No. 2 Fuel Oil Compound Retention and Release by Mytilus edulis:
1983 Cape Cod Canal Oil Spill

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TABLE OF CONTENTS

	<u>Page</u>
FORWARD.....	ii
ABSTRACT.....	iii
ACKNOWLEDGEMENTS.....	iv
I. INTRODUCTION.....	1
II. MATERIALS AND METHODS.....	2
III. RESULTS AND DISCUSSION.....	5
IV. REFERENCES.....	17
V. TABLES.....	20
VI. FIGURE LEGENDS and FIGURES.....	48

FORWARD

Coastal Research Center

Technical Report Series: Buzzards Bay

The Coastal Research Center of the Woods Hole Oceanographic Institution has initiated a series of WHOI/CRC Technical Reports in order to make information generated by CRC-associated researchers available as rapidly as possible to the coastal community.

Buzzards Bay is an important segment of the Massachusetts coast and has been studied by Woods Hole scientists and others for decades. However, only a few systematic studies have been completed. Increasing development pressure in this heavily used area has been followed by an increasing demand for more complete information on coastal environmental quality. In an effort to fill this information gap, and to foster understanding of fundamental processes operative in coastal areas in general, multi-disciplinary research is being pursued in Buzzards Bay. As the results of this become available, this information will be published in the CRC Technical Report Series.

ABSTRACT

Retention and release of No. 2 fuel oil compounds by Mytilus edulis contaminated by a small oil spill in the Cape Cod Canal in 1983 has been studied for the population "in situ" and for a subsample transplanted to a clean laboratory seawater system. Compounds analyzed include C_{13} to C_{24} n-alkanes; pristane; phytane; C_2 -, C_3 -naphthalenes; fluorene; phenanthrene; C_1 -, C_2 -, C_3 -phenanthrenes; fluoranthene; pyrene and dibenzothio-
phene. Biological half-lives were determined for the compounds from Day-3 to Day-29 following the spill and ranged from 1.5 days for C_2 -naphthalenes to 9.9 days for C_2 -phenanthrenes. Biological half-lives for the unresolved complex mixture determined by gas chromatographic analyses were 17 days for the alkane-cycloalkane fraction and 35 days for the aromatic fraction. Results compared favorably with data from a small oil spill contaminating the same mussel population at the same time of the year in 1978, although marked differences were noted for certain parameters.

Gas chromatographic-mass spectrometer analyses of C_2 - and C_3 -phenanthrenes revealed changes in relative abundance of compounds within isomer groups from samples at Day-29 to the time when no further detection of fuel oil was noted. These results suggest a release or metabolism of these compounds which is molecular structure specific.

This study also demonstrated the feasibility of training an analyst unfamiliar with analyses of hydrocarbons in tissues to conduct high resolution glass capillary GC analyses and some aspects of GCMS data systems output within a period of four to five months.

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I. INTRODUCTION

Fossil fuel compound contamination of bivalve populations in the world's coastal areas is a problem of continuing concern from the perspective of possible adverse impacts on human health associated with ingestion of contaminated bivalve tissue and adverse impacts on the viability of local and regional bivalve populations (NRC, 1980; 1985; Widdows et al., 1985, among others). Important aspects of the fate and effects of fossil fuel compounds in bivalves are uptake, retention, and release of compounds under experimental conditions and actual field conditions (Blumer et al., 1970; Farrington and Quinn, 1973; Boehm and Quinn, 1977; Marchand and Cabanne, 1980; NRC, 1980; NRC, 1985).

Several years ago we reviewed the literature about this subject and concluded that controversy surrounds many conclusions because of the wide range of conditions investigated; long-term chronic exposure in the field of months to years and short-term laboratory experiments of days to weeks (Farrington et al., 1982a). At that time, we reported our results from a study of the retention and release of No. 2 fuel oil compounds by a natural population of Mytilus edulis impacted by a small No. 2 fuel oil spill on April 1 and 2, 1978 (Farrington et al., 1982a). Our data did not resolve many of the controversies but did add to the limited data available for individual compound retention and release under actual field conditions at an oil spill where there was short-term exposure to spilled oil.

An unusual opportunity to investigate possible replication of our results occurred April 13, 14, 15, 1983, when the tugboat, Morton B. Bouchard, Jr., sank in the Cape Cod Canal releasing a No. 2 fuel oil slick. This slick impacted the population of Mytilus edulis that had been contaminated by the April 1 and 2, 1978, spill at approximately the same time of the year and under similar conditions. A small slick was sighted intermittently in the vicinity and within the intertidal bed of mussels for three days in 1983 and then disappeared. Presumably the oil slick was not replenished by fresh oil leakage from the tugboat and

the remnants of the slick were flushed from the Canal by strong tidal currents.

We report results of the study of retention and release of fuel oil compounds by Mytilus edulis contaminated by the 1983 spill and compare results and interpretations for both the 1978 and 1983 spills.

II. MATERIALS AND METHODS

Mytilus edulis were sampled on the south side of the Cape Cod Canal, at the railroad bridge abutment (Figure 1). This station was first established for studies of chemical contamination in bivalves as part of the U.S. Mussel Watch program, 1976-1978 (Goldberg et al., 1978; Farrington et al., 1982b; Farrington et al., 1983). Samples, size and other pertinent data are given in Tables 1 and 2. Samples were returned to the laboratory in solvent rinsed aluminum foil and frozen at -10°C to -20°C until shucking and analysis one to two years later.

Shell lengths were determined and meat plus shell were weighed. Bivalves were shucked and the meat plus liquid in the shells combined, weighed, and homogenized in a Virtis blender. Total sample was then stored frozen for a few days and subsequently subsampled for analysis or subsampled immediately for analysis. Dry weights were determined by freeze-drying approximately 60 g wet tissue of each homogenate in a Lab-conco Model Freeze Dry-5 for forty-eight hours. The non-freeze-dried homogenate was subsampled in duplicate for hydrocarbon analysis.

A large subsample of the field population was removed on April 14, 1983 and transported live to the Woods Hole Oceanographic Institution's Environmental Systems Laboratory where the organisms were maintained live in relatively clean seawater drawn from Nantucket Sound (Figure 1). Mussels were maintained in large plastic mesh cages and periodically samples were removed, frozen, and treated as above for samples from the Cape Cod Canal site. The purpose of this experiment was to provide a check on rates of release of compounds after removal to a cleaner water system on the presumption that the Canal site might still be contaminated by oil in the intertidal and subtidal sediments near the study site.

Saponification and Extraction

Approximately 15 g of wet tissue homogenate was placed into a 50 mL centrifuge tube with a Teflon-lined screw cap. Fifteen mL 6N NaOH (1:1 wt./wt., wet tissue: NaOH), prewashed with hexane, was added and the contents were shaken vigorously for two minutes. After an hour, the contents were again shaken vigorously for one minute, then placed into an oven at 30°C for eighteen hours.

Extraction of the hydrocarbons was accomplished by adding three separate aliquots of ethyl ether (1 x 15 mL, 2 x 10 mL). After each addition, the sample was shaken vigorously for one minute and centrifuged for 15 minutes at 3000 rpm to clearly separate the phases. The ethyl ether phase was drawn off using a disposable pipette. All ethyl ether extracts were combined.

If extracts were too thick with an emulsion, a further clean-up step was added. Extracts were transferred into a 250 mL separatory funnel and partitioned using hexane, then MeOH. The hexane was washed two times with H₂O/MeOH (10:1). Total volumes were approximately 100 mL of H₂O/MeOH and 50 mL of hexane.

Total Lipid Weight

Ethyl ether extract was evaporated just to dryness and 5 mL hexane was added. A 5-10 µL aliquot was weighed on a Cahn Model 25 Electro-balance to obtain an extractable non-saponifiable lipid weight.

Column Chromatography

Column chromatography was accomplished using silica gel (60-200 mesh) deactivated 5% with water and topped with alumina (~ 200 mesh) also deactivated 5% with water. The glass column was 10 mm i.d. x 35 cm long with a 250 mL reservoir and initially filled with 100 mL of hexane. The column was prepared by pouring sequentially 5 g of silica gel and 5 g alumina into the column allowing the air to escape and tapping the column for 1-2 minutes to promote settling. The initial hexane was drawn off and the complete column was rinsed with 60 mL clear hexane. The solvent dripped through until level with the top of the packing. The extract was charged

to the top of the column in a small volume (< 1 mL) of hexane. Fraction 1 was eluted with 20 mL of hexane. Fraction 2 was eluted with 20 mL of 10% toluene and 90% hexane followed by 20 mL of 20% toluene and 80% hexane. These fractions were concentrated by rotary evaporation prior to gas chromatographic analysis.

Gas Chromatographic Analysis

Both fractions were analyzed by glass capillary gas chromatography on a 0.32 mm i.d. x 30 m long SE-54 column installed in a Carlo Erba Model 4160 G.C. equipped with an on-column injector. Operating conditions were injection at 70°C holding one minute and programmed 3°C/minute up to 260°C, 4°C/minute from 260°C up to 310°C, and 6°C/minute from 310°C up to 330°C with H₂ as carrier gas at 0.5 kg/cm². Identification of alkane peaks was based on retention indices of compounds and co-injection with standards. Positive identification of aromatic hydrocarbons by mass spectra is described later.

Compounds were quantified by comparison with the peak heights of standards analyzed using the same conditions as sample analysis. The unresolved complex mixture signal (UCM) was measured by planimetry and quantified by using peak area calibrations of alkane and aromatic hydrocarbon standards.

