EFFECTS OF DRILLING MUDS ON LOBSTER BEHAVIOR

A progress report, of work done with grants from
the Environmental Protection Agency (#R 806630010)
and
the Department of Energy (# EY-76-S-02-2546.A003)

from 1 January to 1 October 1979

Jelle Atema

Linda Ashkenas

Eleanore Beale

Leonor Botero

Charles D. Derby

Dana V. Devine

Randall Elgin

Shelley Gibbons

Elisa B. Karnofsky

Susan Oleszko-Szuts

Thomas J. Trott

DISCLAIMER

This book was prepared as an account of work sponsored by an agency of the United States Government. Neither the United States Government nor any agency thereof, nor any of their employees, makes any warranty, express or implied, or assumes any legal liability or responsibility for the accuracy, completeness, or usefulness of any information, apparatus, product, or process disclosed, or represents that its use would not infringe privately owned rights. Reference herein to any specific commercial product, process, or service by trade name, trademark, manufacturer, or otherwise, does not necessarily constitute or imply its endorsement, recommendation, or favoring by the United States Government or any agency thereof. The views and opinions of authors expressed herein do not necessarily state or reflect those of the United States Government or any agency thereof.

MASTER

DISTRIBUTION OF THIS DOCUMENT IS UNLIMITED

DISCLAIMER

This report was prepared as an account of work sponsored by an agency of the United States Government. Neither the United States Government nor any agency Thereof, nor any of their employees, makes any warranty, express or implied, or assumes any legal liability or responsibility for the accuracy, completeness, or usefulness of any information, apparatus, product, or process disclosed, or represents that its use would not infringe privately owned rights. Reference herein to any specific commercial product, process, or service by trade name, trademark, manufacturer, or otherwise does not necessarily constitute or imply its endorsement, recommendation, or favoring by the United States Government or any agency thereof. The views and opinions of authors expressed herein do not necessarily state or reflect those of the United States Government or any agency thereof.

DISCLAIMER

Portions of this document may be illegible in electronic image products. Images are produced from the best available original document.

Table of Contents

	Page
Introduction	1
I. Effects of Drilling Mud on Adult and Subadult Lobsters:	
Feeding and Survival	5
II. Lobster Behavior in a Naturalistic Aquarium	13
III. Field Studies	29
IV. Data Processing	52
V. Experiments on Larval Settling and Substrate Behavior	56
VI. Effects of Drilling Mud on Post-Larval Lobsters:	
Feeding, Molting and Survival	79
VII. Neurophysiological Analysis of the Effects of Drilling	
Mud on the Activity of Chemoreceptors	84
VIII. "Boxing Matches" as a Bioassay for Pollutant Effects	89

Introduction

Drilling muds, used and discarded in great quantities during the drilling phase of exploration and production of oil wells, represent an unknown threat to the marine environment. The compositions of the muds vary greatly with drilling requirements. The toxicity of their components are largely unknown, but can range from apparently harmless to immediately lethal, as found recently in toxicity tests on a number of marine animals. Our laboratory has since late 1978 shifted its focus from the effects of petroleum hydrocarbons to the effects of drilling muds on the behavior of lobsters.

The lobster, Homarus americanus, was chosen for the study of drilling mud effects for a number of reasons: a) It lives in and on the floor of inshore and continental shelf waters, areas now being leased for drilling. Its benthic and shelter dependent existence insures that it will come in contact with drilling muds. b) It supports an important commercial and recreational fisheries, and "the Maine lobster" stands almost as a symbol of the entire New England coast. c) A considerable amount of background data is available on this animal, including oil toxicity studies. This greatly facilitates the interpretation of results from drilling mud toxicity studies.

Toxicity studies must be carried out under strictly controlled laboratory conditions to be able to compare the toxicity levels of different drilling muds and components of the mud, and to compare toxicities between organisms at different stages in their life cycles and in different seasons. To interpret with any confidence the results from strictly controlled toxicity studies in the laboratory, one must understand the animal's field conditions and preferably verify the laboratory results with field studies. Field studies are not always feasible and

are nearly always difficult to control. Therefore an intermediate step can be employed successfully as shown by our recent studies on lobster behavior. These studies employ naturalistic large aquaria, where a small group of animals can be observed for extensive periods of time under semi-natural conditions. Animals of all stages of development can be tested with this technique. Long-term observations of behavior generate a bulk of data which - with the aid of a computer - can be analyzed for effects of size, sex, season, stage in the molt cycle, past experience, etc. Without such basic information on the animal's biology, effects of pollutants on vital behavior such as mating and feeding cannot be interpreted: e.g., what appears as an effect of drilling mud exposure may actually be caused by a combination of pre-molt aggression and lack of food. Our naturalistic studies are complemented and verified by field observations during the entire warm season from March through October.

Lobster larvae spend their first month in the water column. Then, during the fourth larval stage, lobsters must commit themselves to a benthic existence or die. If they do not find the right substrate their chances of survival are greatly diminished. We have therefore begun to analyze some of the factors that are important in settling, such as phototaxis and substrate quality. These studies provide a background which is now used for toxicity studies, where drilling mud is covering the substrate in part or entirely.

Evaluating effects of pollutants on social behavior is important but difficult. Our attempt at a controlled laboratory study is the "boxing match", a technique we have successfully used for the study of drug effects on aggression and neuromuscular control.

Since lobsters use chemical cues from their environment to direct a great number of vital behavioral responses such as feeding, courtship and larval

settlement, we study the effects of pollutants on the two major chemoreceptor systems. The antennular system (i.e., smell) is used normally to make the lobster alert to the presence of food and to identify other lobsters. The latter involves pheromone detection and is essential in social behavior. Antennules also convey important directional information and thus help the spatial orientation of the lobster. The dactyl chemoreceptor system (i.e., taste) is directly concerned with food evaluation and feeding behavior. The neurophysiological assay is relatively quick and can serve to compare pollutant effects. Because physiological mechanisms are similar among animals, the results obtained with lobster chemoreception may be extrapolated with some confidence to other marine animals.

The various components of our study on the effects of drilling muds on lobster behavior are represented in a flow diagram (Fig. 1). We believe that only a multidisciplinary study of all life stages can truly evaluate sublethal toxicity.

This report contains eight sections, each describing an aspect of our studies of lobster behavior, ecology, physiology and the effects of exposure to various levels of different drilling muds. We have received various drilling muds from the EPA which will be referred to as DM-1, DM-2, etc. throughout the paper; they are identified in Materials and Methods of Section I.

Fig. 1. Relationship of projects currently in progress under EPA-DOE sponsorship. Roman numerals refer to projects described in this report. Toxicity studies cannot be understood in vacuo but must be based on an understanding of animal behavior and ecology.

I. EFFECTS OF DRILLING MUD ON ADULT AND SUBADULT LOBSTERS:

FEEDING AND SURVIVAL

Susan Oleszko-Szuts, Shelley Gibbons, and Jelle Atema

Dose-response tests on adult and subadult lobsters (Homarus americanus) are being used to establish toxicity levels of drilling mud and drilling mud effluents; behavioral analysis of feeding is used to describe sublethal effects. These tests are patterned after well-established procedures used successfully in similar work on effects of various petroleum fractions on lobster behavior.

Materials and Methods

Ten lobsters (5 male and 5 female, carapace length 57-88 mm) were placed individually in 100-liter aquaria which contain a 1-inch layer of sand substrate and a glazed clay shelter. They are supplied with running seawater ($T = 8^{\circ} \text{C}$). The flow-through system (Fig. I. 1) consists of one control and one experimental head tank. The experimental head tank supplies six individual 100-liter tanks. The overflow from the individual tanks enters a holding tank where 5 additional lobsters can be individually held to determine the toxic effects of drilling mud effluent (Effluent Exposure). The control head tank supplies four individual 100-liter tanks. Individual tanks, both experimental and control, have inflows ranging from 400-460 ml/min.

The lobsters are allowed to acclimate to laboratory conditions for at least two weeks. They are fed pieces of mussel shortly before introduction of mud and their behavior is observed. Mud is poured into each tank on a one-time-only basis.

In the first test in Dec 78, three experimental tanks (#s 3, 5, 9) received 1700 ml of drilling mud (Heavy Exposure) and three (#s 4, 8, 10) received

Figure I. 1 Flow-through dosing system for adult and subadult lobsters. The system consists of two head tanks, one control and one experimental. The experimental head tank, 2.4 m x 28 cm x 20 cm, is fitted with three baffles to aid in the layering-out of any suspended matter. Its inflow is 4,000 ml/min. Pollutants can be added at the seawater inflow or at any point along the tank. The overflow of the head tank is skimmed off into a collecting tank where further removal of the pollutant can be achieved. From a mid-depth standpipe in the head tank, water enters individually six 100-liter aquaria. The overflow from the six aquaria enters a holding tank where lobsters can be stored for exposure to pollutants. Overflows from the collecting tank and holding tank enter a filter box where pollutant is filtered out with acrylic fiber in a series of compartments before the water enters the drain. The control head tank, 1.2 m x 28 cm x 20 cm, individually supplies four 100-liter aquaria. Its inflow is 2,600 ml/min. Individual aquaria, both experimental and control, have inflows ranging from 400-460 ml/min.

100 ml of drilling mud (Light Exposure). The higher concentration settled out to a layer of about 10-12 mm over the sand substrate; the lower concentration reached a depth of about 1-2 mm. Two control tanks (#s 2 and 6) received 1700 ml of natural ocean bottom mud and two (#s 1 and 7) were left undisturbed. In a subsequent test in Sep 79, three experimental tanks (Heavy Exposure) received 1000 ml drilling mud instead of 1700 ml. Because no behavioral differences were seen between seawater and clean mud controls in Dec 78, controls in the latter experiment were not treated with clean mud. The lobsters are observed at morning and afternoon feedings for four days. The time it takes each lobster to alert to the presence of food (Alert) and find the food (Hit) are noted. After four days of exposure the lobsters are removed from the system and frozen for future chemical analysis. Several drilling muds, provided by Dr. Norman Richards, EPA Laboratory, Gulf Breeze, Florida, have been tested with this experimental design. In this report, the mud tested in Dec 78 will be referred to as DM-1 and in Sep 79 as DM-2. DM-2 is marked "control #16, 10" by the EPA.

