/12/88
CCXXIII	LRE1 <i>Pa</i> 6	LRE1 <i>P.astreoides</i>	13/12/88
CCI	LRE1 <i>Pa</i> 7	LRE1 <i>P.astreoides</i>	13/12/88
CCXV	LRE1 <i>Pa</i> 8	LRE1 <i>P.astreoides</i>	13/12/88
CCXXXIV	LRE1 <i>Pa</i> 9	LRE1 <i>P.astreoides</i>	13/12/88
LIX	LRE2 <i>Ss</i> 1	LRE2 <i>S.siderea</i>	13/12/88
LIV	LRE2 <i>Ss</i> 2	LRE2 <i>S.siderea</i>	13/12/88
LXI	LRE2 <i>Ss</i> 3	LRE2 <i>S.siderea</i>	13/12/88
LIII	LRE2 <i>Ss</i> 4	LRE2 <i>S.siderea</i>	13/12/88
CXI	LRE2 <i>Ss</i> 5	LRE2 <i>S.siderea</i>	13/12/88
CXVIII	LRE2 <i>Ss</i> 6	LRE2 <i>S.siderea</i>	13/12/88

Table F.1 Inventory of samples collected in 1988/89 (continued).

P3 number	Sample name	STRI site name	Collection date
Coral Reef Samples			
LV	LRE2 <i>Ss</i> 7	LRE2 <i>S.siderea</i>	13/12/88
LVI	LRE2 <i>Ss</i> 8	LRE2 <i>S.siderea</i>	13/12/88
LI	LRE2 <i>Ss</i> 9	LRE2 <i>S.siderea</i>	13/12/88
CCXLVII	LRE2 <i>Pa</i> 1	LRE2 <i>P.astreoides</i>	13/12/88
CCXXXVII	LRE2 <i>Pa</i> 2	LRE2 <i>P.astreoides</i>	13/12/88
CCXLIV	LRE2 <i>Pa</i> 3	LRE2 <i>P.astreoides</i>	13/12/88
CCXLV	LRE2 <i>Pa</i> 4	LRE2 <i>P.astreoides</i>	13/12/88
CCXXXVI	LRE2 <i>Pa</i> 5	LRE2 <i>P.astreoides</i>	13/12/88
CCXLI	LRE2 <i>Pa</i> 6	LRE2 <i>P.astreoides</i>	13/12/88
CLXXV	LRE2 <i>Pa</i> 7	LRE2 <i>P.astreoides</i>	13/12/88
CCXXV	LRE2 <i>Pa</i> 8	LRE2 <i>P.astreoides</i>	13/12/88
CCXLVIII	LRE2 <i>Pa</i> 9	LRE2 <i>P.astreoides</i>	13/12/88
LXXV	DONR <i>Ss</i> 1	DONR <i>S.siderea</i>	14/12/88
CXV	DONR <i>Ss</i> 2	DONR <i>S.siderea</i>	14/12/88
CXVI	DONR <i>Ss</i> 3	DONR <i>S.siderea</i>	14/12/88
LXX	DONR <i>Ss</i> 4	DONR <i>S.siderea</i>	14/12/88
CXXII	DONR <i>Ss</i> 5	DONR <i>S.siderea</i>	14/12/88
CXXIV	DONR <i>Ss</i> 6	DONR <i>S.siderea</i>	14/12/88
LXL	DONR <i>Ss</i> 7	DONR <i>S.siderea</i>	14/12/88
LXVIII	DONR <i>Ss</i> 8	DONR <i>S.siderea</i>	14/12/88
LXIX	DONR <i>Ss</i> 9	DONR <i>S.siderea</i>	14/12/88
CLXL	DONR <i>Pa</i> 1	DONR <i>P.astreoides</i>	14/12/88
CCIV	DONR <i>Pa</i> 2	DONR <i>P.astreoides</i>	14/12/88
CCXXIV	DONR <i>Pa</i> 3	DONR <i>P.astreoides</i>	14/12/88
CC	DONR <i>Pa</i> 4	DONR <i>P.astreoides</i>	14/12/88
CCXXVII	DONR <i>Pa</i> 5	DONR <i>P.astreoides</i>	14/12/88
CCXIV	DONR <i>Pa</i> 6	DONR <i>P.astreoides</i>	14/12/88
CLXLII	DONR <i>Pa</i> 7	DONR <i>P.astreoides</i>	14/12/88
CLXLIII	DONR <i>Pa</i> 8	DONR <i>P.astreoides</i>	14/12/88
CXXXVI	DONR <i>Pa</i> 9	DONR <i>P.astreoides</i>	14/12/88
LXXVI	DMA <i>Ss</i> 1	DMA <i>S.siderea</i>	14/12/88
CXIX	DMA <i>Ss</i> 2	DMA <i>S.siderea</i>	14/12/88
LXXXIX	DMA <i>Ss</i> 3	DMA <i>S.siderea</i>	14/12/88
LXXIX	DMA <i>Ss</i> 4	DMA <i>S.siderea</i>	14/12/88