GC-MS-Computer System Analysis

The aromatic/olefinic fraction, f₂, was analyzed on a Finnigan 4500 quadrupole mass spectrometer coupled to a Carlo Erba Model 4160 gas chromatography with on-column injector and a 0.32 mm i.d. x 30 m long SE-54 glass capillary column. The GC operating conditions were similar to those described above; injection at 70°C holding three minutes, 3.5°C/minute to 260°C, 4°C/minute from 260°C to 300°C. The mass spectrometer was operated as follows: 50 eV, 1.2 kV electron multiplier voltage, source temperature 100°C, scan from m/z 50 to 650 in 0.95 seconds with 0.05 second settling time. The INCOS Data System Automatic Quantification Program was utilized for quantification of aromatic hydrocarbons using response curves generated with standard compounds. Standards were not

available for all possible isomers of C_2 -naphthalenes (e.g., all dimethyl or ethylnaphthalenes), C_3 -naphthalenes, C_1 -phenanthrenes, C_2 -phenanthrenes and C_3 -phenanthrenes. Computer assisted manual integration methods were applied to molecular ion chromatogram plots of these compounds to obtain total peak areas for each isomer grouping. Response factors of available standards with each isomer grouping were applied to the entire group to obtain quantitative data for each grouping, e.g. ΣC_2 -naphthalenes, ΣC_3 -naphthalenes.

Internal Standards

Internal standards used in the analyses were: 1) $n-C_{14}$, $n-C_{28}$, androstane, d_{10} -fluorene, d_{14} -terphenyl spiked to the tissue in the extraction tube prior to extraction; 2) d_{10} -biphenyl, d_{10} -anthracene and d_{12} -chrysene added to f_2 aromatic/olefinic fractions after GC and prior to GCMS.

Two analysts carried each duplicate subsample through the extraction, column chromatography and gas chromatography procedures, one was an experienced analyst, the second was learning the procedures for the first time.

III. RESULTS AND DISCUSSION

Recoveries

Recoveries of internal standards are given in Tables 3, 4, 5. The data for the first three sampling dates for the $n-C_{14}$ are presented but cannot be interpreted in a meaningful manner because insufficient internal standard was added relative to the $n-C_{14}$ already in the sample from the No. 2 fuel oil. Androstane internal standard measurement was imprecise for the first three samples because of the low amount added relative to No. 2 fuel oil compounds coeluting with androstane. Results for $n-C_{28}$ did not suffer from interference from No. 2 fuel oil because $n-C_{28}$ is in a higher molecular weight range than that present in No. 2 fuel oil.

Recovery values greater than 100 percent reflect dilution errors due to working with very small volumes of 50 μ L to 400 μ L and errors introduced by volume measurements associated with 5 or 10 μ L syringes used for

GC injections. In general the recovery values after the first three sets of duplicates are good and show no significant difference between analysts.

Recoveries of the aromatic hydrocarbon fraction internal standards d_{10} -fluorene and d_{14} -terphenyl are presented in Tables 4 and 5. There were few interferences noted although it is possible that gas chromatographic measurements of d_{10} -fluorene for the first three samples might have been influenced by other compounds in the fuel oil due to a low amount of internal standard. Recoveries of d_{10} -fluorene were generally lower than d_{14} -terphenyl and occasionally very low, e.g. 24 to 44 percent, reflecting losses of d_{10} -fluorene to volatilization during the concentration steps. Recoveries of d_{14} -terphenyl were anomalously high for the first samples probably reflecting dilution and/or syringe measurement error associated with familiarization or refamiliarization of the analysts with the procedures.

Recoveries of d_{10} -fluorene and d_{14} -terphenyl were lower for GCMS compared to GC reflecting volatilization losses associated with concentration and redilution involving small volumes of solvent on the order of 50 μ L - 500 μ L after GC and prior to GCMS. The data for recoveries do not indicate any systematic differences between analysts for the GC measurements. The GCMS measurement step was performed by a different analyst.

Overall, the recovery data is reasonable based on our general experience in the measurement of individual petroleum compounds in tissue samples (Farrington et al., 1986; Galloway et al., 1983) and agrees with earlier data from our laboratory (Farrington et al., 1982a,b; Galloway et al., 1983; Farrington et al., 1986). There is a need for further improvement in methodology to increase the recoveries of the lower molecular weight aromatic hydrocarbons in the d_{10} -fluorene molecular weight range. However, the data provide a frame of reference for interpretation of other data in our study which have not been corrected for losses during extraction, column chromatography and evaporation-concentration procedures.

No. 2 Fuel Oil Compounds

Alkane and gas chromatographically unresolved complex mixture (UCM) cycloalkane and alkane data for the Cape Cod Canal site mussels from Day-1 to Day-424 after the spill are given in Table 6. Duplicate analyses by the two analysts are in good agreement generally with the exception of the more volatile compounds $n\text{-C}_{11}$ to $n\text{-C}_{15}$ which are influenced by uneven losses to volatilization during solvent evaporation-concentration steps in the extraction and clean-up procedures. There are also some disagreements for alkanes present in low concentrations in mussel tissue outside of the No. 2 fuel oil molecular weight range, e.g. $n\text{-C}_{26}$ - $n\text{-C}_{32}$ due to low peak signal to background UCM signal in GC analyses that were optimized for measurement of higher concentrations of compounds in the No. 2 fuel oil range. The agreement of duplicates is in accord with agreements of other similar replicate measurements for the same compounds in other mussel tissue samples analyzed in our laboratory (Farrington et al., 1982a,b; Farrington et al., 1986).

A comparison of concentrations for alkanes and UCM for the first day after exposure of mussels (Day-1) for 1978 and 1983 reveals a slightly higher concentration of approximately 1.2 to 2 for most compounds in the 1983 mussels (Table 7). Thus, initial contamination of mussels by No. 2 fuel oil alkanes was approximately the same in both spills. There is a slight difference in the relative abundance of compounds in each of the two No. 2 fuel oils which explains the slight differences in relative abundance of alkanes in Day-1 samples for 1983 compared with 1978. These differences in relative abundance are common in comparisons of No. 2 fuel oils and are used to distinguish between different oils (NRC, 1985).

The Day-1 concentrations of the alkanes and UCM are much greater by factors of 10 to 100 than concentrations in mussels from this population sampled in 1976 and 1977 prior to the 1978 oil spill (Farrington et al., 1982a,b). The pre-spill concentrations and mixture of compounds are similar to concentrations of Day-113 and Day-424 after the 1983 spill (Table 6) and Day-87 after the 1978 spill.

Concentrations of most branched alkanes (e.g. pristane and phytane) and UCM increase slightly from Day-1 to Day-3. In part, this increase in concentration may be due to changes in the water to tissue ratios in the

mussels resulting in a decrease in total dry weights of individual animals but not in the No. 2 fuel oil content of individual animals. The net result would be a slight increase in concentration of No. 2 fuel oil compounds in the mussels. This explanation is supported by dry weight/wet weight ratios (Table 1) which change between Day-1 and Day-3, which may be the result of physiological response of the mussels to the No. 2 fuel oil.

The slight increases in concentrations of pristane, phytane, and UCM (Table 6) are not paralleled by increases in concentrations of No. 2 fuel oil n-alkanes. This could be due to differential metabolism resulting in faster loss of these n-alkanes relative to the uptake. Rapid loss of n-alkanes relative to the pristane, phytane and other branched alkanes was noted in the 1978 spill study and is characteristic of other studies as discussed previously (Farrington et al., 1982a). The loss is probably due to metabolism of No. 2 fuel oil n-alkanes by the mussels or indigenous microflora (Stegeman and Teal, 1973; Neff et al., 1976). An alternative explanation to explain the slight increase in concentrations of the pristane, phytane and UCM is further contamination of the mussel population by No. 2 fuel oil which had been partially degraded by microorganisms prior to arrival at the mussel population site. More rapid degradation of No. 2 fuel oil molecular weight range n-alkanes relative to pristane, phytane and UCM compounds is a common phenomena in microbial degradation of oil (NRC, 1985).

There is a rapid loss of n-alkanes, and a less rapid loss of pristane, phytane and UCM constituent compounds after Day-3 (Table 6). This phenomenon is illustrated by the representative gas chromatograms of Figure 2 and concentration-time plots of representative compounds in Figure 3. The gas chromatograms are very similar to those for the 1978 spill (Figure 4). There are other isoprenoid compounds, e.g. farnesane, in No. 2 fuel oil and these probably are represented by unidentified peaks that persist longer than n-alkanes in the gas chromatograms of f_1 (Figures 2 and 4).

The very rapid loss of n-alkanes after Day-3 is most likely the result of a combination of metabolism, microbial degradation by indigenous microflora as previously discussed, and release from tissue back to clean water after oil contaminated water has been swept from the area.

Concentrations of aromatic hydrocarbons determined by GC and GCMS are compared in Table 8. The measurements by GC are subject to interferences by co-eluting compounds and small GC peak signal relative to the UCM compounds, especially for samples taken after Day-3 (Figure 5). GCMS analyses combined mass spectral confirmation of compound identity with quantitative measurements of molecular ions of the specific compounds to provide a more discriminatory, accurate determination of the amount of individual aromatic compounds. Thus, it is no surprise that concentrations determined by GCMS are lower than concentrations determined by GC. The differences are often a factor of two to four and are not due to the respective differences in recoveries between GC and GCMS noted for internal standards earlier in this report. The comparisons of the GC and GCMS data (Table 8) demonstrates that caution should be exercised in the interpretation of aromatic hydrocarbon data from GC measurements. This caution applies for tissue samples with the high concentrations of No. 2 fuel oil compounds, e.g. Day-1, Day-2 and Day-3, as well as for tissue samples with lower concentrations of aromatic hydrocarbons.

Concentrations of isomer groupings of aromatic hydrocarbons (e.g. sum of all C_2 -phenanthrenes) determined by combined manual integration of molecular ion currents and INCOS automated data system processing are presented in Table 9. Time-concentration plots of representative data from Table 9 are illustrated in Figure 6.