Results

DM-1:

The most severe effects of DM-1 appeared in those lobsters which were exposed to the highest concentrations of drilling mud and to drilling mud effluents.

Heavy Exposure: After only four hours from the addition of the drilling mud, two of the three lobsters showed serious effects: one was lying on his back moving only his walking legs (Fig. I. 2); the other had apparently tail-flipped into a corner and was stuck standing on his head with his rostrum buried up to his eyestalks, with apparently no capability of righting himself. Neither of these animals regained mobility over the course of the experiment, but be-

Figure I. 2. Response of lobster to Heavy Exposure of DM-1.¹⁰⁰
The lobster was in this position four hours after the addition of the drilling mud. He displayed only feeble movements of his walking legs and antennules, even after prodding, and did not recover in four days.

came weaker as time progressed. Even poking them provoked only the most feeble response from a few legs. The third lobster of the heavy exposure group showed a reduction in physical coordination after six hours of exposure and severe effects similar to the other two after eight hours. While this animal was always more active than the other two heavily exposed ones (the water in his tank was always turbid, the mud never having a chance to settle), he also weakened progressively over the four days. None of these three animals ate after the addition of mud and initially gave only weak signs of alerting to the presence of food, even when the food was dropped directly on their antennules and dactyls. It is important to note the rapidity of response of the animals to a realistic dose of this drilling mud (10-12 mm bottom cover). Faced with a sudden deluge of this mud, the animals would have difficulty walking away from the exposed area within four hours.

Effluent exposure: Two of the lobsters exposed to the drilling mud effluent in the holding tank died before the end of the experiment. Three lobsters of this group showed increasing inability to coordinate movements and at the end of the experiment were essentially comatose.

Light exposure: The lobsters exposed to light concentrations of DM-1 were less affected than the others. They remained active to varying degrees throughout the experiment, digging and bulldozing the substrate so that after 6 hr no mud was obvious as a layer on top of the sand. Compared to pre-exposure behavior, however, they appeared to be less well-coordinated. One obvious effect of the mud exposure was the cessation of feeding activity. All three animals alerted to the presence of food, but failed to find it - either because they never searched or because, if they did touch the food, they seemed to be

unaware of it. Two of these animals did, however, recover: one was eating after 30 hr and the other after 54 hr from the time of addition of drilling mud.

Controls: None of the control lobsters, neither those in clean sand, nor those in sand with the addition of nonpolluted mud, changed their behavior over the four days. The "mudded" lobsters, especially, seemed very active (Fig. I. 3), their tanks were always turbid, and they always alerted to and ate the pieces of mussel fed to them.

DM-2:

The drilling mud tested in Sep 79 (DM-2) gave very different results. Even the lobsters exposed to the higher concentration of the pollutant showed little or no effects. All animals ate normally and the only difference in behavior was that animals exposed to drilling mud were found more often in their shelters.

Discussion

These experiments have demonstrated that some drilling muds and their effluents can be extremely toxic and that others may have negligible effects on lobsters. In addition to the lethal effects of high concentrations of the toxic muds, lower concentration suppressed the feeding behavior of the exposed animals. Although this effect was reversible, alterations of feeding behavior could have longer-lasting implications for sublethally exposed populations. The need for chemical analysis to determine the toxic additives or components of drilling mud is obvious. Our results indicate that caution should be exercised in disposing of drilling muds near marine areas of high biological productivity. There are many questions to be answered before we understand the biological implications

Figure I. 3. Response of lobster to exposure to clean ocean mud (Control). Although the carapace of this lobster was completely covered with mud, and visibility in the aquarium was only 3 cm, this animal remained very active and fed regularly.

of toxic drilling muds; for instance, will mobile animals avoid the mud? In our experiments, effects from toxic drilling mud exposure occurred very rapidly, much too rapidly to permit escape by the lobsters, especially if the mud were to cover an area of ocean floor larger than the lobsters' normal range. In our field work, this range is often within 10-50 meters. Less mobile species and those living in the substrate, including post-larval lobsters, would have virtually no chance of escaping toxic effects.

Obviously, physical questions such as how much the mud disperses when released at the surface must also be addressed. Both in our large tank experiments (Section II) and in the field (Section III), lobsters do much bulldozing and moving of substrate; in the field we have also observed lobsters living in mud shelters. This digging activity could lead to their re-exposure to still toxic drilling mud incorporated in sediment over a much longer period of time than would be expected from natural breakdown rates. Generally, the results we obtained with the toxic mud were far more severe than we had anticipated.

II. LOBSTER BEHAVIOR IN A NATURALISTIC AQUARIUM

Elisa B. Karnofsky, Susan Oleszko-Szuts, Randall Elgin, Linda Ashkenas,
Charles D. Derby, Dana V. Devine, Leonor Botero, Thomas J. Trott.
Shelley Gibbons, Eleanore Beale and Jelle Atema

Laboratory tests of toxicity levels of pollutants are usually made in very limited environments which do not reflect the normal life of an organism. In order to understand the diverse implications of the effects of a pollutant on an organism, one must first have a good understanding of the life of that organism, including not only its biological life cycle but also the general behavior pattern exhibited by each developmental stage. We have studied sub-mature and mature lobster behavior in the laboratory in large naturalistic tanks where detailed observations are possible in a fairly natural environment. These studies are complemented by field studies (Section III).

Methods

Eight lobsters (4 males, 4 females) (see Table II. 1) have been observed for 18 months in a naturalistic environment to establish a seasonal base line for lobster behavior, i.e., activity levels and rhythms, foraging, shelter use and construction, molting behavior, courtship, territoriality, and dominance relationships. The aquarium (Fig. II. 1) was 6.7 m long, 1.5 m wide and 0.9 m deep, and had a substrate with mud, sand and pebble areas. Two blocks of peat were added due to reports that post-larval lobsters have been found in peat. Nine rocks with macroalgae attached were placed in the central areas of the aquarium. They were changed frequently to replenish the algae, which did not survive well in the tank despite the presence of

Grolux with wide spectrum lights placed above them. Eel grass was planted in the mud section. Eight cinder blocks and three molded concrete shelters were provided. The molded shelters had a window on the glass to allow observations within the shelters. A variety of prey items was supplied on an irregular basis. Two potential predators, a tautog and a dog fish, were added but did not prey on the lobsters and were removed after one month. Unfiltered seawater from Woods Hole Harbor at ambient temperatures flowed through the tanks at 40 l/min.

Behavioral observations were made three times daily for one half hour, in the morning, at nightfall and during the night. Observations on interactions between lobsters, and between lobsters and other species were recorded, as well as shelter-related activity and general movement in the tank. During periods of molting and mating more frequent observations were made (as much as 24 hr per day). Every 2-4 months 24-hr recordings were made to determine if the existing recording schedule was representative of activity in the tank. Behavior was broken down into units for easy notation. These units form the basis of data analysis (see Section IV for list of units).

Results

One aspect of this study, courtship and mating behavior, has been analyzed and published (Atema et al., Mar. Behav. Physiol., in press). Lobsters were shown to exhibit female mate selection and pair formation. (See Appendix II for abstract).

Other aspects of the data were analyzed by hand to formulate questions answerable in detail with the aid of a computer: a) A representative 18-day

Table II. 1. Description of lobsters in naturalistic tank (1978)

Sex	Size (mm)	Comments
♀ RR	77-85	molt, 6 July, mated w. LL(1), eggs 18 Aug
♂ LL(1)	76	removed for other experiment
(2)	75-90	molt, 8 Oct
♀ PS	60-66	molt, 22 Aug, mated w. LL(1)
♂ L	57-67-80	molt, 12 July; 21 Oct
♂ R	55-65-71	molt, 4 July; 12 Oct, lost claw to RL
♀ RL(1)	48	removed for other experiment
(2)	50-61	molt, 29 Oct
♂ P	46-52-58	molt, 27 June, lost claw to R; 25 Sep
♀ S	36-42-48	molt, 30 July; 10 Nov

Figure II.1. Diagram of naturalistic aquarium. Lined area at left represents the mud substrate area, which also includes pots of eelgrass; the dotted area is the sand substrate. The remainder of the tank has a pebble substrate. The rock garden is in the center right. Peat blocks are located at the back of the mud and sand sections. Cinder blocks are along the back of the tank; large molded concrete shelters are along the front windows.

period was analyzed for aggressive interactions and a 9-day period for lobster range and home territory. Each lobster is individually described with respect to these data. b) Residency in preferred shelter D is then described for a one-year period to illustrate changes in dominance over an extended period. An anecdotal description of c) molting and d) feeding behavior is also included.

a) Analysis of representative time period

During 01-18 Oct 78 the large dominant male (LL) molted on 08 Oct and the second smallest male (R) molted on 12 Oct. The resident of the preferred shelter (D) changed three times - from the dominant male (LL) to the subdominant male (L) to the largest female (RR) carrying eggs.

Figure II. 2 illustrates the aggressive interactions over this period. In general, there were fewer aggressive interactions during the day than at night. The completely nocturnal activity pattern seen in the field is less complete under laboratory conditions. Small individuals such as female S seemed to take advantage of the relative nonactivity of larger animals to do their foraging during the day (Fig. II. 3a). As a result, their chances of interacting with others are greatly diminished. Females appeared to have fewer aggressive interactions than males. This may be an intrinsic sex difference as described by Scrivener (1971, Fish. Res. Bd. Can. Tech. Rep. 235), or the two male molts during this period may have abnormally increased the number of male interactions. Analysis over longer periods with computer will clarify this relationship. Premolt lobsters typically shows an increase in successful aggressive interactions (see male LC who molted on 08 Oct and male L who molted on 21 Oct; Fig. II. 2). The females in the tank became more active following the male molts.

Figure II. 2. Lobster aggression in naturalistic aquarium during 01-18 Oct 78.

Interactions which each lobster won are shown above each abscissa; those lost are below. Intense interactions are shown in black and represent charges, attacks and/or scissors for wins; tail flips for losses. The cross-hatched regions are for mild interactions: approach, follow for wins; retreat, flee for losses. A. Daytime interactions. B. Nighttime interactions.