Table F.1 Inventory of samples collected in 1988/89 (continued).

P3 number	Sample name	STRI site name	Collection date
Coral Reef Samples			
LXXX	DMA <i>Ss</i> 5	DMA <i>S.siderea</i>	14/12/88
CXXVIII	DMA <i>Ss</i> 6	DMA <i>S.siderea</i>	14/12/88
CXVII	DMA <i>Ss</i> 7	DMA <i>S.siderea</i>	14/12/88
LXXXI	DMA <i>Ss</i> 8	DMA <i>S.siderea</i>	14/12/88
CXX	DMA <i>Ss</i> 9	DMA <i>S.siderea</i>	14/12/88
CLXXI	DMA <i>Pa</i> 1	DMA <i>P.astreoides</i>	14/12/88
CLXLVII	DMA <i>Pa</i> 2	DMA <i>P.astreoides</i>	14/12/88
CLXXXVII	DMA <i>Pa</i> 3	DMA <i>P.astreoides</i>	14/12/88
CCXXIX	DMA <i>Pa</i> 4	DMA <i>P.astreoides</i>	14/12/88
CCXII	DMA <i>Pa</i> 5	DMA <i>P.astreoides</i>	14/12/88
CXXX	DMA <i>Pa</i> 6	DMA <i>P.astreoides</i>	14/12/88
CLXVII	DMA <i>Pa</i> 7	DMA <i>P.astreoides</i>	14/12/88
CCXXII	DMA <i>Pa</i> 8	DMA <i>P.astreoides</i>	14/12/88
CXXXII	DMA <i>Pa</i> 9	DMA <i>P.astreoides</i>	14/12/88
LXLI	PAYN <i>Ss</i> 1	PAYN <i>S.siderea</i>	15/12/88
LXXXIII	PAYN <i>Ss</i> 2	PAYN <i>S.siderea</i>	15/12/88
LXXXVI	PAYN <i>Ss</i> 3	PAYN <i>S.siderea</i>	15/12/88
LXXXIV	PAYN <i>Ss</i> 4	PAYN <i>S.siderea</i>	15/12/88
LXXXVIII	PAYN <i>Ss</i> 5	PAYN <i>S.siderea</i>	15/12/88
LXLIX	PAYN <i>Ss</i> 6	PAYN <i>S.siderea</i>	15/12/88
CXXVII	PAYN <i>Ss</i> 7	PAYN <i>S.siderea</i>	15/12/88
CXXVI	PAYN <i>Ss</i> 8	PAYN <i>S.siderea</i>	15/12/88
CXXV	PAYN <i>Ss</i> 9	PAYN <i>S.siderea</i>	15/12/88
CCX	PAYN <i>Pa</i> 1	PAYN <i>P.astreoides</i>	15/12/88
CLXXXIII	PAYN <i>Pa</i> 2	PAYN <i>P.astreoides</i>	15/12/88
CLXXXVI	PAYN <i>Pa</i> 3	PAYN <i>P.astreoides</i>	15/12/88
CLXXXV	PAYN <i>Pa</i> 4	PAYN <i>P.astreoides</i>	15/12/88
CLXXXIV	PAYN <i>Pa</i> 5	PAYN <i>P.astreoides</i>	15/12/88
CLXXIII	PAYN <i>Pa</i> 6	PAYN <i>P.astreoides</i>	15/12/88
CCV	PAYN <i>Pa</i> 7	PAYN <i>P.astreoides</i>	15/12/88
CLXXIV	PAYN <i>Pa</i> 8	PAYN <i>P.astreoides</i>	15/12/88
CXXXIII	PAYN <i>Pa</i> 9	PAYN <i>P.astreoides</i>	15/12/88
CXLI	PAYN <i>Ds</i> 1	PAYN <i>D.strigosa</i>	15/12/88
CXLIII	PAYN <i>Ds</i> 2	PAYN <i>D.strigosa</i>	15/12/88