Retention and release of aromatic hydrocarbons followed the general time trends of alkanes and f_1 UCM. Concentrations of several aromatic hydrocarbons fluctuated between Day-1 to Day-5, increasing and then decreasing. No. 2 fuel oil had been subjected to different combinations of volatilization, water column concentration, and microbial degradation over the period of Day-1 to Day-5, and coupled with the changes in dry tissue to wet tissue ratios as previously discussed, could account for the fluctuations in tissue concentrations of the aromatic hydrocarbons, alkanes and f_1 UCM and f_2 UCM during this period. Following Day-5, there are more rapid losses of the lower molecular weight aromatic hydrocarbons such as C_1 - and C_2 -naphthalenes compared to higher molecular weight compounds such as the C_1 - and C_2 -phenanthrenes. Some of the compounds con-

stituting the f_2 UCM seem to be retained over time to a greater extent compared to the C_1 -, C_2 -, C_3 -naphthalenes, and C_1 -, C_2 -, C_3 -phenanthrenes. The molecular weight distribution of the f_2 UCM shifts to higher molecular weights as time progresses as is illustrated in representative gas chromatograms in Figure 5.

Initial concentrations of some of the compounds present in the mussels on Day-1 of the 1978 and 1983 two spills were different (Table 10), perhaps reflecting differences in the aromatic hydrocarbon content and relative abundances of aromatic compounds in each of the fuel oils. The Day-1 mussels in the 1978 oil spill had higher concentrations of naphthalenes similar to concentrations of phenanthrenes and methylphenanthrenes, but lower concentrations of C_2 -phenanthrenes compared to Day-1 mussels of the 1983 spill (Table 10). However, the patterns of retention and release of aromatic hydrocarbons were similar for both spills as is indicated by comparison of gas chromatograms of aromatic compounds extracted from samples from the two oil spill studies (Figures 5 and 7).

Concentrations of No. 2 fuel oil compounds in mussels transferred on Day-2 to ESL (Environmental Systems Laboratory at W.H.O.I.) are given in Tables 11 and 12. GCMS analyses were not undertaken for these samples because of lack of funds. Thus, we cannot compare the Cape Cod Canal field site data with ESL data for aromatic compounds. Comparisons of GC data for f_2 could be subject to error due to interferences as noted previously. Table 13 compares data from the field site with data from ESL for alkanes and f_1 UCM. Concentrations are lower for ESL samples throughout the comparison period indicating that the field site mussel population was still exposed to oil in the habitat for a few days after Day-2. The ESL seawater system draws seawater from Nantucket Sound at a location removed from any direct influence of urban harbors or water from the Cape Cod Canal and thus would be expected to be relatively clean. The pattern of release of the alkanes and f_1 UCM in the ESL system is similar to the pattern of release for the field site (Figures 3 and 8). The differences are due to significant release starting a day or so later at the field site because of the continued presence of spilled oil in the field as indicated by the visual observations of the slick

and the probability of some residual oil in the substrate of the mussel population after the surface slick disappeared.

Fossato and Canzonier (1976) and Ernst (1977) discussed treatment of data for uptake, retention and release of hydrocarbons by bivalves in terms of a biological half-life concept. An exponential loss of compounds or decrease in concentration is assumed and an equation of the form, $C_t = C_0 e^{-bt}$ is fit to the data where C_t is concentration in the mussel at time t after removal of the exposure, C_0 is concentration of compound at time of removal of exposure conditions (e.g. flushing of an oil slick out of the canal), and b is a constant. This is a specific form of the general exponential equation $y = ae^{bx}$ and in treatment of our data x = time in days, a = intercept on y axis, b = slope of the line in a semilog plot, y = concentration of No. 2 fuel oil compound or compounds in mussel tissue. A biological half-life ($T_b 1/2$) of a compound is defined as the time taken for the concentration of a given compound to decrease to one-half the concentration at time 0 and is calculated as $T_b 1/2 = \ln 2/b = 0.693/b$. We have applied an exponential curve fitting program package of a Hewlett Packard Model 14C electronic calculator to the data for the 1983 and 1978 oil spills. Semilog plots of data are given in Figures 3, 6 and 9-19. We have used two different time intervals over which the 1983 data is calculated, Day-1 to Day-29 and Day-3 to Day-29 for purposes of comparison. The time interval Day-3 to Day-29 is the correct interval to use because that is the time interval beginning with Day-3 when the majority of the spilled oil was swept out of the canal during the 1983 spill.

Results of the biological half-life ($T_b 1/2$) calculations are given in Tables 14 to 18 for the 1983 spill. The correlation fits of data to the exponential equation are excellent for Day-3 to Day-29 (Tables 14-18). We did not use data after Day-29 in these calculations because the semilog plots showed significant departure from linearity for some compounds (Figures 3 and 6), concentrations were below detection limits for some compounds, and this is the closest interval of time to the $T_b 1/2$ over 21 days calculated for the 1978 oil spill.

Comparisons of the 1978 spill data and 1983 spill data for representative compounds are presented in Figures 9-19 and $T_b 1/2$ data are com-

pared in Table 19. There are some marked differences between the 1978 and 1983 spill data for n-C₁₆ and C₂-naphthalenes and reasonably good agreement for other data (Figures 9-19 and Table 19). This is the first comparison of this type to our knowledge and provides an estimate of the extent to which one set of biological half-lives quantifying retention and release of oil compounds in bivalves in one field situation might be extrapolated to the same population of bivalves at another point in time. This also provides limited guidance to the more general case of extrapolations from one study to any population of bivalves contaminated by fossil fuel compounds as a result of short-term exposures to spilled oil.

GCMS analyses during our study of the 1978 oil spill revealed that a pair of C₂-phenanthrene peaks in the mass chromatograms changed in relative abundance for the Day-21 samples and continued to change until concentrations approached background concentrations. We have been able to obtain better gas chromatographic resolution of the C₂-phenanthrene isomers and also data for C₃-phenanthrene isomers and this more clearly reveals these changes for the 1983 spill samples (Figure 20). The relative abundances of the isomers of C₂- and C₃-phenanthrenes are changed in the Day-29 samples compared to the Day-8 samples. This observation agrees with the 1978 spill data where changes in ratios were noted for the Day-21 samples but were not significant for the Day-11 samples. Similar changes in relative abundance of C₂-naphthalenes and C₃-naphthalenes have not been demonstrated because C₂- and C₃-naphthalenes are present in very low concentrations by Day-21.

Several studies of the effects of short-term and long-term chronic exposures of No. 2 fuel oil on Mytilus edulis and other molluscs have been completed since our study of the 1979 spill (Livingstone et al., 1985; and references therein). Livingstone et al. (1985) reported on the response of cytochrome P-450 monooxygenase system of Mytilus edulis to exposure to diesel oil (very similar to No. 2 fuel oil). The mussels in their study rapidly accumulated No. 2 fuel oil compounds from an eight-day exposure to water containing 29 ppb (10^{-6} g/L) No. 2 fuel oil to achieve a tissue concentration of 13.8×10^{-6} g/g wet weight of 2-ring aromatic compounds measured as 2,3-dimethylnaphthalene equivalents and 2.40×10^{-6}

g/g wet weight of 3-ring aromatic compounds measured as 1-methylphenanthrene equivalents. These concentrations are within approximately factors of two to four higher than those we have measured for the total of naphthalenes and phenanthrenes although strict comparisons are difficult because Livingstone estimated concentrations by an HPLC U.V.-fluorescence measurement method (Donkin and Evans, 1984). Livingstone et al. noted a general trend for molluscs exposed to petroleum hydrocarbons of increase in cytochromes P-450 and b₅, NADPH cytochrome c reductase, and NADPH neotetrazolium reductase activity but not much change, if any, in benzo-pyrene hydroxylase activity. Stegeman (1985) has demonstrated activity of P-450 systems and benzopyrene hydroxylase activity in Mytilus edulis. Thus, there is strong evidence of enzymatic activity in Mytilus edulis capable of metabolizing aromatic hydrocarbons, and there are indications of increases in enzymatic activity capable of metabolism of aromatic hydrocarbons linked to exposure of mussels to fuel oil. However, there is no proof, as yet, of the induction of elevated levels of the complete enzymatic system capable of metabolizing aromatic hydrocarbons in contrast to fish and crustacea where such an induction has been demonstrated (Stegeman, 1985; Livingstone et al., 1985; and references therein).

The changes in relative abundance of C₂-, and C₃-phenanthrenes could be explained by enzyme specific metabolism with some isomers being more susceptible to metabolism than others due to influence of substitution on steric hindrance effects or electrophilic substitution effects in substrate-enzyme interactions. The lack of changes in relative abundance prior to Day-29 could be due to a lag time required for induction of greater enzymatic activity for metabolizing the compounds. Another possibility is that the level of aromatic hydrocarbon metabolizing activity is approximately the same throughout the entire period of time but is much slower than the rate of release of unmetabolized C₂- and C₃-phenanthrenes back to the water. When concentrations of these compounds become low enough in tissue then the rate of release decreases to the point where enzymatic activity has an effect of sufficient magnitude to cause measurable changes in relative abundance in tissue. We cannot distinguish between these alternatives with our data nor do we offer proof that the

changes in relative abundance are due to metabolic transformations. Rather, we submit the changes in relative abundance are consistent with an influence by metabolism and consistent with the biochemical investigations of Livingstone et al. (1985) and Stegeman (1985).