Figure II. 3. Ranges of lobster activity in naturalistic aquarium during 10-18 Oct 78. Each rectangle represents a schematic floor area of the tank, without the mud area (see Fig. II. 1) which was opened on 31 Dec 78. A grid was formed over the floor: there were three divisions front to back; the divisions from left to right correspond to the area in front of each cinder block and standpipe. Shaded areas indicate grid squares entered at least once by each lobster during each day (AM) and each nightfall and night (PM) recordings. In some cases (e.g. Fig. II. 3d, PM of 12 Oct) there are noncontiguous shaded areas. These represent different recordings made during one evening. The "X" in the shaded area is the area considered "home" shelter by each animal.

FIGURE II.2.b.

10 OCT

11 OCT

12 OCT

13 OCT

14 OCT

15 OCT

16 OCT

17 OCT

18 OCT

♀ S (42 min)

A

10 OCT

11 OCT

12 OCT

13 OCT

14 OCT

15 OCT

16 OCT

17 OCT

18 OCT

♀ RL. (50 mm)

B

♀ PS (66 mm)

C

AM

SP 1 2 3 SP 4 5 6 SP 7 8 9 SP

PM

SP 1 2 3 SP 4 5 6 SP 7 8 9 SP

10 OCT

♀ RR (85 min)

11 OCT

12 OCT

13 OCT

14 OCT

15 OCT

16 OCT

17 OCT

18 OCT

D

AM

PM

SP (58 mm)

E

10 OCT

11 OCT

12 OCT

13 OCT

14 OCT

15 OCT

16 OCT

17 OCT

18 OCT

♂ R (65 mm)

F

10 OCT

11 OCT

12 OCT

13 OCT

14 OCT

15 OCT

16 OCT

17 OCT

18 OCT

♂ L (67 mm)

G

♂ 1.1. (75 mm)

H

Maps (Fig. II. 3) show a daily range and home shelter for each animal over a nine-day period (10-18 Oct). The extent and stability of range varied greatly for each individual. Small female S had a limited range and fixed shelter during the entire time (Fig. II. 3a) whereas small male P had neither fixed range nor shelter (Fig. II. 3e). In the field, given unlimited space, an animal such as P would probably leave the area. Initially female PS did not have a fixed shelter, but she established one on 14 Oct, after the molt of male R (Fig. II. 3c). Female RR who was carrying eggs was resident in smaller molded concrete shelter B until she moved to preferred shelter D vacated by newly molted male LL (Fig. II. 3d). Newly molted animals, such as males R and LL, are not able to maintain a residence for several days or even weeks until their exoskeletons start to harden (Fig. II. 3f and h). Male R, who was the dominant male during this period, had the largest range of all the lobsters.

Results from these sets of data are summarized in a description of each lobster.

S ♀ was the smallest lobster in the tank. She had almost no interactions, probably because she only moved around the tank occasionally. She had a fixed shelter under one of the cinder blocks, too small to fit a large lobster, and she rarely foraged any great distance from it. She covered a larger range during the daytime, perhaps to avoid other lobsters active at night.

RL ♀ also had few interactions. Most occurred after the male R molted. This increase in encounters may have been due to the unusual presence of the newly molted male within her range. She had a fixed shelter and fixed range.

PS ♀ did not win many interactions until male R molted, after which her percentage of winning encounters increased (Fig. II. 2). Her shelter changed frequently at the beginning of this period and then stabilized again after the molt of male R; she had an extended range in the center of the tank.

RR ♀ was the largest female and carried eggs. She had relatively few interactions compared to males but won a large proportion of them. She usually had a fixed shelter but during this period she moved from B to choice shelter D. After moving to D she seemed to do much patrolling of the area. This might be necessary to establish and maintain residence in the preferred shelter in the tank.

P ♂ was the smallest male. He almost never won interactions and had a variable range, variable shelters, or no shelter at all. It has been suggested that lobsters without fixed shelters in the field would leave the area.

R ♂ molted during this period, but did not show the customary premolt aggression peak. He may have been too small in relation to the other lobsters in the tank to display aggression. He had a stable shelter before molting but moved to the opposite side of the tank following his molt. As his exoskeleton hardened his range slowly extended toward his previous residence.

L ♂ showed a large aggression peak which may have been due either to his premolt condition (he molted 21 Oct), or to the molt of the other two males (R and LL), or to a combination of the two. His range shifted

from the area around D to the area around B when female RR took over D. He established a stable residence in B.

LL ♂ was the largest male in the tank. He showed a very clear premolt aggression peak as well as a loss of dominance following his molt; surprisingly, he still won a large percentage of his encounters, probably due to his size and aggressivity. Before molting he had a stable residence in D. After his molt he was evicted from D and moved to the center of the tank. As he hardened his range extended back to the area around D. This extension of range also correlates with female RR's takeover of D and male L's eviction from the area.

Further analysis of the data using these techniques, but with the aid of the computer, will help confirm our observations of the effects of molting on aggression, loss of dominance and change in home range. It may also bring out complex relationships between behavior patterns of different lobsters and their causes. The reality of this behavioral complexity is a necessary baseline to describe possible effects of pollutants.

b) Residence in Shelter D

Changes in residents of shelter D (the preferred shelter in the tank for larger animals) demonstrate changing dominance and the effects of molting and egg-carrying. Shelter D is the largest shelter and is located on a pebble substrate with a front and back entrance. It has an opening on the glass to allow observations within the shelter.

The largest dominant male (LL) inhabited D from the time he entered the tank in Jul 78. All matings took place in D with LL. Smaller animals often took refuge on top of D, particularly following a molt. Following LL's molt,

08 Oct 78, he was evicted from D and second largest male L moved in. For two days L barricaded the doors. On 10 Oct the largest female (RR), who carried eggs, then approached D. Although she did not enter, L no longer stayed in D but constantly checked nearby shelters and stood near or on top of D. On 12 Oct RR moved into D and L took over her old residence, B. On 21 Nov, LL, now hardened after his molt and having shown superiority over the other animals in the tank, evicted RR and moved into D. Interactions between RR and LL during the preceeding days were out of the ordinary and more frequent than usual. They would meet in the center of the tank and stand still facing each other with claws closed. Occasionally there would be pushing. No violent interactions were observed between the two, i.e., lunge, snap, or attack. However, both LL and RR were seen to attack L (the second largest male) who retreated readily. RR took up residence in B but the next day continued to check D. There were still no violent interactions between RR and LL.

On 02 Sep 79, LL molted again. This time he showed a violent aggression period of 7-10 days before molting. After molting he was attacked and finally cannibalized by males R and L. R took over D. L was much larger than R, but L had recently molted. On 18 Sep, L dug R out of D and established residence there himself.

From this simplified outline of residence in shelter D, one can see that a molted animal loses dominance but can regain it when hardened. It also appears that females with eggs relate differently to males than to females without eggs or other males. With computer aid we will quantify our data to efficiently determine the complex and changing relationships between lobsters.

c) Molting Behavior

Molting behavior has been well described in the laboratory. A large percentage of molts occurred during the day. In the field in particular, this would give the lobster several hours to gain strength before being evicted by other lobsters during their nocturnal foraging and shelter checking activity. The cinder blocks appeared to be too small for molting. Although some lobsters barricaded themselves into cinder blocks, molting took place in the large molded shelters, under the peat, or in craters dug out in the substrate.

Molts could often be predicted from shelter behavior (barricading, bulldozing) and premolt aggression peak. Lobsters, both males and small females, build elaborate barricades in front of a shelter or dig large craters in the substrate. Construction of barricades is not haphazard: sponges, scallops and small rocks were collected by lobsters from other areas of the tank and carefully placed to form barricades. The so-called premolt aggression peak has been documented experimentally for post-larval lobsters by Tamm and Cobb (1978, *Science* 200:79-81), and is often observed in our naturalistic tanks for sub-adult and adult animals. In these larger animals it can last as long as two weeks.

In spite of the extreme vulnerability of lobsters during molting, we have never seen a lobster actually being disturbed during the molting process itself. The process, from the time the lobster falls on its side until it tailflips out of the abdomen, takes about 10 min for this size class. In one case a lobster molted in a large crater it had dug in the middle of the rock garden. A slightly smaller lobster resided under a rock within one foot of this crater. The premolt lobster was seen lunging at his neighbor up to 5 min premolt. The

neighbor did not disturb him during his molt or for four hours following the molt. At sunset, however, he was immediately chased away. Thus, decreased activity during daylight hours is probably an important factor in post-molt protection. There may also be an association between the smell of a premolt-molting lobster and his aggression peak.

d) Feeding behavior

Lobsters are not simple scavengers as they have been reputed to be, but attack and eat a wide variety of live prey. They readily eat relatively sedentary prey such as mussels, scallops, snails and echinoderms. In the large aquaria they have been seen mulling mussels out of mussel clumps, crushing and eating them. The larger lobsters were able to crush large bay scallops, gaining access by crushing them specifically at the adductor muscle. Snails were cracked in the crusher with the seizer held over the snail to prevent it from shooting out of the crusher. Sea urchins and starfish were also eaten but did not seem to be a preferred food. Sea urchins are simply crushed. Lobsters crush only one leg of a starfish without killing the animal because the starfish appears to autotomize the crushed leg, leaving it to the lobster predator.

Faster moving prey are also caught, but here lobsters do not appear to be efficient predators. Small fish were only rarely seized. This usually occurred when the lobster was eating something else and the fish would duck in to steal scraps. The fish were snapped at with the seizer claw - perhaps as a scare tactic - and apparently accidentally were caught. Crabs were often eaten, but the chase, especially with green crabs (Carcinus maenas) and lady crabs (Ovalipes ocellatus), was often unsuccessful. Successful predation usually occurred when

the lobster dug the green crab or lady crab out from under a rock or out of the sand. Rock crabs (Cancer irroratus, C. borealis), lady crabs, hermit crabs (Pagurus longicarpus, P. polycaris), and spider crabs (Libinia emarginata) were often eaten. Lobster feeding appears to be chemically rather than visually guided. In pursuing a crab they follow the chemical trail whether or not the crab is in sight. Crabs may escape after losing only one or two legs.

Lobsters also spend long periods of time "dactyl feeding", i.e. probing the substrate with their walking legs, which are covered at the tips with sensory hairs for both mechano- and chemoreception. From time to time they will stop and take a clump of mud or a pebble into their maxillipeds and manipulate it with their mouthparts. They were also seen drawing strands of eel grass or pieces of macroalgae, i.e. Fucus, through their mouthparts. It is possible that they are gathering small invertebrates such as small snails and amphipods, or actually eating bits of plant material.