Table F.1 Inventory of samples collected in 1988/89 (continued).

P3 number	Sample name	STRI site name	Collection date
Coral Reef Samples			
CXXXIV&CII	PAYN <i>Ds</i> 3	PAYN <i>D.strigosa</i>	15/12/88
CI	PM <i>Ss</i> 1	PM <i>S.siderea</i>	15/12/88
LXLII	PM <i>Ss</i> 2	PM <i>S.siderea</i>	15/12/88
CXIV	PM <i>Ss</i> 3	PM <i>S.siderea</i>	15/12/88
LXLIV	PM <i>Ss</i> 4	PM <i>S.siderea</i>	15/12/88
LXXXVII	PM <i>Ss</i> 5	PM <i>S.siderea</i>	15/12/88
LXLVI	PM <i>Ss</i> 6	PM <i>S.siderea</i>	15/12/88
LXLIII	PM <i>Ss</i> 7	PM <i>S.siderea</i>	15/12/88
C	PM <i>Ss</i> 8	PM <i>S.siderea</i>	15/12/88
LXXXV	PM <i>Ss</i> 9	PM <i>S.siderea</i>	15/12/88
CCXVII	PM <i>Pa</i> 1	PM <i>P.astreoides</i>	15/12/88
CCXIX	PM <i>Pa</i> 2	PM <i>P.astreoides</i>	15/12/88
CCXXXV	PM <i>Pa</i> 3	PM <i>P.astreoides</i>	15/12/88
CLXIX	PM <i>Pa</i> 4	PM <i>P.astreoides</i>	15/12/88
CCVII	PM <i>Pa</i> 5	PM <i>P.astreoides</i>	15/12/88
CCII	PM <i>Pa</i> 6	PM <i>P.astreoides</i>	15/12/88
CCXXXIX	PM <i>Pa</i> 7	PM <i>P.astreoides</i>	15/12/88
CLXVIII	PM <i>Pa</i> 8	PM <i>P.astreoides</i>	15/12/88
CCIX	PM <i>Pa</i> 9	PM <i>P.astreoides</i>	15/12/88
CXXXVII	PAYN <i>Ds</i> 4	PAYN <i>D.strigosa</i>	16/12/88
CXXXV	PAYN <i>Ds</i> 5	PAYN <i>D.strigosa</i>	16/12/88
CXXXIX	PAYN <i>Ds</i> 6	PAYN <i>D.strigosa</i>	16/12/88
CXXXVIII	PAYN <i>Ds</i> 7	PAYN <i>D.strigosa</i>	16/12/88
CXL	PAYN <i>Ds</i> 8	PAYN <i>D.strigosa</i>	16/12/88
CXLII	PAYN <i>Ds</i> 9	PAYN <i>D.strigosa</i>	16/12/88
CLXVI	PAYN <i>Ap</i> 1	PAYN <i>A.palmata</i>	16/12/88
	PAYN <i>Ap</i> 2	PAYN <i>A.palmata</i>	16/12/88
CLXIII	PAYN <i>Ap</i> 3	PAYN <i>A.palmata</i>	16/12/88
CLVI	PAYN <i>Ap</i> 4	PAYN <i>A.palmata</i>	16/12/88
CLXI	PAYN <i>Ap</i> 5	PAYN <i>A.palmata</i>	16/12/88
CLXV	PAYN <i>Ap</i> 6	PAYN <i>A.palmata</i>	16/12/88
CLVII	PAYN <i>Ap</i> 7	PAYN <i>A.palmata</i>	16/12/88
CLXIV	PAYN <i>Ap</i> 8	PAYN <i>A.palmata</i>	16/12/88
CLXII	PAYN <i>Ap</i> 9	PAYN <i>A.palmata</i>	16/12/88