GENERAL DISCUSSION

There was reasonably good agreement between data collected for retention and release of fuel oil compounds by the same population of Mytilus edulis impacted by two No. 2 fuel oil spills five years apart at about the same time of year. A general rapid loss of n-alkanes, followed by a less rapid loss of the isoprenoid alkanes pristane and phytane is shown. Lower molecular weight aromatic hydrocarbons (e.g. C₁- and C₂-naphthalenes) are released to the water more rapidly than higher molecular weight aromatic hydrocarbons (e.g. C₁- and C₂-phenanthrenes). It appears from these results and those of other investigators that partitioning between tissue and the water is a controlling factor in the first several weeks of release when animals are exposed to clean water in their habitat. The relative rate of release seems to be roughly correlated with solubility with the more soluble, lower molecular weight compounds released more rapidly than the less soluble, higher molecular weight compounds based on our data and the aromatic hydrocarbon solubility data of Whitehouse (1984), May (1980) and May and Wasik (1978).

The changing slopes of lines in the semilog plots of concentrations in tissue versus time (Figures 3, 6 and 9-19) are consistent with the multicompartment conceptual model of partitioning controlling uptake and release of petroleum compounds (NRC, 1980, 1985, and references therein). In this model some parts of the animals' tissue are more accessible to uptake and release of oil compounds back to water. The rapid depletion of the first type of tissue is followed by a slower release from the second type of tissue. There probably are many compartments or tissue types, each with a different rate of loss.

The changes in relative abundance of isomers in the C₂- and C₃-phenanthrene groups indicates a structure specificity control in later stages

of loss of compounds from mussel tissue. This is probably related to metabolism but we cannot rule out an influence of specificity related to molecular structure for partitioning between tissue and seawater. For example, isomers of chlorobiphenyls, similar in hydrophobic properties to aromatic hydrocarbons, have different octanol-water (K_{ow}) partition coefficients and partitioning between tissue and water has been correlated with K_{ow} s (Mackay, 1982 among others). More research is needed to identify the specific isomers involved in the changing C_2 - and C_3 -phenanthrenes before we can determine which processes are involved in changing the relative abundance of isomers.

The f_1 and f_2 UCM signal is the longest persisting of several parameters measured from the No. 2 fuel oil contamination in tissue samples. Further GCMS investigations of the compounds constituting these complex mixtures would be helpful in elucidating specific compounds which might be used as quantitative or semiquantitative long-term spill contamination indicators as we and others have suggested previously (Farrington et al., 1982a; Anderlini et al., 1980; NRC, 1985). Investigations of this type were beyond the scope of our current investigation due to funding constraints.

Results from this study support our earlier conclusions following the 1978 study (Farrington et al., 1982a). We paraphrase these comments here. Rate of uptake and release by mussels depends on the various petroleum hydrocarbon molecular types, molecular weights, and molecular configurations, the type of exposure (acute or chronic) and duration of exposure. These variables place constraints on monitoring programs involving the measurement of petroleum hydrocarbons in bivalve molluscs (Farrington et al., 1982b, 1983; Topping, 1983; IFREMER, 1985, among others). In both spills, a single measurement in time for the Mytilus edulis population would have yielded very different indications of the extent of petroleum pollution depending on the time of sampling. Thus, while measurements of petroleum compounds in bivalves are probably good indicators for long-term oil contamination in a given area (Farrington et al., 1982b, 1983; Topping, 1983, among others), interpretation must be made cautiously and confirmed by repeated measurements to elucidate the pos-

sible roles of small spills as contributors to the concentrations of petroleum compounds.

A very important aspect of this study was the demonstration that a scientist with limited experience in the high resolution analyses of hydrocarbons (one of the co-authors, Jia Xiaoping) was able to learn the methods for complicated glass capillary GC analyses and a few aspects of GCMS analyses within a period of four to five months and to conduct many analyses yielding high quality data for this study. This was accomplished by intensive interactions between the person being educated and trained, and personnel actively involved in ongoing projects in a research laboratory. This is an important consideration within the context of future international training programs and international programs in coastal and oceanic environmental quality research and monitoring efforts for organizations such as IOC-UNESCO (Intergovernmental Oceanographic Commission) and the United Nations Environment Program (UNEP).

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Table 1. Cape Cod Canal Railroad Bridge No. 2 Fuel Oil Spill:
Mytilus edulis Samples

Sample ^a	No. of Individuals	Length (cm)		Total Shelled Weight (g)	Average Shelled Weight (g)
		Range	Average		
CCCRB-830413	67	3-7	5.8	1300	19.4
CCCRB-830414	52	4-8	6.6	1512	29.1
CCCRB-830415	62	2-8	5.9	1597	25.8
CCCRB-830417	54	6-8	6.6	1795	33.2
CCCRB-830420	55	2-8	4.7	1202	21.8
CCCRB-830511	51	5-8	6.4	1412	27.7
CCCRB-830519	49	4-8	5.9	1465	29.9
CCCRB-830526	73	3-7	5.5	1853	25.4
CCCRB-830803	53	2-8	5.9	1582	29.8
CCCRB-840610	65	2-8	5.7	1664	25.6
ESL, Depuration Experiment					
830415	65	3-7	5.7	1615	24.8
830417	78	3-7	5.4	1685	21.6
830420	87	3-7	5.5	1948	22.4
830511	50	4-7	5.6	1263	25.3
830519	50	5-8	6.4	1324	26.5

^aCode Number 830413 is: Year (1983), Month (04 April), Day (13).

Table 1. (continued)

Total Wet Weight (g)	Average Wet Weight (g)	Percent Water (%)	Ratio Dry Weight/ Wet Weight
525	7.84	82.5	0.175
781	15.0	82.8	0.172
760	12.2	87.5	0.125
757	14.0	84.6	0.154
602	10.9	83.4	0.166
748	14.7	82.8	0.172
702	14.3	82.5	0.175
876	12.0	83.4	0.166
754	14.2	86.6	0.134
944	14.5	86.7	0.133
841	12.9	84.1	0.159
876	11.2	88.8	0.112
961	11.0	89.0	0.110
690	13.8	86.2	0.138
682	13.6	86.4	0.136

Table 2. Cape Cod Canal Railroad Bridge No. 2 Fuel Oil Spill:
Subsamples of Mytilus edulis

Field Sample	Sample Wet Weight (g)		Sample Dry Weight (g)		Dry Weight/ Wet Weight (%)	Total Non-saponifiable Lipid Weight (mg)	
	A ^a	B ^a	A	B		A	B
CCCRB-830413	14.90	14.90	2.61	2.61	17.5	374.1 ^b	— ^c
CCCRB-830414	15.39	15.39	2.65	2.65	17.2	242.8 ^b	— ^c
CCCRB-830415	13.30	13.30	1.66	1.66	12.5	250.2 ^b	— ^c
CCCRB-830417	14.55	15.60	2.24	2.40	15.4	73.15	49.10
CCCRB-830420	14.58	15.20	2.42	2.52	16.6	47.16	40.06
CCCRB-830511	14.85	15.20	2.55	2.61	17.2	60.82	54.88
CCCRB-830519	15.40	16.34	2.70	2.86	17.5	51.00	55.54
CCCRB-830526	14.68	15.76	2.44	2.62	16.6	35.31	37.06
CCCRB-830803	15.33	15.39	2.05	2.06	13.4	39.29	41.98
CCCRB-840610	15.79	15.39	2.10	2.05	13.3	47.69	37.18
ESL Depuration Experiment							
830415	15.28	15.45	2.43	2.46	15.9	41.77	44.76
830417	15.63	15.38	2.77	2.72	11.2	56.18	58.28
830420	15.83	15.08	2.60	2.47	11.0	238.7 ^a	28.09
830511	15.83	15.56	2.18	2.15	13.8	160.19 ^a	31.23
830519	15.49	15.44	2.15	2.15	13.6	208.53 ^a	28.09

^aA and B are subsamples of the homogenate.

^bBack extraction - not done for these samples.

^cTotal non-saponifiable lipid weight of these samples was not measured.

Table 3. Recoveries of Selected Hydrocarbons Added to Samples
Hydrocarbon Analysis by GC

Sample	n-C ₁₄ (%)		Androstane (%)		n-C ₂₈ (%)	
	A	B	A	B	A	B
CCCRB-830413 ^a	777	146	149	247	126	113
CCCRB-830414 ^a	911	1250	122	279	88	93
CCCRB-830415 ^a	588	596	208	250	93	90
CCCRB-830417 ^a	135	170	122	120	101	99
CCCRB-830420 ^a	103	131	124	123	99	106
CCCRB-830511 ^a	108	100	118	102	109	101
CCCRB-830519	98	101	111	97	107	101
CCCRB-830526	98	101	111	108	107	106
CCCRB-830803	---	81	84	109	99	109
CCCRB-840610	57	78	100	85	92	77

^aAmount of internal standard added was too little in comparison to natural occurrence in sample from No. 2 fuel oil contamination. Data reflects contributions by compounds in tissue from oil contamination. Therefore cannot be used to estimate recoveries.

Table 4. Recoveries of Selected Aromatic Hydrocarbons:
Analysis by GC/MS

Sample	D ₁₀ -Fluorene			D ₁₄ -Terphenyl		
	Added (ng)	Recovered (ng)	%	Added (ng)	Recovered (ng)	(%)
<u>Analyst A</u>						
CCCRB-830413	291	16.7	5.7	250	98	39
CCCRB-830414	336	186	55	288	308	107
CCCRB-830415	478	260	54	408	378	93
CCCRB-830417	3020	1580	52	2600	2340	90
CCCRB-830420	2800	1190	42	2410	1860	77
CCCRB-830511	2660	1130	42	2280	664	73
CCCRB-830519	2520	1030	41	2160	1470	68
CCCRB-830526	2780	—	— ^a	2390	—	—
CCCRB-830803	2760	815	30	2370	1270	54
CCCRB-840610	1350	321	24	1160	782	68
<u>Analyst B</u>						
CCCRB-830413 ^b	291	225	77	250	386	154
CCCRB-830413 ^b	291	236	89	250	419	167
CCCRB-830414	336	152	45	288	251	87
CCCRB-830415	478	202	42	408	423	104
CCCRB-830417	2282	1780	63	2430	2290	94
CCCRB-830420	2730	1450	53	2350	1980	84
CCCRB-830511	2680	1170	44	2300	1630	71
CCCRB-830519	2410	935	39	2080	1480	72
CCCRB-830526	2590	952	37	2230	1450	65
CCCRB-830803	2750	763	28	2360	1210	51
CCCRB-840610	1380	345	25	1190	879	74

^aThis sample was not analyzed by GCMS because of loss during storage.