Detailed and quantitative analysis of the data bulk will be used to answer such questions as whether lobsters select certain food over others or whether they are strictly opportunistic feeders, and whether selection is related to season, molt stage, previous experience, etc.

Discussion

The naturalistic tank observations have been essential in understanding the relative importance of different parameters in the life of a lobster. The lobsters can exhibit potentially complex relationships with their environment, which are not possible in small tank studies, while permitting lengthy, detailed observations of behavior which are not possible in field work. Hand analysis of

a short time period has helped us to organize the data and formulate questions for computer analysis now in progress.

Appendix II

Marine Behaviour and Physiology, 1979, Vol. 1, pp. 000-000
0000-0000/79/0604-0000\$04.50/0
© 1979 Gordon and Breach Science Publishers Inc.
Printed in Great Britain

Pair Formation in the Lobster, *Homarus americanus*: Behavioral Development, Pheromones and Mating

JELLE ATEMA, STEWART JACOBSON, ELISA KARNOFSKY,
SUSAN OLESZKO-SZUTS and LAUREN STEIN

*Boston University Marine Program, Marine Biological Laboratory,
Woods Hole, Massachusetts 02543*

(Received March 10, 1978)

Female-initiated pair formation and mating were observed during long term observations of lobsters (*Homarus americanus*) in seminatural environments. Females left their solitary shelters and selected the company of dominant, territorial males as much as 7 days before molting. During subsequent pair formation and shelter sharing a number of new behavior patterns were observed, some previously not reported for other Crustacea. Both behavioral displays and a female sex pheromone appear to be involved in pair formation. Mating took place one half hour after the female molted. After mating, pairs remained together in the male's shelter for up to an additional 7 days. During this time males fed on the molt shell and actively defended the area. Females of 58-70 mm carapace length showed varying levels of incomplete pair formation and mating behavior, indicating that courtship and mating behavior may require a number of molts to develop. This size class forms a behaviorally defined transition stage. Females larger than 75 mm were behaviorally mature.

Pair formation in lobsters appears to protect mature, post-molt females against predation and cannibalism and to insure the reproductive success of dominant males. These observations provide a more natural behavioral context for the function of a sex pheromone in *Homarus americanus*.

Acknowledgements

We thank Camas Lott and Gail Burd for help with the 1974 observations, Randall Elgin for help with the 1978 observations, and Sherman Goodell and Peter Sachs for obtaining lobsters for us. This research was supported by Energy Research and Development Administration grant #E(11-1) 2546, and by grants from the U.S. Department of Energy (EY.76.S.02.2546) and the U.S. Environmental Protection Agency (R-803833).

III. FIELD STUDIES

Elisa B. Karnofsky, Jelle Atema and Randall Elgin

Observations of lobsters in the laboratory, even under "naturalistic" conditions, do not necessarily reflect the life of lobsters in the ocean. In an aquarium they are confined as a fixed, small group of individuals; water turnover is negligible compared to the tidal movement and currents in the field; large predators are absent; and there are not the natural seasonal changes in abundance of prey species. It is important to observe behavior, distribution in the field, and the reality of ocean bottom existence in order to evaluate the more detailed observations possible under laboratory conditions.

Methods

From Apr-Oct 79 field observations of lobsters have been made. The field site is a small cove 140 m across, 13 km from Woods Hole on one of the Elizabeth Islands. The water depth in the observation area varies from 0.3-2.4 m at low tide. The substrate is eelgrass, sand or rock. The fauna consists of several species of fish, crabs, and molluscs.

Rocks with painted numbers identified 116 lobster burrows. The burrows were located on a map by triangulation using stadimeter readings and distinct distance measurements (Fig. III. 1). Daytime observations were made to monitor the state of the burrows for lobster activity and changes in shape and possible occupancy. A 4-m square plot was delineated by a chain for photographic documentation of monthly changes of the macroflora. An observation raft was constructed for long term observation of a small area. It was placed near the mating burrow (see below).

The lobsters in the beginning of the season were trapped in standard lobster traps and banded using large rubber bands placed behind the dactyl hinge of the large chelae - on the left for females and on the right for males. Later 48 individuals were identified by numbers written in magic marker on the rubber bands. The lobsters were sexed and carapace length (CL) was measured; handedness of crusher as well as distinguishing characteristics were noted at the time of banding.

Nighttime observations on lobster behavior were made by snorkeling 1-2 hr, usually beginning at sunset, with low intensity lights. Location of lobsters and behavior, including interactions with other lobsters, shelters and prey items, were noted.

Results

Over the 1979 observation period, 117 burrows were labeled and 52 lobsters were individually marked. Observations have been made of social behavior, burrow-related behavior and foraging behavior. After a qualitative summary of these three aspects of their behavior, a more detailed account of individuals and shelters follows.

Besides the field confirmation (Karnofsky et al. 1979, Biol. Bull. in press) of the recently published courtship and mating sequence from the naturalistic study (Atema et al. 1979, Mar. Behav. Physiol., in press), aggressive social interactions were observed. These showed that small lobsters generally avoid encounters with approaching larger ones. Long, intense interactions with snapping and facing-off were seen between lobsters of similar size. However, in other cases lobsters were seen to pass within 15 cm of each other without observable interaction. Occasionally, seemingly purposeful attacks over long

distances were observed, i.e., eviction by a mating male of another male lobster from its burrow 5 m away.

Lobsters appear to be constantly modifying their burrows. Sand is bulldozed out of the interior as storm waves and tidal currents disturb the substrate and the shelters. Rocks and other objects are brought to the burrow openings, possibly as defense. Some lobsters are shelter bound while others are very active, often checking various shelters along their nightly course.

Surprisingly little information has been collected on foraging behavior. It may be that foraging is more easily disturbed by our use of lights. Lobsters have, however, been observed scavenging, e.g., chewing on molt shells from various crustacea, and dead animals lying on the substrate, such as bait fish killed during a bluefish feeding frenzy. They have also been seen manipulating hermit crabs and attempting to catch fish. While walking over the substrate they may be constantly feeling ("dactyl feel") for small invertebrates. Lobsters are also often seen combing through the macroalgae on rocks.

To indicate the type of information obtained from this study, the sequence of major events observed at four burrows located on the photomosaic (Fig. III. 2) is summarized below. Following this, a summary of four regularly observed lobsters is given. Details on all shelters and events can be found in Appendix III.

Shelter #29 was a mating burrow. It consisted of a large space under a rock with a main entrance facing SSW and a side entrance facing SE. The banded male resident of about 80-mm CL cohabited in sequence with two females, one unbanded and one banded. Both females molted and - based on our knowledge from the naturalistic study (Part II) - probably mated. Other females visited the burrow, standing under a small rock nearby (WR), occasionally approaching the entrances of #29, then leaving. Males, large and small, were also seen approaching the entrances and leaving quickly. The resident male was seen leaving the mating burrow, running 4 m North to shelter #110S, entering the burrow, and evicting a

Figure III. 2. Diagram of plot drawn from photomosaic. Lobster burrows are labelled. See text for details.

male of about 70-mm CL. A small lobster "satellite" (about 30-mm CL) lived under a rock directly behind the mating burrow. A yearling lobster was observed later in the season under one side of the doorway (see Appendix III for details). Male-female cohabitation, general interest in the mating burrow, male evictions by the mating male and small satellite lobsters have all been described, but with far more detail, in the naturalistic tanks. This indicates the great degree of realism that exists in the aquarium situation.

Shelter #81 housed a lobster of about 50-60 mm CL for 5-6 weeks. He was only occasionally observed outside his burrow. His leaving became more frequent in September and may have reflected his pre-molt activity and aggression peak. In mid-September a new lobster was seen in the shelter. The original occupant may have molted and been displaced. (This is the normal course of events in the naturalistic tanks.) The opening was very narrow and what appeared to be a back door early in the season was closed in later.

Shelter #110S was a large burrow with various lobsters seen in it. It was under a large flat rock with the front door facing E and the back door facing W. Molt shells were observed in front on several occasions. Both females and males were seen in the opening. In the beginning of the season, it may have been a mating burrow. While #29 was an active mating burrow, #110S was the burrow from which other males were evicted. In early September it was the residence of an egg-carrying female who also spent some time in #29.

Shelter #110N was opened up on 27 Jul when a 57-mm CL female was seen within. She was the sole occupant through 15 Sep. She was occasionally seen foraging in the vicinity.

Lobster II (female LC, 63 mm, cut seizer tips) was observed beginning 17 Jul when she showed great interest in the mating burrow (#29). She

approached the burrow and stood under a rock (WR) nearby for an hour after sunset. She visited daily until 31 Jul when the male disappeared. She then entered the burrow on short visits for several evenings and finally was no longer observed in that area. Her extensive activity range is shown on the map (Fig. III. 1). She was observed on a regular basis from 17 Jul through 15 Sep.

Lobster III (female RC, 61 mm, no crusher) was a one-clawed lobster with a large deformed crusher limb bud and a right antenna bud. She was seen from 31 Jul, when she was banded, until 17 Aug. She probably molted at that time because 12 days later, on 29 Aug, lobster XL (female RC, 68 mm, regenerate crusher) appeared in the area with a regenerate twisted crusher and a short right antenna. She was observed in the Big Rock area until 03 Sep when she was seen 12 m further South than she had ever been described, and she has not been seen since. Next year telson clips will accompany banding in order to be able to identify lobsters following the molt (see Appendix III for details).

Lobster XII (female LC, 53 mm, no right antenna) is an example of an animal which is very shelter bound - since 01 Aug she has been in #38 or foraging nearby. On only two occasions was she seen in nearby shelters.

Lobster XVI (male RC, 60 mm, barnacles on seizer) is a male who has been seen interacting with female III on several occasions. On 08 Aug he moved into shelter #72 where female III had been seen previously and was able to defend it against her when she tried to re-enter. He is always observed in the Big Rock area (see Appendix III for details).

Discussion

We have shown that valuable information on lobster behavior can be collected in the field. In the shallow cove we studied there is a stable population of lobsters. Certain individuals have been followed nearly daily over a 3-month period. It was encouraging to see that our method of banding and observing has not discouraged individuals from staying in the area. Some lobsters were only seen once. This may be due to fishing pressure or territoriality of residents. Large (60-80 mm CL) and small (20-40 mm CL) lobsters live in close proximity to each other with only low key (approach-avoid) interactions. The large incidence of damage to lobsters (missing claws, walking legs or antennae) may be related to molting, aggression, predation or trapping. Some wounds appear clearly the result of lobster aggression.