Table F.1 Inventory of samples collected in 1988/89 (continued).

P3 number	Sample name	STRI site name	Collection date
Coral Reef Samples			
CLIII	PAYN <i>At</i> 1	PAYN <i>A.tenuifolia</i>	16/12/88
CLIV	PAYN <i>At</i> 2	PAYN <i>A.tenuifolia</i>	16/12/88
CLII	PAYN <i>At</i> 3	PAYN <i>A.tenuifolia</i>	16/12/88
CL	PAYN <i>At</i> 4	PAYN <i>A.tenuifolia</i>	16/12/88
CLV	PAYN <i>At</i> 5	PAYN <i>A.tenuifolia</i>	16/12/88
CLIX	PAYN <i>At</i> 6	PAYN <i>A.tenuifolia</i>	16/12/88
CLVIII	PAYN <i>At</i> 7	PAYN <i>A.tenuifolia</i>	16/12/88
CLI	PAYN <i>At</i> 8	PAYN <i>A.tenuifolia</i>	16/12/88
CLX	PAYN <i>At</i> 9	PAYN <i>A.tenuifolia</i>	16/12/88
CVII	NARS <i>Ss</i> 1	NARS <i>S.siderea</i>	17/12/88
CIV	NARS <i>Ss</i> 2	NARS <i>S.siderea</i>	17/12/88
CIII	NARS <i>Ss</i> 3	NARS <i>S.siderea</i>	17/12/88
CX	NARS <i>Ss</i> 4	NARS <i>S.siderea</i>	17/12/88
CVIII	NARS <i>Ss</i> 5	NARS <i>S.siderea</i>	17/12/88
CV	NARS <i>Ss</i> 6	NARS <i>S.siderea</i>	17/12/88
CVI	NARS <i>Ss</i> 7	NARS <i>S.siderea</i>	17/12/88
CXII	NARS <i>Ss</i> 8	NARS <i>S.siderea</i>	17/12/88
CIX	NARS <i>Ss</i> 9	NARS <i>S.siderea</i>	17/12/88
CXLV	NARS <i>Pa</i> 1	NARS <i>P.astreoides</i>	17/12/88
CCXXI	NARS <i>Pa</i> 2	NARS <i>P.astreoides</i>	17/12/88
CLXXIX	NARS <i>Pa</i> 3	NARS <i>P.astreoides</i>	17/12/88
CLXXVII	NARS <i>Pa</i> 4	NARS <i>P.astreoides</i>	17/12/88
CLXXXIX	NARS <i>Pa</i> 5	NARS <i>P.astreoides</i>	17/12/88
CLXLI	NARS <i>Pa</i> 6	NARS <i>P.astreoides</i>	17/12/88
CXLVIII	NARS <i>Pa</i> 7	NARS <i>P.astreoides</i>	17/12/88
CXLIX	NARS <i>Pa</i> 8	NARS <i>P.astreoides</i>	17/12/88
CXLVI	NARS <i>Pa</i> 9	NARS <i>P.astreoides</i>	17/12/88
CXXIX	PAYW <i>Ss</i> 1	PAYW <i>S.siderea</i>	19/12/88
CXXI	PAYW <i>Ss</i> 2	PAYW <i>S.siderea</i>	19/12/88
CXXIII	PAYW <i>Ss</i> 3	PAYW <i>S.siderea</i>	19/12/88
CCCI	PAYW <i>Ss</i> 4	PAYW <i>S.siderea</i>	03/05/89
CCCI	PAYW <i>Ss</i> 5	PAYW <i>S.siderea</i>	03/05/89
CCLXLIX	PAYW <i>Ss</i> 6	PAYW <i>S.siderea</i>	03/05/89
CCCIV	PAYW <i>Ss</i> 7	PAYW <i>S.siderea</i>	03/05/89