^bMeasured twice by GCMS.

Table 5. Cape Cod Canal Railroad Bridge No. 2 Fuel Oil Spill:
Comparison of Recoveries of Internal Standards Determined by
GC and GC/MS

Sample	D ₁₀ -Fluorene				D ₁₄ -Terphenyl			
	A		B		A		B	
	GC%	GC/MS%	GC%	GC/MS%	GC%	GC/MS%	GC%	GC/MS%
CCCRB-830413	44	5.7	62	77	237	39	174	154
CCCRB-830414	58	55	55	45	130	107	139	87
CCCRB-830415	27	54	31	42	56	93	121	104
CCCRB-830417	84	52	106	63	108	90	113	94
CCCRB-830420	69	42	104	53	101	77	107	84
CCCRB-830511	78	42	82	44	101	73	104	71
CCCRB-830519	71	41	64	39	100	68	97	71
CCCRB-830526	63	— ^c	76	37	89	— ^c	110	65
CCCRB-830803	63	30	65	28	101	54	106	51
CCCRB-840610	53	24	34	25	118	67	91	74
$\bar{X}$	61	43 ^a	68	45	100 ^b	74	110	78
$\pm$ s.d.	± 17	± 11	± 25	± 16	± 20	± 21	± 14	± 16

^aExcluding 5.7%

^bExcluding 237%, 174%, 154%

^cThis sample was not analyzed by GC/MS because of loss during storage.

Table 6. Cape Cod Canal Railroad Bridge No. 2 Fuel Oil Spill:
Alkane and Cycloalkane Concentrations in Mytilus edulis

(10^{-9} g/g dry weight)

Compound	CCCRB-830413 Day-1			CCCRB-830414 Day-2			CCCRB-830415 Day-3		
	A ^a	B ^a	$\bar{X}$	A ^a	C ^a	$\bar{X}$	A	B	$\bar{X}$
C ₁₁	305	— ^c	305	65	—	65	138	492	314
C ₁₂	423	—	423	193	60	127	140	215	178
C ₁₃	1680	209 ^b	1680	1260	616	937	764	858	811
C ₁₅	5340	4670	5010	4450	3880	4170	2310	2420	2460
C ₁₆	6180	6440	6310	4420	2960	3690	2560	1710	2140
C ₁₇	5690	5970	5830	2960	2840	2901	1650	1580	1610
Pristane	5340	5940	5640	7300	6800	7050	8770	7240	8000
C ₁₈	4440	5180	4810	2130	2070	2100	976	869	923
Phytane	3080	3670	3380	4290	4090	4190	4830	4310	4570
C ₂₀	3630	3320	3470	3150	2960	3060	1900	1360	1630
C ₂₂	1520	1860	1690	2090	2020	2060	2040	1830	1930
C ₂₄	425	550	488	674	573	624	693	735	714
C ₂₆	102	133	118	138	137	138	162	172	167
C ₃₀	—	61	61	45	53	49	69	66	68
C ₃₂	55	55	55	56	50	53	98	80	89

(10^{-6} g/g dry weight)

UCM	179	164	172	237	199	218	296	242	269
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^aA, B; A, C are subsamples of the homogenate analyzed by two different analysts.

^bData not included in $\bar{X}$ because of low recoveries or unusually low value compared to other data. n-Alkanes - C₁₁, C₁₂, etc.

^cBelow detection limit < 0.1×10^{-9} g/g.

^dContaminant interference - phthalate?

Table 6. (continued)

Compound	CCCRB-830417 Day-5			CCCRB-830420 Day-8			CCCRB-830511 Day-29		
	A	B	$\bar{X}$	A	B	$\bar{X}$	A	B	$\bar{X}$
C ₁₁	—	—	—	—	—	—	—	—	—
C ₁₂	147	98	123	60	52	56	32	44	38
C ₁₃	589	478	534	193	149	171	42	54	48
C ₁₅	1470	1530	1500	856	715	786	320	394	357
C ₁₆	1520	917	1220	519	458	489	252	260	256
C ₁₇	926	1140	1030	691	658	675	732	804	768
Pristane	6340	6030	6190	5270	4620	4950	790	722	756
C ₁₈	576	538	557	319	375	347	54	71	63
Phytane	3600	3430	3520	1770	2660	2710	322	375	349
C ₂₀	576	562	569	294	189	242	—	—	—
C ₂₂	1210	1280	1240	735	716	726	34	44	39
C ₂₄	483	562	523	410	477	444	90	63	77
C ₂₆	157	179	168	153	146	150	85	70	78
C ₃₀	100	121	111	92	113	103	84	93	89
C ₃₂	75	68	72	67	67	67	52	47	50
(10 ⁻⁶ g/g dry weight)									
UCM	236	238	237	220	193	207	110	125	118

Table 6. (continued)

Compound	CCCRB-830519 Day-37			CCCRB-830526 Day-44			CCCRB-830803 Day-113		
	A	B	$\bar{X}$	A	B	$\bar{X}$	A	B	$\bar{X}$
C ₁₁	--	71	71	--	85	85	--	--	--
C ₁₂	--	65	65	37	98	68	17	--	17
C ₁₃	37	--	37	55	59	57	16	--	16
C ₁₅	271	304	288	512	627	570	276	--	276
C ₁₆	143	175	159	234	212	223	66	--	66
C ₁₇	397	479	438	790	786	788	208	110	159
Pristane	538	541	540	443	504	474	184	97	141
C ₁₈	34	46	40	69	86	78	20	20	20
Phytane	225	273	249	215	260	238	54	26	40
C ₂₀	--	--	--	24	--	24	--	--	--
C ₂₂	--	--	--	--	--	--	10	--	10
C ₂₄	37	43	40	53	50	52	21	^d	21
C ₂₆	59	64	62	69	68	69	58	63	61
C ₂₈	88	94	91	89	101	96	112	84	98
C ₃₀	44	56	50	54	62	58	32	40	36
(10 ⁻⁶ g/g dry weight)									
UCM	100	118	109	97	119	108	36	27	32

Table 6. (continued)

Compound	CCCRB-840610 Day-424		
	A	B	$\bar{X}$
C ₁₁	48	19	34
C ₁₂	47	17	32
C ₁₃	20	—	20
C ₁₅	259	210	235
C ₁₆	70	82	76
C ₁₇	246	286	266
Pristane	131	161	146
C ₁₈	21	47	34
Phytane	31	50	41
C ₂₀	13	29	21
C ₂₂	19	26	23
C ₂₄	30	22	26
C ₂₆	50	68	59
C ₂₈	55	89	72
C ₃₀	59	69	64
(10 ⁻⁶ g/g dry weight)			
UCM	34	47	41

Table 7. Cape Cod Canal Railroad Bridge No. 2 Fuel Oil Spills:
Alkane and Cycloalkane Concentrations in Mytilus edulis:
Comparison of 1978 and 1983 Spill Data for
Representative Alkanes and f_1 UCM

$\bar{X}$, 10^{-9} g/g dry weight)

Compound	Day-1	
	1978	1983
C ₁₃	2200	1680
C ₁₅	4690	5010
C ₁₆	3715	6310
C ₁₇	2570	5830
Pristane	4570	5640
C ₁₈	2215	4810
Phytane	1730	3380
C ₂₄	587	488
$\bar{X}$, 10^{-6} g/g dry weight)		
UCM	199	172

Table 8. Cape Cod Canal Railroad Bridge No. 2 Fuel Oil Spill:
Aromatic Hydrocarbon Concentrations in Mytilus edulis

(10⁻⁹ g/g dry weight)

Compound	CCCRB-830413 Day-1						CCCRB-830414 Day-2					
	GC			GC/MS			GC			GC/MS		
	A ^a	B ^a	$\bar{X}$	A	B	$\bar{X}$	A	C	$\bar{X}$	A	C	$\bar{X}$
Naphthalene	— ^c	—	—	—	—	—	—	—	—	—	—	—
1-Methyl-naphthalene	69	—	69	38	—	38	—	108	108	36	45	40
2,6-Dimethyl-naphthalene	544	66 ^b	544	348	3.0 ^b	348	988	868	928	384	471	428
2,3,6-Tri-methylnaphthalene	1010	528	769	634	28 ^b	634	574	791	683	604	741	672
Fluorene	444	192 ^b	444	132	4.8 ^b	132	596	609	602	121	148	134
Phenanthrene	1060	816	938	598	151 ^b	598	1460	1230	1350	597	732	664
2-Methyl-phenanthrene	1340	980	1160	576	183 ^b	576	1200	1080	1140	685	841	763
1-Methyl-phenanthrene	44 ^b	674	674	274	85 ^b	274	1170	956	1060	343	420	382
3,6-Dimethyl-phenanthrene	406	500	453	278	72 ^b	278	545	559	552	314	386	350
Dibenzothio-phene	485	304	394	148	34 ^b	148	506	614	560	140	172	156
Fluoranthene	438	421	430	123	34 ^b	123	602	608	605	93	114	104
Pyrene	320	401	360	109	28 ^b	109	176	182	179	91	112	102
Chrysene	140	36 ^b	140	29	7.3 ^b	29	332	307	320	18	22	20

(10⁻⁶ g/g dry weight)

UCM	109	96	102				140	124	132
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^aExtractions, column chromatography, and gas chromatography by two analysts A and B or A and C for duplicates. GC/MS by one analyst.