Despite the use of lights during night observations, useful information has been collected on burrow-related behavior, social interactions, including courtship and mating, and foraging behavior. The short disjointed observations characteristic of the field were understandable because of correlation with the detailed descriptions made in the naturalistic tanks (Section II.). We are confident that only this approach (i.e., laboratory studies combined with field observations) can lead to effective evaluation of pollution effects in the lobster's real world.

II

labelled 7/31. female LC 63 mm cut seizer tips (CST)
caught out of small rock burrow N of plot

7/17 ? tried to band $\frac{1}{2}$ on
7/18 fixed band (did not note CST)
7/20 walk direct to #63 turn, leave, not disturbed by light
7/23 enter #75; check burrow next door to #75 enter #75; not
there later
7/24 ? in #67
7/24 at NE corner of plot walking about
7/26 foraging near big rock
7/27 under waiting rock - several passes at #29 leave toward #81
7/28 under waiting rock; made several passes at #29 moved off toward #81
9/29 under waiting rock
9/30 approach side door #29 enter as male (CST) exits from front door
walk North female back into front door male returns enter
front door female exit new back door approach small female who
avoids II walk SW
7/31 banded with number
8/1 under waiting rock
8/2 entered #29 from waiting rock no male inside stayed in #29
8/4 approach #72 III inside back door try to duck in probably
because of lights then come out III follow but does not leave
burrow II walk around rock and duck under ledge where she stayed
facing #67 ? near #96
8/5 in #65; in #29
8/7 walk South of big rock enter between 2 flat rocks near #43
8/8 fight with male LC about same size in #98 both meral spread,
scissor II finally walk away male also female enter flat
rocks near #43
8/9 hang out flat rock crevass
8/10 ♀ LC CST
8/12 near #67
8/16 at back of #81 walk to front then East then North enter exit
#63 enter #65

8/17 walk by #81
 8/20 walk across front of #81 to West then South enter #61 later
 at back of #81 (VII approach from North check)
 8/21 walk in plot toward NE
 8/22 walk around rocks SW anchor line West of #61 then proceed
 to #81 try to enter did not; went North
 8/27 near #61 enter #61 passed cm from XXXIII no interactions
 8/27 enter #63 stay long time eating piece of jelly fish or
 weird sea weed
 8/28 walk toward East near #63
 8/29 in #67 leave walk to large rock West of Big Rock
 in #98 leave walk to same large rock West of Big Rock
 8/30 in area of #61
 8/31 near #60
 enter #65
 enter #63
 in burrow between #58 and Big Rock
 9/3 run into back #72 then appear at front
 9/4 in #65
 9/5 in #75
 9/12 shy under rock other side of #66
 9/13 dig at space between 2 rocks outside chain behind #81
 light doesn't seem to affect later seen under back of #81
 then walk around check #81 back in turn abruptly
XIX in #81 seizer wide open II stand off then turn walk
 Northeast short time later seen again at #81
 front door light → walk to back under rock
 9/15 near #58 hide in crack

[III] banded 7/31 female RC 61 mm no crusher
 medium large crusher limb bud getting dark - deformed
 also coiled right antenna bud

8/2 in #75
 8/3 in #72
 8/4 in #72
 8/5 in #75
 8/8 under rock 6' south of #72
 8/9 approach back door of #72 ♂ [XVI] in #72 at front door [III]
 enter back door [XVI] turn [III] exit walk to front door
 approach jump snap male scissor face off [III] leave
 walk to #63 back door
 later male RC [XVI] approach her at back of #63 #65 rock
 male lunge - scissor [III] retreat; male return to #72
 [III] later seen hanging out of crevass in flat rocks where [II] female
 usually is
 [III] later tail seen hanging out of #110S 2 jerks then back out
 move to north then west then I left no lobster came
 forward in #110S

8/10 seen on rock scared by light climb off rock - run toward
 female RC [XVII] swinging claw - she might have been pushing
 algae out of the way or scraping her claw [XVII] moved away north

8/11 in #65
 8/12 in #101
 8/6 under rock north east of #80 bulldoze
 8/17 under same rock bulldoze

XII banded 8/1 female LC 53 mm no right antenna

8/2 in #38 later on fan of #100 facing door backed into #38
with light

8/3 under rock about 10' south of #100

8/4 in #38

8/5 under rock near #80

8/7 in #101

8/9 in #100

8/11 in #38

8/12 in #38

8/17 in #38

8/20 in #38

8/30 in #38

8/31 in #38

9/3 walk north of #38

9/4 hang out #38

9/8 in #38

9/13 back into #38

XVI banded 8/1 male RC 60 mm barnacles on seizer
 8/1 caught and banded near #65
 8/5 in #66
 8/8 possibly in #72
 8/9 female III approach XVI in #72 XVI turn to check back door
 where she attempted to enter, then front door when she
 approached scissor, snap, Face off XVI may have scissored or
 snapped ♀ leave. XVI later approach scissor ♀ III under
 large rock of #63 and #65 ♀ retreat XVI walk around rock,
 return to #72
 8/10 male possibly same in #63
 8/11 approach rock south of #75 then back into #75 with light
 (II female later seen under that rock)
 8/12 under other side of #66
 displaced X male from #63
 8/17 leave #60 enter rock facing #60
 8/26 under rock near #65
 8/31 in #72 confronted by XL female push, jab, fight XL leaves
 9/3 in #72
 9/5 in #78 area walk enter rear of #78
 9/7 enter #95 leave, enter #97
 9/12 in #78
 9/13 run near #78 carrying something in maxillipeds (I can't see what)
 enter #95
 9/15 walk west of big rock into #78N leave walk to #95 enter
 walk near about 40 mm lobster both no response

XL banded 8/29 female RC $\frac{1}{2}$ C 68 mm crusher has large dent in center
of topside - also it's twisted and deformed caught
south west of #67 right antenna slightly short

8/29 in #60

8/30 approach XVI male in burrow continue walk across #66
enter #60

8/31 approach XVI male in #72 after leaving burrow near #72 tries
to enter push jab fight leaves walk to #78 enter #66

9/2 in #72 bulldoze

9/3 in #75 later walking south west of #61

Appendix III. B. Field records of lobster shelters

#29

- 6/10 2 doors; big cunner under rock
- 6/17 molt claw just inside corner of plot.
- 6/18 molt claw - lobster antenna sticking out door - caved in area at north side
- 6/28 still has big fans slightly caved in part under back of rock
- 7/3 (SSW) giant, 2 holes deep fans
- 7/10 (SSW) active
- 7/12 nearby about 50 mm RC lobster and about 30 mm lobster
- 7/17 am banded RC male and RC female? no band missing crusher
pm no molt claw seen in area, small (20-30 mm) at rear of #29, male in entrance, broad femalish tail of the other lobster, male boxed her claws, pushing her against back wall
- 7/19 RC male and 2 clawed, somewhat smaller lobster; no molt shells seen nearby or inside; both animals fanning, male at entrance
- 7/20 raft observations of activity in burrow: male fan sway walk turn male lunge female turn retreat, present rear; male box jab, female push stand retreat; male came to entrance female stands mostly in the back
- 7/21 rear of #29 a (20-30 mm) lobster in small burrow
- 7/22 from raft, activity in burrow: male approach, run; female stand push. Male retreat. Male stand away. Female claws seen out of SE entrance
- 7/23 female looked, stretched, regenerate crusher
(male LC, large 1.5 cm red limb bud crusher, cut thumb seizer, digging at rear through tiny lobster's home, leave with light)
male at front, female occasionally disappears at rear, she often faces back
- 7/24 male as usual between entrances, female on W side, female has regenerate small RC (first regenerate size after loss), battered and punctured left molt seizer found in SW entrance. No other molt remains seen; only male seen.
male and new female RC banded inside both at 20:15 and 21:30.
New female slightly smaller than male.

- 7/25 only male seen from free dives, came always forward; once I thought I saw other molt arm remains, but next time not seen. No evidence of other lobsters. SW entrance has several small rocks in it (barricade?)
from raft: only male seen, but he could disappear out of sight; female could be there too.
two lobsters walked in area: RC(50) from #29 to plot area, walks with meral spread and a (30) walks along to plot
- 7/26 male and large unbanded RC (female?), no evidence of molt shell. Tooth distinction on seizer propus, she was at entrance several times. She is perhaps slightly smaller than male. She looks very clean with orange edges of claws, does not act post-molt. RC(30 MM) behind #29, entered main shelter from rear.
male plus female (?) no band on RC, LS not seen, no molt shell parts inside, or in area.
male plus female RC unbanded, tooth on seizer, no molt shell parts inside, or in area.
- 7/27 am molt crusher (RC) found 6 ft S of #29; male plus RC female inside (deep in - face forward); male on tail piece of molt shell (eating?)
pm male RC banded female RC against W wall facing center; female comes to door; about 30 RC lobster under rock at back
female LC CST (II) walk under raft see attached diagram for female approaches
- 7/28 am female usual place. male in front, comes to door; cunner and sculpin inside; no molt remains
pm about 30 mm lobster behind burrow. See diagrams for female CST (II) movement
- 7/29 am can only see male; also did not come forward as usual
pm female RC came out, grab bait head
unbanded about 70 mm approach side door, turn, flee, meral spread #70 near rock to East saw female LC recap 50-60 mm foraging; later she approached #29 side door, turn, walk back to East rock, stand facing #29; male CST walk around back to front door then walk away East about 30 mm lobster under waiting rock (WR), then II female CST under Waiting Rock (WR); RC about 50 mm lobster walk by #29 headed for #81