Table F.1 Inventory of samples collected in 1988/89 (continued).

P3 number	Sample name	STRI site name	Collection date
Coral Reef Samples			
CCC	PAYW <i>Ss</i> 8	PAYW <i>S.siderea</i>	03/05/89
CCCII	PAYW <i>Ss</i> 9	PAYW <i>S.siderea</i>	03/05/89
CXLIV	PAYW <i>Pa</i> 1	PAYW <i>P.astreoides</i>	19/12/88
CCXL	PAYW <i>Pa</i> 2	PAYW <i>P.astreoides</i>	19/12/88
CCXLIII	PAYW <i>Pa</i> 3	PAYW <i>P.astreoides</i>	19/12/88
CCXLII	PAYW <i>Pa</i> 4	PAYW <i>P.astreoides</i>	19/12/88
CCLXLVIII	PAYW <i>Pa</i> 5	PAYW <i>P.astreoides</i>	03/05/89
CCLXLVI	PAYW <i>Pa</i> 6	PAYW <i>P.astreoides</i>	03/05/89
CCLXLIV	PAYW <i>Pa</i> 7	PAYW <i>P.astreoides</i>	03/05/89
CCLXLV	PAYW <i>Pa</i> 8	PAYW <i>P.astreoides</i>	03/05/89
CCLXLVII	PAYW <i>Pa</i> 9	PAYW <i>P.astreoides</i>	03/05/89
CCLXI	PALW <i>Ss</i> 1	PALW <i>S.siderea</i>	28/04/89
CCLXIII	PALW <i>Ss</i> 2	PALW <i>S.siderea</i>	28/04/89
CCLX	PALW <i>Ss</i> 3	PALW <i>S.siderea</i>	28/04/89
CCLXIV	PALW <i>Ss</i> 4	PALW <i>S.siderea</i>	28/04/89
CCLXVI	PALW <i>Ss</i> 5	PALW <i>S.siderea</i>	28/04/89
CCLVIII	PALW <i>Ss</i> 6	PALW <i>S.siderea</i>	28/04/89
CCLXII	PALW <i>Ss</i> 7	PALW <i>S.siderea</i>	28/04/89
CCLIX	PALW <i>Ss</i> 8	PALW <i>S.siderea</i>	28/04/89
CCLXV	PALW <i>Ss</i> 9	PALW <i>S.siderea</i>	28/04/89
CCLXVIII	PALW <i>Pa</i> 1	PALW <i>P.astreoides</i>	02/05/89
CCLXXIII	PALW <i>Pa</i> 2	PALW <i>P.astreoides</i>	02/05/89
CCLXVII	PALW <i>Pa</i> 3	PALW <i>P.astreoides</i>	02/05/89
CCLXXIV	PALW <i>Pa</i> 4	PALW <i>P.astreoides</i>	02/05/89
CCLXIX	PALW <i>Pa</i> 5	PALW <i>P.astreoides</i>	02/05/89
CCLXXI	PALW <i>Pa</i> 6	PALW <i>P.astreoides</i>	02/05/89
CCLXXII	PALW <i>Pa</i> 7	PALW <i>P.astreoides</i>	02/05/89
CCLXXV	PALW <i>Pa</i> 8	PALW <i>P.astreoides</i>	02/05/89
CCLXX	PALW <i>Pa</i> 9	PALW <i>P.astreoides</i>	02/05/89
CCLXXX	PALW <i>Ap</i> 1	PALW <i>A.palmata</i>	28/04/89
CCLXXXVII	PALW <i>Ap</i> 2	PALW <i>A.palmata</i>	28/04/89
CCLXXXVI	PALW <i>Ap</i> 3	PALW <i>A.palmata</i>	28/04/89
CCLXXXIV	PALW <i>Ap</i> 4	PALW <i>A.palmata</i>	28/04/89
CCLXXVIII	PALW <i>Ap</i> 5	PALW <i>A.palmata</i>	28/04/89