^bData not included in $\bar{X}$ because of low recoveries or unusually low value compared to other data.

^cBelow detection limit < 0.1 x 10⁻⁹ g/g.

Table 8. (continued)
(10^{-9} g/g dry weight)

Compound	CCCRB-830415 Day-3						CCCRB-830417 Day-5					
	GC			GC/MS			GC			GC/MS		
	A	B	$\bar{X}$	A	B	$\bar{X}$	A	B	$\bar{X}$	A	B	$\bar{X}$
Naphthalene	—	—	—	—	—	—	—	—	—	—	—	—
1-Methyl-naphthalene	—	37	37	—	36	36	53	35	44	32	20	26
2,6-Dimethyl-naphthalene	110	358	234	87 ^b	419	419	650	470	560	424	305	364
2,3,6-Trimethyl-naphthalene	370	246	308	446	556	501	440	418	425	605	397	501
Fluorene	378	276	328	116	122	119	199	234	217	128	108	118
Phenanthrene	1630	612	1120	786	675	730	1400	1420	1410	691	629	660
2-Methyl-phenanthrene	1400	700	1050	990	832	911	950	1030	990	810	739	774
1-Methyl-phenanthrene	868	430	649	504	408	456	771	786	778	432	388	410
3,6-Dimethyl-phenanthrene	563	314	438	470	380	425	519	476	498	394	381	388
Dibenzothio-phene	128	278	203	189	162	176	640	573	606	167	154	161
Fluoranthene	698	302	500	171	136	154	545	535	540	162	155	159
Pyrene	354	151	252	160	123	142	313	257	285	137	128	133
Chrysene	365	168	266	29	24	27	—	—	—	25	28	27
(10 ⁻⁶ g/g dry weight)												
UCM	75	144	109				134	100	117			

^bData not included in $\bar{X}$ because of low recovery.

Table 8. (continued)
(10⁻⁹ g/g dry weight)

Compound	CCCRB-830420 Day-8						CCCRB-830511 Day-29					
	GC			GC/MS			GC			GC/MS		
	A	B	$\bar{X}$	A	B	$\bar{X}$	A	B	$\bar{X}$	A	B	$\bar{X}$
Naphthalene	--	--	--	--	--	--	--	--	--	--	--	--
1-Methyl-naphthalene	--	--	--	3.5	7.1	5.3	--	--	--	0.8	--	0.8
2,6-Dimethyl-naphthalene	244	320	282	115	184	150	--	--	--	4.0	5.0	4.5
2,3,6-Trimethyl-naphthalene	397	320	358	501	391	446	40	115	78	39	42	41
Fluorene	173	97	135	39	56	48	52	28	40	4.1	3.7	3.9
Phenanthrene	884	1060	972	345	434	390	247	--	247	49	51	50
2-Methyl-phenanthrene	745	738	742	458	596	527	137	--	137	86	83	85
1-Methyl-phenanthrene	622	665	643	261	336	298	210	134	172	64	60	62
3,6-Dimethyl-phenanthrene	502	461	482	272	350	311	205	400	302	137	142	140
Dibenzothio- phene	374	104	239	--	--	--	113	461	287	--	--	--
Fluoranthene	452	494	473	94	119	106	267	200	234	79	81	80
Pyrene	244	249	247	84	104	94	106	98	102	41	42	42
Chrysene	310	350	330	16	25	21	165	167	166	19	19	19
(10 ⁻⁶ g/g dry weight)												
UCM	102	122	112				61	78	70			

Table 8. (continued)
(10⁻⁹ g/g dry weight)

Compound	CCCRB-830519 Day-37						CCCRB-830526 Day-44					
	GC			GC/MS			GC			GC/MS		
	A	B	$\bar{X}$	A	B	$\bar{X}$	A	B	$\bar{X}$	A ^d	B	$\bar{X}$
Naphthalene	--	--	--	--	--	--	--	--	--	--	--	--
1-Methyl-naphthalene	--	--	--	--	0.5	0.5	--	--	--	--	--	--
2,6-Dimethyl-naphthalene	--	--	--	1.6	--	1.6	--	--	--	--	--	--
2,3,6-Trimethyl-naphthalene	19	16	18	14	25	20	--	28	28	--	--	--
Fluorene	28	18	23	2.9	2.9	2.9	--	--	--	--	--	--
Phenanthrene	164	174	169	34	40	37	56	132	94	--	19	19
2-Methyl-phenanthrene	66	--	66	40	40	40	--	--	--	--	24	24
1-Methyl-phenanthrene	110	86	98	33	37	35	22	73	47	--	16	16
3,6-Dimethyl-phenanthrene	180	332	256	98	95	97	780	230	504	--	62	62
Dibenzothio- -phene	84	180	132	--	--	--	25	206	116	--	--	--
Fluoranthene	105	86	96	77	79	78	26	112	69	--	55	55
Pyrene	--	--	--	33	35	34	72	87	80	--	23	23
Chrysene	117	108	112	18	18	18	113	137	125	--	16	16
(10 ⁻⁶ g/g dry weight)												
UCM	49	60	54				44	58	51			

^dSample not analyzed. Solvent evaporated to dryness in vial.

Table 8. (continued)
(10⁻⁹ g/g dry weight)

Compound	CCCRB-830803 Day-113						CCCRB-830610 Day-424					
	GC			GC/MS			GC			GC/MS		
	A	B	$\bar{X}$	A	B	$\bar{X}$	A	B	$\bar{X}$	A ^d	B	$\bar{X}$
Naphthalene	--	--	--	--	--	--	--	--	--	--	--	--
1-Methyl-naphthalene	--	--	--	--	0.2	0.2	--	--	--	--	--	--
2,6-Dimethyl-naphthalene	--	--	--	--	--	--	--	--	--	--	--	--
2,3,6-Trimethyl-naphthalene	--	--	--	--	--	--	--	--	--	--	--	--
Fluorene	--	--	--	0.8	--	0.8	--	--	--	1.3	1.8	1.6
Phenanthrene	--	--	--	5.9	8.2	7.1	--	--	--	13	16	15
2-Methyl-phenanthrene	--	--	--	3.8	5.7	4.8	--	--	--	5.4	7.4	6.4
1-Methyl-phenanthrene	--	--	--	3.0	2.8	2.9	--	--	--	4.3	5.2	4.8
3,6-Dimethyl-phenanthrene	--	--	--	3.9	4.3	4.1	--	--	--	4.7	8.0	6.4
Dibenzothio-phene	--	--	--	--	--	--	--	--	--	1.2	--	1.2
Fluoranthene	30	27	29	25	25	25	51	44	48	38	48	43
Pyrene	52	32	42	16	17	17	72	--	72	17	21	19
Chrysene	--	--	--	6.0	6.8	6.4	23	29	26	8.1	10	90
(10 ⁻⁶ g/g dry weight)												
UCM	4	19	12				3	12	8			

Table 9. Cape Cod Canal Railroad Bridge No. 2 Fuel Oil Spill:
Aromatic Hydrocarbon Concentrations in Mytilus edulis

(10^{-9} g/g dry weight)^a

Compound	CCCRB-830413 Day-1			CCCRB-830414 Day-2			CCCRB-830415 Day-3		
	A	B	$\bar{X}$	A	C	$\bar{X}$	A	B	$\bar{X}$
Naphthalene	—	— ^b	—	—	—	—	—	—	—
C ₁ -Naphthalenes	91	—	91	254	111	182	—	71	71
C ₂ -Naphthalenes	1310	11 ^c	1310	3160	1650	2400	369	1350	1350
C ₃ -Naphthalenes	2520	185 ^c	2520	5670	4590	5130	3250	4110	3680
Phenanthrene	598	151 ^c	598	597	732	664	786	675	730
C ₁ -Phenanthrenes	1920	519 ^c	1920	3270	2550	2910	3050	2570	2820
C ₂ -Phenanthrenes	2340	567 ^c	2340	3790	3020	3410	3520	3160	3340

Compound	CCCRB-830417 Day-5			CCCRB-830420 Day-8			CCCRB-830511 Day-29		
	A	B	$\bar{X}$	A	B	$\bar{X}$	A	B	$\bar{X}$
Naphthalene	—	—	—	—	—	—	—	—	—
C ₁ -Naphthalenes	86	49	68	11	22	17	—	—	—
C ₂ -Naphthalenes	1430	—	1430	402	632	517	—	—	—
C ₃ -Naphthalenes	4770	—	4770	1970	2950	2460	218	474	346
Phenanthrene	691	629	660	345	434	390	49	51	50
C ₁ -Phenanthrenes	2510	2350	2430	1510	1930	1720	340	352	346
C ₂ -Phenanthrenes	3020	2750	2882	1930	2410	2170	687	690	689

^aGC/MS: Manual search series.

^bNot detected: generally less than 0.1×10^{-9} g/g dry weight.

^cData not included in $\bar{X}$, recoveries low.