- 7/29 pm male hovers at door then leaves; heads quickly North, approach
(cont) back door #110S. Enter and large male RC (no seizer, about
70 mm) leave by front door (no band). I did not see male return
to #29.
Female CST (II) back under Waiting Rock, walk South under raft
- 7/30 am Male RC came to door
pm male RC banded NEW MALE (CST) hang out side door, back in with
light. Leave side door, walk around back, back in side door, exit
front door at same time large lobster approach, half enter side
door, back out, turn to back in. Male CST back in front door.
Large lobster flee walk SE. I caught. It's a male, crusher
has newly broken dactyl, still white wound; seizer has tooth in
middle on under side of dactyl - egg shell very soft. New back door
at #29, no about 30 mm lobster under back rock.
Female LC CST (II) approach side door, enter as male (CST) leaves
front door, walks to rocks N of plot, approach back door #110S
then walk to front doot, back and forth 3 times then stand on
chain facing #29
Female LC (IV) walk in back of #29 to Waiting Rock
Male walks from North past female (IV) who approaches male and
enters front door #29
Female LC CST (II) exit from back door, approach female (IV) avoid
female (II), approach Waiting Rock, then walk to southwest. Female
(IV) returns to Waiting Rock, stand in front for rest of recording
Male (CST) in and out front door, sometimes rearing on algae beside
door
- 7/31 IV female LC 52 mm behind shelter #29 in Waiting Rock
- 8/1 Nearby male RC 40 mm not banded
Waiting Rock II - in #29 RC (about 50 mm)
- 8/2 Active 2 front doors, 1 back door, probably not inhabited
(II) enter #29 from Waiting Rock, male not inside, stayed inside)
about 30 mm lobster inside front door; later male RC (CST); later
II from Waiting Rock
- 8/3 IV female inside
- 8/5 II female CST inside

- 8/7 XXIII male LC ? about 60-70 mm at front door
- 8/8 ϕ later male RC about 60 mm
- 8/9 new bulldozing closing front door XXIII inside
- 8/10 RC male about 70 mm XXIII ?
- 8/11 XXIII in side door doors are more closed than before
front door almost completely shut
- 8/12 XXIII at side door
- 8/16 XXIII very shy at side door bulldoze
about 20-30 mm at front door right under rock
- 8/17 XXIII inside
- 8/20 XXIII enter front door from around back
VII enter side door #29, exit SW door, short period stay
- 8/21 Active
XXIII inside
- 8/22 beginning ϕ end male RC about 60 mm (XXIII ?)
- 8/26 ϕ later banded male about 60 mm inside all same time
about 25 mm lobster enter
- 8/27 ϕ
- 8/28 ϕ
- 8/29 XXIII inside
- 8/30 LC about 60-70 mm went inside;
later RC female XLIII inside XXXV in rear tiny burrow of
#29; later #29 ϕ
- 8/31 XLIII egger inside; X male approach, short confrontation,
X leaves 5 min later ϕ
- 9/2 XXXV in rear tiny burrow climbing on #29
- 9/3 side door more closed than last night
XXXV behind it later female about 80 RC inside
bulldoze side door XLIII
- 9/4 XLVII approach meral spread enter #29 (LC female 70 mm)
- 9/5 ϕ
- 9/7 ϕ ; later female 70-80 mm RC XLIII in #29 side door
leave run directly to #110S enter back door not seen again
tail curled

- 9/8 female RC egger large hang out side door XLIII
XXXV? male about 40 mm behind rock duck under back rock;
later ø
- 9/12 Active side door more closed than usual
later XLVII approach side door large female RC inside
XLIII? cannot see # both have open claws XLVII retreat
- 9/13 RC female inside seems small for egger but cannot tell
XXXV under rock behind #29; later ø
- 9/15 XLIII inside tiny white tipped juvenile (about 15 mm ?) in
separate hole under #29 East wall (exactly as in shelter B of
Tank II in lab)

#81

7/2 front & back door pictures
 7/19 RC (about 80 ? mm) inside nearby male RC 76 mm has CST
 7/21 Active; $\emptyset$
 7/23 about 50 mm LC inside
 7/24 RC (about 70 mm) inside
 RC (about 50 mm) inside
 7/26 RC about 60 mm inside
 7/28 am $\emptyset$
 pm RC about 60 mm in door
 7/29 RC about 60 mm inside
 female CST walk toward #81 RC about 50 mm approach #81
 fan female banded walk around back of #81 RC about
 50 mm head back toward #29
 8/1 RC (about 70-80 mm) inside
 8/2 Active; RC about 80 mm inside
 8/3 RC about 60 mm inside
 8/4 RC about 50 mm inside
 8/7 RC about 60 mm inside
 8/8 Active fuzzy algae in door RC about 60 mm inside
 8/9 Photograph
 8/16 male RC regenerate seizer at front door another lobster
 inside also RC
 male RC $\frac{1}{2}$ S left with light II female CST at back about
 30-40 mm within 18" of her
II walk to front then E then N enter exit #63 #65
 8/17 RC 50-60 mm in door
 8/20 II walk across front to rock to West then South enter #61
 later II at back CII approach from North check #81 RC about
 50-60 mm inside
 8/22 RC about 60 mm II try to enter turn went North
 8/27 RC about 60 mm XXXII ran by
 8/28 $\emptyset$
 8/29 $\emptyset$
 8/30 RC about 60 mm inside

- 8/31 RC about 60 mm inside
- 9/2 female XLV inside later "Cagy" RC blond about 60 mm
 approach #81 enter $\frac{1}{2}$, back out XLV came out, ran away
 "Cagy" went in and stayed
- 9/3 Active; later "Cagy" inside
- 9/4 I approach fight "Cagy" I back off and walk NE direct to
 #110N later XLVII try to enter walk back meet "Cagy" both
 jump "Cagy" return to #81 enter
 XLVII leave later "Cagy" walk past Waiting Rock directly
 enter #81 tail first stood at door awhile
- 9/5 $\emptyset$
- 9/7 $\emptyset$
- 9/8 $\emptyset$ later LC male about 50-60 mm in door could not read # on band
- 9/12 Active; big excavation behind just outside chain later $\emptyset$
- 9/13 $\emptyset$ CST II dig under rock behind and outside chain later XIX enter
 CST II approach try to enter XIX wide open seizer II lunge
 try again; later walk around sit under back

110N

7/27 about 50 mm run at light - enter - new fan at North end of 110 (110N)
 7/29 RC about 60 mm
 7/30 about 50-60 mm
 7/31 ☐ female RC 57 banded from 110N
 8/1 ☐ inside
 8/2 ☐ inside
 8/3 ☐ XVII in area - ☐ inside
 8/4 ϕ
 8/5 RC about 60 mm inside ☐ ?
 8/7 ☐ inside
 8/8 ☐ inside
 8/9 ☐ enter
 8/20 ☐ inside
 8/26 ☐ inside
 8/27 ϕ
 8/28 ϕ
 8/29 ☐ inside
 8/30 ☐ inside
 9/2 ☐ bulldoze inside
 9/3 ☐ duck in at light
 9/4 ☐ enter
 9/5 ϕ
 9/7 ☐ enter
 9/8 ϕ
 9/10 ☐ inside
 9/13 ☐ inside
 9/15 ☐ inside

110S

6/2 am 2 claws on fan
 pm 1 claw gone; carapace & merus present
 RC male, hang out, duck in (fairly large)
 6/3 both claws there (LC) + parts of abdomen
 7/4 male 50 mm RC eggshell nearby - didn't band
 7/10 door open
 7/11 door closed with rocks deep in
 7/12 door open
 7/18 small rocks in door
 7/24 RC female in back door
 7/26 RC about 60 mm in hole RC about 30 mm nearby
 7/27 am large fan; molt shell pieces outside
 pm filled in about 40 mm walk nearby
 7/25 ϕ
 7/29 male from mating burrow #29 approach enter back door &
 large male RC no S about 70 mm leave by front door
 8/1 ϕ
 8/3 door has been upened up - algae pushed aside
 8/4 closed with algae
 8/5 male (cut seizer tips) approach 110S front & back door 10-15
 times pushing. Finally XX (male LC 74 mm) came out, ran West
 male (CST) stays inside
 8/7 LC about 80 mm ducks into 110S with light
 8/9 III only tail visible - jerk - jump - exit - walk North
 then West - no other lobster seen in burrow
 8/21 Active
 8/27 large lobster
 8/30 female LC about 60 double band IV? inside
 9/2 full of weeds
 9/3 Active some weeds gone
 9/4 43 (female RC egger) enter
 9/5 ϕ
 9/8 ϕ
 9/12 eel grass covered later RC female 43 inside back door
 9/13 ϕ

IV. DATA PROCESSING

Eleanore Beale and Jelle Atema

The need for computerized data analysis is apparent when one considers the huge volume of collected data in the behavioral studies described in the pertinent sections of this report. For 18 months written recordings have been made three times a day, seven days a week on two separate tanks. Each tank has been set up as a semi-natural environment and is populated with eight lobsters plus all their shelters and food items. Careful analysis of this wealth of data will establish a unique baseline description of lobster behavior that will be used to interpret effects of pollutants on behavior under conditions approaching the "real world" of lobsters.

We have made an agreement with Boston University's Dr. Ivan Valiela to share the use of his Hewlett-Packard System 45 computer, to which we have added a Flexible Disk Drive Master and a Mass Storage ROM to facilitate data analysis. A research assistant has been hired to help in the observation of lobsters, to handle the data, and to operate the computer.

Steps have been taken toward computerizing the data. The shorthand descriptions of lobster behavior have been organized into a succinct code of behavior units (see Table IV. 1). Essential information such as date, time, temperature, lobster identification and location have also been coded. We expect to use this approach for the handling of laboratory, and later field, data to describe sublethal changes in behavior between exposed and non-exposed lobsters.