Table F.1 Inventory of samples collected in 1988/89 (continued).

P3 number	Sample name	STRI site name	Collection date
Coral Reef Samples			
CCLXXXI	PALW <i>Ap</i> 6	PALW <i>A.palmata</i>	28/04/89
CCLXXIX	PALW <i>Ap</i> 7	PALW <i>A.palmata</i>	28/04/89
CCLXXXII	PALW <i>Ap</i> 8	PALW <i>A.palmata</i>	28/04/89
CCLXXXIII	PALW <i>Ap</i> 9	PALW <i>A.palmata</i>	28/04/89
CCLXLIII	PALW <i>At</i> 1	PALW <i>A.tenuifolia</i>	02/05/89
CCLXXXVIII	PALW <i>At</i> 2	PALW <i>A.tenuifolia</i>	02/05/89
CCLXLII	PALW <i>At</i> 3	PALW <i>A.tenuifolia</i>	02/05/89
CCLXLI	PALW <i>At</i> 4	PALW <i>A.tenuifolia</i>	02/05/89
CCLXXXVI	PALW <i>At</i> 5	PALW <i>A.tenuifolia</i>	02/05/89
CCLXXXV	PALW <i>At</i> 6	PALW <i>A.tenuifolia</i>	02/05/89
CCLXL	PALW <i>At</i> 7	PALW <i>A.tenuifolia</i>	02/05/89
CCLXXXVII	PALW <i>At</i> 8	PALW <i>A.tenuifolia</i>	02/05/89
CCLXXXIX	PALW <i>At</i> 9	PALW <i>A.tenuifolia</i>	02/05/89
CCLIV	PALW <i>Ds</i> 1	PALW <i>D.strigosa</i>	28/04/89
CCLV	PALW <i>Ds</i> 2	PALW <i>D.strigosa</i>	28/04/89
CCXLIX	PALW <i>Ds</i> 3	PALW <i>D.strigosa</i>	28/04/89
CCLI	PALW <i>Ds</i> 4	PALW <i>D.strigosa</i>	28/04/89
CCLVII	PALW <i>Ds</i> 5	PALW <i>D.strigosa</i>	28/04/89
CCLII	PALW <i>Ds</i> 6	PALW <i>D.strigosa</i>	28/04/89
CCL	PALW <i>Ds</i> 7	PALW <i>D.strigosa</i>	28/04/89
CCLVI	PALW <i>Ds</i> 8	PALW <i>D.strigosa</i>	28/04/89
CCLIII	PALW <i>Ds</i> 9	PALW <i>D.strigosa</i>	28/04/89

As the Nation's principal conservation agency, the Department of the Interior has responsibility for most of our nationally owned public lands and natural resources. The includes fostering the wisest use of our land and water resources, protecting our fish and wildlife, preserving the environmental and cultural values of our national parks and historical places, and providing for the enjoyment of life through outdoor recreation. The Department assesses our energy and mineral resources and works to assure that their development is in the best interest of all our people. The Department also has a major responsibility for American Indian reservation communities and for people who live in Island Territories under U.S. Administration.