Table 9. (continued)
(10⁻⁹ g/g dry weight)

Compound	CCCRB-830519 Day-37			CCCRB-830526 Day-44			CCCRB-830803 Day-113		
	A	B	$\bar{X}$	A	B	$\bar{X}$	A	B	$\bar{X}$
Naphthalene	—	—	—	—	—	—	—	—	—
C ₁ -Naphthalenes	—	—	—	—	—	—	—	—	—
C ₂ -Naphthalenes	—	—	—	—	—	—	—	—	—
C ₃ -Naphthalenes	—	—	—	—	—	—	—	—	—
Phenanthrene	34	40	37	—	19	19	5.9	8.2	7.1
C ₁ -Phenanthrenes	124	172	148	—	81	81	11	9.1	10
C ₂ -Phenanthrenes	405	378	392	—	201	210	14	14	14

Compound	CCCRB-840610 Day-424		
	A	B	$\bar{X}$
Naphthalene	—	—	—
C ₁ -Naphthalenes	—	—	—
C ₂ -Naphthalenes	—	—	—
C ₃ -Naphthalenes	—	—	—
Phenanthrene	13	16	15
C ₁ -Phenanthrenes	16	24	20
C ₂ -Phenanthrenes	18	28	23

Table 10. Cape Cod Canal Railroad Bridge No. 2 Fuel Oil Spills:
Aromatic Hydrocarbon Concentrations in Mytilus edulis:
Comparison of 1978 and 1983 Spill Data for Representative
Aromatic Compounds and f₂ UCM

$\bar{X}$ in 10⁻⁹ g/g dry weight)

Compounds	Day-1	
	1978	1983
C ₂ -Naphthalenes	5620	1310
C ₃ -Naphthalenes	7850	2520
Phenanthrene	490	598
C ₁ -Phenanthrenes	1970	1920
C ₂ -Phenanthrenes	940	2340

Table 11. ESL Depuration Experiment: Alkanes and Cycloalkanes in *Mytilus edulis*(10⁻⁹ g/g dry wt.)

	830415 Day-3			830417 Day-5			830420 Day-8			830511 Day-29			830519 Day-37		
	A	B	$\bar{X}$	A	B	$\bar{X}$	A	B	$\bar{X}$	A	B	$\bar{X}$	A	B	$\bar{X}$
C ₁₁	153	— ^a	153	91	—	91	2070	1500	1790	2300	429	1370	2040	687	1360
C ₁₂	294	173	234	124	60	92	147	151	159	110	67	89	111	70	91
C ₁₃	921	728	825	223	141	182	115	136	126	46	41	44	28	—	28
C ₁₅	1690	1780	1740	595	661	628	424	466	445	327	332	329	194	210	202
C ₁₆	1500	1420	1460	325	298	312	206	226	216	249	174	211	166	126	146
C ₁₇	809	993	901	437	520	479	465	538	502	442	448	445	517	509	513
Pristane	4190	4200	4200	2980	2930	2960	2020	1870	1950	350	351	351	228	182	205
C ₁₈	457	539	498	185	123	154	84	67	76	49	45	47	31	—	31
Phytane	2300	2390	2350	1630	1450	1540	1040	962	1000	166	170	168	101	82	92
C ₂₀	729	539	634	154	84	119	—	—	—	—	—	—	—	—	—
C ₂₂	847	930	889	418	418	418	157	136	147	35	—	35	80	—	80
C ₂₄	295	392	344	269	218	244	152	143	148	54	—	54	133	—	133
C ₂₆	129	232	181	120	107	114	118	107	113	56	48	52	165	—	165
C ₃₀	118	136	127	94	104	99	104	65	85	63	50	57	178	—	178
C ₃₂	57	79	68	53	55	54	51	43	47	44	—	44	127	—	127
C ₃₄	44	—	44	42	—	42	—	87	87	—	—	—	56	—	56

(10⁻⁶ g/g dry weight)

UCM	170	236	203	164	178	171	128	124	126	68	57	63	51	44	48
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^aNone detected. Less than 0.1 x 10⁻⁹ g/g dry wt.

Table 12. ESL Depuration Experiment: Aromatic Hydrocarbon Concentration in Mytilus edulis
(10^{-9} g/g dry wt.)

	850415 Day-3				850417 Day-5				850420 Day-8				850511 Day-29				850519 Day-37			
	A ^a	B	$\bar{X}$	A	B	$\bar{X}$	A	B	A	B	$\bar{X}$	A	B	A	B	$\bar{X}$	A	B	$\bar{X}$	
2,6-Dimethyl-naphthalene	261	722	722	333	524	429	175	—	175	—	—	—	—	—	—	—	—	—	—	
2,3,6-Trimethyl-naphthalene ^b	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	
Fluorene	78	715	715	117	120	119	138	—	138	—	—	25	—	25	—	25	26	—	26	
Dibenzothio- phen	41	1050	1050	271	530	401	348	732	540	46	115	80	102	144	123	—	—	—	—	
Phenanthrene	521	1730	1730	714	1020	867	741	581	661	140	208	174	91	—	91	—	—	—	91	
2-Me-phenan- threne	460	1130	1130	530	748	639	454	531	493	142	—	142	72	—	—	—	—	—	72	
1-Me-phenan- threne	412	1140	1140	493	643	568	401	513	457	127	157	142	99	99	99	—	—	—	99	
3,6-Dimethyl- phenanthrene	224	978	978	282	335	309	260	270	265	121	247	184	99	250	175	—	—	—	175	
Fluoranthene	288	1080	1080	308	438	373	316	332	324	143	170	157	116	148	132	—	—	—	132	
Pyrene	152	741	741	134	211	173	165	163	164	95	—	95	64	90	77	—	—	—	77	
(10 ⁻⁶ g/g dry weight)																				
UCM	40	73	57	40	87	64	65	53	59	35	42	39	30	36	33	—	—	—	33	

^aData not included in $\bar{X}$ because of low recoveries or unusually low value compared to other data.

^bNone detected. Less than 0.1×10^{-9} g/g dry wt.

Table 13. Comparison of Cape Cod Canal Field Site and ESL Data for Alkanes and UCM

($\bar{X}$, 10^{-9} g/g dry weight)^a

	Day-3		Day-5		Day-8		Day-29		Day-37	
	CCC	ESL	CCC	ESL	CCC	ESL	CCC	ESL	CCC	ESL
C ₁₅	2460	1740	1500	628	786	445	357	329	288	202
C ₁₇	1610	901	1030	479	675	502	768	445	438	513
Pristane	8000	4200	6190	2960	4950	1950	756	351	540	205
C ₁₈	923	498	557	154	347	76	63	47	40	31
Phytane	4570	2350	3520	1540	2710	1000	349	168	249	92
C ₂₂	1930	889	1240	418	726	147	39	35	—	80
C ₂₄	714	344	523	244	444	148	77	54	40	133

(10^{-6} g/g dry weight)

UCM	269	203	237	171	207	126	118	63	109	48
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^aData for $\bar{X}$ from Tables 6 and 11.

Table 14. Biological Half-lives (Tb 1/2)^a of No. 2 Fuel Oil Compounds:
1983 Oil Spill

Compounds	<u>Day-1 to Day-29</u>			Tb 1/2
	a	b	r ²	
n-C ₁₆	3022	-0.10	0.70	6.9
Pristane	8458	-0.08	0.96	8.7
Phytane	5045	-0.09	0.97	7.7
n-C ₂₃	1480	-0.10	0.98	6.9
Σ C ₂ -Naphthalenes	4747	-0.36	0.98	1.9
Σ C ₃ -Naphthalenes	4895	-0.09	0.89	7.7
Phenanthrene	852	-0.10	0.97	6.9
Σ C ₁ -Phenanthrenes	3029	-0.07	0.94	9.9
Σ C ₂ -Phenanthrenes	3400	-0.05	0.92	14
UCM f ₁	235	-0.02	0.66	35
UCM f ₂	122	-0.02	0.79	35

^aEquation is $y = ae^{bx}$; y = concentration in mussel tissue, x = time in days, a = intercept y axis, b = slope of line in semilog plot, r² = correlation fit of data to equation, Tb 1/2 = $\frac{\ln 2}{b}$ is biological half-life of rate of loss of compound from mussels.

Table 15. Biological Half-lives ($T_{b\ 1/2}$)^a of No. 2 Fuel Oil Compounds:
1983 Oil Spill

Compounds	a	<u>Day-1 to Day-29</u>		$T_{b\ 1/2}$
		b	r^2	
1-Methylnaphthalene	42	-0.14	0.91	5.0
2,6-Dimethylnaphthalene	600	-0.17	0.98	4.1
2,3,6-Trimethylnaphthalene	808	-0.11	0.98	6.3
Phenanthrene	842	-0.10	0.97	6.9
2-Methylphenanthrene	933	-0.08	0.93	8.7
1-Methylphenanthrene	453	-0.07	0.89	9.9
3,6-Dimethylphenanthrene	390	-0.03	0.80	23

^aSee footnote, Table 14.

Table 16. Biological Half-lives ($T_{b\ 1/2}$)^a of No. 2 Fuel Oil Compounds:
1983 Oil Spill

Compounds	a	<u>Day-3 to Day-29</u>		$T_{b\ 1/2}$
		b	r^2	
n-C ₁₆	1496	-0.08	0.77	8.7
Pristane	8914	-0.11	1.00	6.3
Phytane	5181	-0.12	1.00	5.8
n-C ₂₃	1349	-0.12	1.00	5.8
$\Sigma\ C_2$ -Naphthalenes	4345	-0.46	0.98	1.5
$\Sigma\ C_3$ -Naphthalenes	5149	-0.12	0.97	5.8
Phenanthrene	868	-0.12	1.00	5.8
$\Sigma\ C_1$ -Phenanthrenes	3134	-0.10	1.00	6.9
$\Sigma\ C_2$ -Phenanthrenes	3503	-0.07	1.00	9.9
UCM f ₁	267	-0.04	0.99	17
UCM f ₂	121	-0.02	0.93	35

^aSee footnote, Table 14.

Table 17. Biological Half-lives ($T_{b\ 1/2}$)^a of No. 2 Fuel Oil Compounds:
1983 Oil Spill

Compounds	<u>Day-3 to Day-29</u>			$T_{b\ 1/2}$
	a	b	r^2	
1-Methylnaphthalene	28	-0.14	0.89	5.0
2,6-Dimethylnaphthalene	514	-0.18	1.00	3.9
2,3,6-Trimethylnaphthalene	667	-0.10	0.98	6.9
Phenanthrene	813	-0.10	1.00	6.9
2-Methylphenanthrene	977	-0.09	1.00	7.7
1-Methylphenanthrene	495	-0.08	1.00	8.7
3,6-Dimethylphenanthrene	429	-0.04	0.99	17

^aSee footnote, Table 14.