Table IV. 1. Behavior units for lobsters

alt	alert	an initial response to a stimulus
afl	antenna feel	contact
apo	antenna point	directional
awa	antenna wave	general activity; search
awp	antenna whip	lashing at another animal
awi	antenna wipe	groom antenna with maxillipeds
abb	antennule burst	sudden increase in rate of beating
anf	antennule feel	contact
anp	antennule point	directional
app	approach	frontal, either toward another animal or object
avd	avoid	
att	attack	claws forward, body shoots forward; some combination of snap, lunge, scissor, rush
bkp	back up	backing a short distance
bar	barricade	building a barricade in front of or in shelter entrance
ble	block entrance	male quickly blocks entrance with claws from inside
box	box	post- and pre-molt ritual, agonistic
bdz	bulldoze	pushing or pulling substrate with maxillipeds & 1st legs
bur	bury	bulldoze substrate over a dropped piece of food or molt
car	carry	with claws or maxillipeds
chs	chase	fast following of an object that is moving away
chk	check	brief inspection of shelters - tips of claws in
clt	claw lift	mating ritual : ♂ lifts ♀ claws
clk	claw lock	open crushers interlocked
clc	claws closed	
clo	claws open	
cld	claws down	in ♀ shelter approach, down & closed (as a shield), tips in substrate
cmb	climb	walking to top of shelters
cop	copulate	mating
cwl	crawl	very submissive : body on substrate
cru	crush	grinding food to eat
dfl	dactyl feel	manipulation of substrate

dsw	dactyl sway	walking legs slowly swaying with or without touching substrate
def	defensive posture	raised front, opened claws
dig	dig	digging with walking legs, claws down & closed
dmt	dismount	mating
ent	enter	into a shelter or rock garden
ecy	entrance ceremony	pairing ritual at entrance
fcg	facing	one animal notices another
fco	face off	facing each other within one body length
ffd	fake feed	dactyl or maxilliped in mouth without food
fan	fan	pleiopod beating, creating a strong current
fed	feed	eat
fle	flee	fast forward or sideways running away from scene
fol	follow	one animal walks behind another walking animal
frz	freeze	motionless stance in an interaction
gha	ghost attack	attacking a space where another animal had been
gbb	gill bailer burst	sudden rapid beating of gill bailers
gos	goose	one animal coming up on another's tail
gsp	grasp	with claws or dactyls
grm	groom	cleaning with dactyls and maxillipeds
hgo	hang out	half in and out of shelter
hes	hesitate	momentary halt in forward motion
hol	high on legs	standing on tips of walking legs
jab	jab	poke with a claw
knt	knight	♀ mating ritual
lev	leave	exit from
lun	lunge	thrusting one or both claws forward
man	manipulate	food, substrate, algae moved with mouth parts
mrs	meral spread	claws raised and spread sideways
mnt	mount	mating
mov	move	general movement, often inside of a shelter
ogd	on guard	seizer forward, crusher as body shield
ocr	open crusher	claw open
osz	open seizer	claw open

pnb	pebble nibble	manipulate pebble with walking legs, often prior to fake feed
plb	pleiopod beat	fan
pok	poke	claws pushed into a shelter and withdrawn
pnc	pounce	quick collapse onto substrate with mouth
prp	present rear	mating posture of ♀
psh	push	with claws, mostly social
rer	rear	head up, stand on walking legs
rpo	rest position	quiet stand (usually in or on a shelter)
ret	retreat	backing a long distance
run	run	fast forward, non-social
rsh	rush	fast forward, social
scs	scissor	bringing claws together rapidly from meral spread
scr	scrape	rubbing one or both claws against substrate or rock or shelter
stl	steals	grab food from another animal
spo	submissive posture	body flat on substrate, claws forward
sjb	substrate jab	ritual in mating
snp	snap	rapid closing/opening of seizer, usually with contact
swy	sway	same as dactyl sway including the claws
tlf	tail flip	sudden backwards escape, one or several times
tch	touch	contact with antennae or claws
trn	turn	180° or to follow action
wlk	walk	

V. EXPERIMENTS ON LARVAL SETTLING AND SUBSTRATE SELECTION

Leonor Botero and Jelle Atema

Lobsters live the first weeks of their lives as pelagic larvae before settling at the fourth larval stage into a benthic life style. It is important to know what type of substrate larval lobsters select if one hopes to understand the effect of drilling mud on the process of settlement. One must also understand the mechanism of settlement so that alterations in this behavior in the presence of drilling mud are apparent. Potentially, drilling muds could make large areas unsuitable for settlement and/or survival.

This study has been divided into four parts. For all parts, fourth-stage larval lobsters were obtained from the State Lobster Hatchery in Martha's Vineyard, Massachusetts, and from Marine Research Inc. in Sandwich, Massachusetts. When fourth-stage lobsters were not directly available from the suppliers, first-, second- and third-stage lobsters were reared to fourth stage in our own laboratory in plankton kreisels and fed brine shrimp (Artemia salina) three times a day.

Only larvae that had molted overnight from third to four stage were used to assure that the right stage was being used. The selection of late fourth- and early fifth-stage lobsters for some experiments was determined on the basis of length of time since molting to fourth stage, on the basis of size, and on the presence of two white pigmented areas on the pleura of the first abdominal segment in fifth-stage lobsters.

A. Phototaxis

The pelagic environment in which lobsters live their first weeks is lit brightly in contrast to the benthic world to which they settle. Phototaxis reverses at the time of settling to direct the larvae to shelter and substrate.

These experiments were designed to quantify larval responses to light before and after settling. Changes in the responses caused by pollutants could endanger the very process of settling - a crucial stage in the lobster life cycle.

1. Materials and Methods

Four hundred fourth-stage lobsters were released into the naturalistic 4000-liter aquarium described in Section II. The location of the fourth-stage lobsters in the water column with reference to depth and to section of the tank was monitored. During the night, a white light was flashed toward the water column both from above and from the side of the tank. The response and orientation to the light rays of the pelagic fourth-stage lobsters was observed. After all the lobsters had settled to the bottom, a white light was flashed again toward the water column and toward different spots on the bottom and observations were made on the response to light of the newly settled lobsters.

To test quantitatively the response of fourth- (unsettled) and fifth- (settled) stage lobsters to light, a rectangular transparent plastic tray (50 cm long, 20 cm wide, 10 cm deep) was divided into three continuous sections, each with a different light level, by covering one third of the tray with black plexiglass, one third with red plexiglass, and one third with transparent plexiglass. The lobster could move freely across the tray by either walking or swimming. Twenty lobsters were individually tested; the lobster was introduced into the center of the tray and the plexiglass covers were placed around it in the sequence black, red, transparent. A 20-min adaptation period was allowed, and after that the lobster was watched for 15 min. The approximate time each lobster remained at rest in the black, red or transparent section and the time spent moving around the tray were recorded. Two sets of trials were made, each one with

ten lobsters. For the first set of trials only fourth-stage lobsters were used and for the second set, late fourth- or early fifth-stage lobsters were used.

The data were analyzed first with a simple ANOVA to test for significant differences in the time spent in each situation (black, red, transparent, randomly swimming), and later with the Scheffe multiple comparison procedure to find out where the differences were.

2. Results

Pelagic fourth stage lobsters were strongly photopositive. During the daytime they congregated near the surface of the water column in areas under the light sources that were illuminating the tank. During the night time whenever a white light was flashed toward the water column, an immediate positive response was obtained and they would all swim toward the light source.

After a period of four days the lobsters had disappeared from the water column and settled to the substrate. Because of their small size and the great number of possible hiding places, it was difficult to follow them. When a white light was flashed toward the water column and toward several spots of the substrate, no aggregation of lobsters near the light source was observed. Instead, the few visible lobsters were seen avoiding the light by moving away from it or by hiding under stones or seaweed.

Tables V. 1 and V. 2 present the results of the responses of pre- and post-settling stage lobsters to light when using the tray with three different light regions. Numbers in the table are the approximate time in minutes spent by the lobsters in each situation.

Table V. 1: Approximate time in minutes spent by fourth-stage lobsters in different light exposures or in random movement during an observation period of 15 minutes.

Trial	R e g i o n			Random Movement
	Light	Red	Black	
1	12	1	2	0
2	10	0	1	4
3	12	3	0	0
4	9	2	1	3
5	7	5	1	2
6	7	3	0	5
7	9	1	3	2
8	8	2	2	3
9	7	3	3	2
10	9	2	0	4

Table V. 2: Approximate time in minutes spent by late fourth- and early fifth-stage lobsters in different light exposures and in random movement during an observation period of 15 minutes.

Trial	R e g i o n			Random Movement
	Light	Red	Black	
1	3	5	7	0
2	2	5	8	2
3	3	6	6	1
4	2	4	9	2
5	4	6	5	2
6	4	5	6	3
7	2	3	10	3
8	1	7	7	4
9	0	8	7	5
10	0	5	10	4

A simple ANOVA determined significant differences among the times spent by early fourth-stage lobsters in each of the four situations ($P < .001$). With Scheffe's multiple comparison procedure all the possible paired comparisons between situations were made. The time spent in the light region was greater than the time in either red or black region or in random movement ($P < .05$). The times spent in red and black regions and in movement were equal ($P > .05$). These results show that early fourth-stage lobsters spent significantly more time in the lighted region, confirming the photopositive results obtained from the big tank studies.

For the late fourth- and early fifth-stage lobsters a simple ANOVA determines significant differences among the times spent by the lobsters in each situation ($P < .001$). Scheffe's multiple comparison procedure shows that late fourth- and early fifth-stage lobsters spent significantly more time in the darker region ($P < .05$ in all pair comparisons except lighted vs. random movement where $P > .05$). This confirms the photonegative results obtained earlier for settled lobsters.

3. Discussion

The results indicate that phototactic responses of larval lobsters are correlated with their benthic or pelagic life style. Pollutants, such as drilling mud, could alter these responses through chemical or physical pathways. Changes in the perception of the environment might cause erratic behavior detrimental to the settling process, thus endangering the health of the exposed generation of animals. Studies with drilling mud must be done to show whether this part of the settling behavior is affected.

B. Substrate Choice

Lobsters live most of their lives in the benthic environment and are necessarily intimate with the substrate; both food and shelter are provided by an appropriate bottom. Making a correct choice during settling probably determines the success of an individual lobster. One must understand the patterns of behavior during settling, as well as the ultimate preferences, in order to understand the effects of substrate pollutants on this process.

1. Materials and Methods

The bottom of each of two 96-liter aquaria (55 cm x 55 cm x 35 cm) was divided into four quadrants with 7-cm high pieces of plexiglass. Each quadrant was provided with a different type of substrate. A central standpipe served as a drain and a screen in its hole prevented the lobsters from escaping. Water entered the aquarium through four plastic hoses, one at each corner. Water was allowed to run for 12 hr prior to an experiment to stabilize the conditions in the aquarium. Four types of substrate were used: 1) flat sand, 2) mud collected in Buzzards Bay, 3) gravel, and 4) rocks of approximately 10 cm in diameter with seaweed growing on them (Fucus sp., Ulva sp., Enteromorpha sp.), on top of a one-inch layer of sand. This last section will be referred to as the rock garden.