Table 18. Biological Half-lives ($T_{b\ 1/2}$)^a of No. 2 Fuel Oil Compounds:
1978 Oil Spill

Compounds	<u>Day-1 to Day-21</u>			$T_{b\ 1/2}$
	a	b	r^2	
n-C ₁₆	6724	-0.7	1.00	0.99
Pristane	5315	-0.09	0.98	7.7
Phytane	1976	-0.10	0.97	6.9
n-C ₂₃	1228	-0.15	1.00	4.6
$\Sigma\ C_2$ -Naphthalenes	5616	-0.12	0.99	5.8
$\Sigma\ C_3$ -Naphthalenes	8169	-0.09	0.99	7.7
Phenanthrene	451	-0.04	0.88	17
$\Sigma\ C_1$ -Phenanthrenes	1886	-0.07	0.93	9.9
$\Sigma\ C_2$ -Phenanthrenes	891	-0.01	0.32	69
UCM f_1	171	-0.05	0.87	14
UCM f_2	110	-0.05	0.93	14

^aSee footnote, Table 14.

Table 19. Comparison of Biological Half-lives of No. 2 Fuel Oil Compounds: 1978 and 1983 Oil Spill

Compounds	<u>Tb 1/2 in Days</u>	
	1978 ^a Day 1 - Day 21	1983 Day 3 - Day 29
n-C ₁₆	.99	8.7
Pristane	7.7	6.3
Phytane	6.9	5.8
n-C ₂₃	4.6	5.8
Σ C ₂ -Naphthalenes	5.8	1.5
Σ C ₃ -Naphthalenes	7.7	5.8
Phenanthrene	17	5.8
Σ C ₁ -Phenanthrenes	9.9	6.9
Σ C ₂ -Phenanthrenes	69 ^b	9.9
UCM f ₁	14	17
UCM f ₂	14	35

^aRecalculated for 1978 data. These are slightly different from those reported in Farrington et al. (1982a) because of an error in a calculator program discovered since that publication.

^bCorrelation fit of data to equation is poor.

Figure Legends

- Figure 1. Chart of the Massachusetts Coast with locations of sampling stations for U.S. Mussel Watch Program 1976-1978 and Environmental Systems Laboratory (ESL), Woods Hole Oceanographic Institution.
- Figure 2. 1983 Oil Spill. Glass capillary gas chromatograms of alkanes and cycloalkanes of sample of No. 2 fuel oil from the tugboat and extracted from tissue of Mytilus edulis sampled at the Cape Cod Canal Railroad Bridge. I.S. indicates internal standard. QS indicates quantitation standard. Abbreviated notations are used for n-alkanes, e.g. C₁₅ is n-pentadecane. CCCRB is abbreviation for Cape Cod Canal Railroad Bridge. Code numbers such as 830413 indicate year, month day. f₁ Indicates column chromatography fraction one. Details of analyses are given in Materials and Methods section of report.
- Figure 3. Semilog plot of concentrations of representative alkanes and unresolved complex mixture (UCM) in tissue of Mytilus edulis as a function of time for 1983 No. 2 fuel oil spill. Abbreviated notation used for n-alkanes, e.g. n-C₁₆ is n-hexadecane. Average concentrations plotted from data of Table 6. Dashed line indicates extrapolation to non-detectable concentrations.
- Figure 4. 1978 Oil Spill. Glass capillary gas chromatograms of alkanes and cycloalkanes extracted from tissues of Mytilus edulis sampled at the Cape Cod Canal Railroad Bridge. Numbers on the x-axis at scale marks indicate position of elution of corresponding n-alkanes, e.g. 18 indicates n-octadecane. PRIS indicates pristane and PHYT indicates phytane, UCM indicates unresolved complex mixture, CCC is an abbreviation for Cape Cod Canal, sample codes such as 78-04-02 f₁ indicate year, month, day. f₁ Indicates column chromatography fraction one. Figure from previous study of 1978 oil spill and analysis details are in Farrington et al. (1982a).
- Figure 5. 1983 Oil Spill. Glass capillary gas chromatograms of aromatic compounds of No. 2 fuel oil from the tugboat and extracted from tissue of Mytilus edulis sampled at the Cape Cod Canal Railroad Bridge. QS indicates quantitation standard. Abbreviations are: 1-meN, 1-methylnaphthalene; 2,6-dimeN, 2,6-dimethylnaphthalene; 2,3,6-trimeN, 2,3,6-trimethylnaphthalene; 2-meP, 2-methylphenanthrene; 1-meP, 1-methylphenanthrene; 3,6-dimeP, 3,6-dimethylphenanthrene. I.S. indicates the internal standards deuterated (D₁₀) fluorene and deuterated (D₁₄) terphenyl. Peaks eluting after QS are probably alkenes or cycloalkenes of presumed biogenic origin. Sample codes are the same as for Figure 2.

- Figure 6. Semilog plot of concentrations of representative aromatic hydrocarbons and unresolved complex mixture (UCM) in Mytilus edulis tissue as a function of time for 1983 No. 2 fuel oil spill. Data from Table 9. Dotted line indicates extrapolation to non-detectable concentrations.
- Figure 7. 1978 Oil Spill. Glass capillary gas chromatograms of aromatic compounds extracted from tissue of Mytilus edulis sampled Cape Cod Canal Railroad Bridge. Numbers and scale on x axis indicate position of elution of n-alkanes for reference. Peaks eluting between 20 and 22 carbon n-alkanes and peaks eluting after 22 are alkenes (olefins) or cycloalkenes of presumed biogenic origin. UCM indicates unresolved complex mixture.
- Figure 8. Semilog plot of concentrations of representative alkanes and unresolved complex mixture (UCM) in tissue of Mytilus edulis as a function of time for 1983 No. 2 fuel oil spill ESL depuration experiment. Also see legend Figure 3.
- Figure 9-19. Semilog plots of concentrations of hydrocarbons in tissue of Mytilus edulis vs. time for 1978 and 1983 oil spills. Dashed lines are extrapolations to non-detectable concentrations.
- Figure 20. Compilation of tracings of computer reconstructed molecular ion plots from GCMS data showing relative abundances of resolved or partially resolved isomers of C₂-phenanthrenes (m/z 206) and C₃-phenanthrenes (m/z 220). Stippled peaks decrease in abundance relative to clear or open peaks. Lined peaks, also indicated by x, are m/z 220 signals from compounds other than C₃-phenanthrenes.

Figure 1.

FLAME IONIZATION DETECTOR RESPONSE

No. 2 Fuel Oil f₁

CCCRB 830413 f₁
DAY 1

CCCRB 830414 f₁
DAY 2

CCCRB 830415 f₁
DAY 3

Figure 2.

FLAME IONIZATION DETECTOR RESPONSE

CCCRB 830511 f₁
DAY 29

CCCRB 830519 f₁
DAY 37

CCCRB 840610 f₁
DAY 424

Figure 2.

Concentrations of Alkanes and UCM After Spill

Figure 3.

FLAME IONIZATION DETECTOR RESPONSE

Figure 4.

FLAME IONIZATION DETECTOR RESPONSE

Figure 4.

FLAME IONIZATION DETECTOR RESPONSE

No. 2 Fuel Oil f_2

CCCRB 830413 f_2
DAY 1

CCCRB 830414 f_2
DAY 2

CCCRB 830415 f_2
DAY 3

Figure 5.

FLAME IONIZATION DETECTOR RESPONSE

Figure 5.

Concentrations of Aromatic Compounds After Spill (Quantitative GCMS Analysis)

Figure 6.

FLAME IONIZATION DETECTOR SIGNAL

Figure 7.

FLAME IONIZATION DETECTOR SIGNAL

Figure 7.

ESL Depuration Experiment
Concentrations of Alkanes and UCM
After Spill

Figure 8.

Figure 9.

Figure 10.

Figure 11.

Figure 12.

Figure 13.

Figure 14.

Figure 15.

Figure 16.

Figure 17.

Figure 18.

Figure 19.

GCMS MASS CHROMATOGRAMS f_2

Figure 20.

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16. Abstract (Limit: 200 words) Retention and release of No. 2 fuel oil compounds by <u>Mytilus edulis</u> contaminated by a small oil spill in the Cape Cod Canal in 1983 has been studied for the population "in situ" and for a subsample transplanted to a clean laboratory seawater system. Compounds analyzed include C ₁₃ to C ₂₄ n-alkanes; pristane; phytane; C ₂ -, C ₃ -naphthalenes; fluorene; phenanthrene; C ₁ -, C ₂ -, C ₃ -phenanthrenes; fluoranthene; pyrene and dibenzothiophene. Biological half-lives were determined for the compounds from Day-3 to Day-29 following the spill and ranged from 1.5 days for C ₂ -naphthalenes to 9.9 days for C ₂ -phenanthrenes. Biological half-lives for the unresolved complex mixture determined by gas chromatographic analyses were 17 days for the alkane-cycloalkane fraction and 35 days for the aromatic fraction. Results compared favorably with data from a small oil spill contaminating the same mussel population at the same time of the year in 1978, although marked differences were noted for certain parameters. Gas chromatographic-mass spectrometer analyses of C ₂ - and C ₃ -phenanthrenes revealed changes in relative abundance of compounds within isomer groups from samples at Day-29 to the time when no further detection of fuel oil was noted. These results suggest a release of metabolism of these compounds which is molecular structure specific. This study also demonstrated the feasibility of training an analyst unfamiliar with analyses of hydrocarbons in tissues to conduct high resolution glass capillary GC analyses and some aspects of GCMS data systems output within a period of four to five months.			
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