Ten fourth-stage lobsters were released into each aquarium and their general behavior before and after settling was observed. In each of five trials, the number of fourth-stage lobsters in the water column was recorded at one hour intervals until total settlement.

After total settlement, the number of lobsters on each substrate was determined. The quadrants were first checked visually for any lobsters or burrows easily visible. The number, size, and shape of mud burrows was recorded and the

activity of the lobster living in it was noted. Plexiglass dividers, 20-cm high, were then placed between the quadrants and most of the water was drained with a siphon, taking care to disturb the substrate as little as possible. By moving the rocks and gravel, lobsters were made to come out of their shelters, were counted and were removed from the aquarium. Lobsters living on the sand were easily counted and removed since they had no place to hide. In the mud section the burrows were disturbed to evict the lobsters for counting and removal. If fewer than 10 lobsters were recovered up to this point, the mud was sieved through a 4-mm mesh size screen.

After all the lobsters were found, counted and removed, the four quadrants were rearranged, water was allowed to run for 12 hr and the next 10 fourth-stage lobsters were released. A total of 13 trials were run, each one with 10 lobsters and the same lobster was never used in more than one trial.

The results for the total number of lobsters settled on each substrate were graphed and analyzed statistically using a Chi-square test of significance.

2. Results

After introduction into the aquaria, fourth-stage lobsters spent some time swimming in the water column before settling to the bottom. Swimming was accomplished by fast movement of the swimmerets. During swimming, three positions of the chelipeds were observed: chelipeds pointing straight forward, crossed one over the other in a scissors pattern, or extended to the sides at a 45° angle with the body line. The first two positions were mostly observed in fast and directional swimming while the third position was observed when the lobsters were "relaxing" in the water column, moving their swimmerets very slowly

and showing no directionality of movement. Fast and steep directional "dives" to the bottom were frequent, followed either by fast immediate ascents or by walking on the bottom for short periods of time. A permanent stay at the bottom was never observed after a first descent; several descents and ascents (depending on the individual) were made before permanent settlement occurred.

The first body parts to touch the substrate are the claws and first two pairs of walking legs. The abdomen remains elevated at an angle of about 45° with the body line and while in this position there is always a constant movement of the swimmerets. In this position the lobster "walks" around the bottom. After a while it either starts to swim again or to walk using all four pairs of walking legs, with its whole body in a horizontal position. These walking patterns, interrupted by swimming periods, were observed on all four substrates.

When settling permanently on one substrate, different patterns of activity were observed:

Sand: During the walking and swimming periods, the lobsters commonly descended to the sand. However, except for three of the 130 lobsters tested, they never remained on the sand for more than a few seconds. These three were the only ones that settled permanently on the sand. A small depression dug by them at the corner provided them with some shelter. Attempts to build a burrow under the sand were not successful due to an immediate collapse of the substrate.

Gravel: The lobsters that settled permanently on the gravel used spaces between or under the pebbles for shelter. No digging or active manipulation of the substrate was observed.

Rock garden: A greater number of lobsters settled permanently on this section than on the other three sections. Walking on the rocks

and seaweed followed by swimming periods was also observed. When permanently settled, the lobsters were seen in burrows under the rocks which presumably had been dug by them, since small piles of sand could be observed at the entrances. Some lobsters occupied spaces or crevices between the rocks. However, because of their small size and the number of possible shelters available, only a few of them were seen before the sampling of the substrate took place.

Mud: Due to the similarity of the behavior in this section to the one observed in the mud aquarium described in Part V. C, description will be presented in that section.

Figure V. 1 represents the variation in the number of fourth stage lobsters in the water column through time for these five trials. An average period of 46.6 hours was required for total settlement of the lobsters. A definite trend toward a decrease in the number of lobsters in the water column is observed, as one would expect. Before permanent settlement, there is a great deal of upward and downward movements by the still pelagic lobsters.

Table V. 3 presents the results of the number of lobsters settled on each substrate during each of the 13 trials. A total of 130 lobsters were released and 112 were recovered after sampling the substrates. The total percentages given for each substrate are based on the number of lobsters recovered. A Chi-square value of 71.84 shows a significant difference between the number of lobsters settled on each substrate ($P < .001$).

3. Discussion

It is apparent that a great deal of effort is expended by larval lobsters in choosing a settling environment. This experiment will be modified to test

Figure V. 1. Number of lobsters in the water column over a variety of substrates. Means $\pm$ standard deviation of three trials.

Table V. 3: Number of lobsters recovered from each substrate in each of the 13 trials.

Trial	Substrate				Number Recovered
	Rock Garden	Gravel	Mud	Sand	
1	1	0	8	1	10
2	0	0	6	2	8
3	4	1	1	1	7
4	5	0	2	3	10
5	5	1	0	1	7
6	7	0	0	2	9
7	4	0	3	1	8
8	6	0	1	0	7
9	8	0	0	0	8
10	6	0	1	2	9
11	7	1	2	0	10
12	5	0	3	1	9
13	<u>6</u>	<u>1</u>	<u>1</u>	<u>2</u>	<u>10</u>
Total	64	4	28	16	112
% recovered	49.2	3.07	21.5	12.3	100

pollutants such as drilling mud in order to determine whether the young animals can avoid settling on a contaminated substrate, even if the original surface is as attractive as a rock garden. Since mud is perceived as a suitable if not preferable surface, failure to detect polluted mud could induce serious long-term effects during the constant exposure of post-settling lobsters.

C. Effect of Different Substrates on the Behavior and Time of Settlement

The previous experiments showed how settling larval lobsters respond, given a choice of substrates. It is also important to know how they react when presented with only one choice, since in their pelagic life the larvae can be swept over widely homogeneous parts of the ocean floor. With the disposal of drilling mud, larvae could encounter wide areas covered by this pollutant; it would be to their advantage to be able to postpone settlement until they could swim from or were swept away from the area.

1. Materials and Methods

The same four aquaria, flow conditions and stabilizing times described in Part V. B were used in this experiment. However, here only one substrate was provided in each.

Ten fourth-stage lobsters were introduced simultaneously into each of the four aquaria. Observations on the behavior of the lobsters were made for 20 min after introduction and then for a few minutes at approximately 2-hr intervals until total settlement and establishment. There were two 8-hr intervals during which observations were not made. The behavioral observations are presented as one continuous description for each aquarium. Counts of the

number of lobsters present in the water column were also made every 2 hr (except for the two 8-hr intervals already mentioned). The time required for total settlement of all the lobsters in each aquarium was recorded. Three trials were run for each aquarium, each one with 10 lobsters per aquarium, and the same lobster was never used for more than one trial. Trials were always started at the same time of day; after every trial the aquaria were rearranged and water was allowed to run for 12 hr prior to the start of the next trial.

The results of the number of lobsters present in the water column at each 2-hr interval were analyzed with a Chi-square test of significance to look for differences in the time required for settlement of the lobsters on the different substrates. Fisher's exact probability test was used to determine differences between pairs of substrates.

2. Results

The following observations were made on general behavior:

Sand aquarium: The same swimming and walking patterns described in Part V. B were observed. After descent to the bottom, walking on the sand only lasted for a few minutes. Occasional bulldozing of sand at the corners of the aquarium resulted in small depressions being formed. No burrows were built and usually the lobster would resume swimming promptly. During all observation periods, at least 50% of the lobsters remained in the water column. Two weeks after the lobsters on all other substrates were settled and the recording period of the experiment was finished, 70% of the lobsters on sand substrate were still unsettled.

Mud aquarium: After introduction of the lobsters, the same swimming patterns described in Part V. B were observed. Frequent dives to the bottom were made; each lobster would either initiate an immediate ascent or walk on the mud for some seconds with the abdomen elevated at an angle with the body line as described before. After several seconds of walking it would initiate a fast and directional ascent to the surface and would then swim around the tank. Several minutes later it dived to the bottom, walked around and ascended to the surface again. This diving-walking-ascending sequence was repeated several times until it was ready to settle permanently. At this time, the lobster started walking on the mud for a longer period of time, stopping occasionally to feel the substrate with its dactyls. It would pleopod fan, forming a cloud of mud. Walking was resumed and the lobster would then stop again at one of the corners of the aquarium; here it felt the mud with its dactyls and proceeded to loosen the mud with its claws, in this manner starting its digging activity.

Two lobsters started their digging by diving from the surface very fast and directionally and thrusting their claws into the mud upon reaching the bottom. The loosening of the mud with the claws was followed by strong pleopod fanning and the loose mud was bulldozed out of the hole that was being formed. The lobster then turned, faced the hole and started to dig again. The sequence digging-pleopod fanning-bulldozing was repeated many times until the burrow started taking shape and a clear tunnel was formed. The lobster walked on the surface of the mud to a

distance of about 5-7 cm away from the burrow entrance and started to dig another tunnel in the same manner as described above. The two tunnels eventually connected, forming a U-shaped burrow against the glass wall of the aquarium which could be clearly seen by the observer. Each burrow was occupied by only one lobster. The size and shape of all the burrows observed followed a very similar pattern. Figure V. 2 represents a typical burrow; the measurements obtained from 10 of these burrows are also presented in Table V. 4. The burrows were built initially in a period of 4-5 hr; the inhabitant continued to pleopod fan and to bulldoze mud out of the burrow, forming a small pile of mud outside the entrances. After a period of about 3 days the burrow started to disintegrate and usually collapsed. It was then abandoned by the lobster, who started looking for another spot in which to start a new burrow. Burrows were also built in the central part of the mud substrate. In this case, only the two entrances, usually separated by a distance of 5-8 cm, were visible.

Pebble aquarium: The swimming and walking patterns are the same as already described. After settlement lobsters were found living under the rocks or in between pebbles.

Rock garden aquarium: Settlement to this substrate was very fast compared to the other three aquaria. Lobsters lived in crevices or spaces between the rocks or in burrows under the rocks. Bulldozing of sand from under the rocks was frequent. No lobsters were found living among the seaweed.

Figure V. 2. Diagram of mud burrow dug by a newly settled lobster. 1) Depth of burrow in mud substrate. 2) Distance between entrances. 3) Diameter of tunnel. Note pile of mud at entrances. Measurements of 10 burrows are found in Table V. 4. This diagram represents burrow #3 of Table V. 4.

Table V. 4. Measurements (in cm) of ten burrows of newly settled lobsters. See Figure V. 2. for clarification of measurement location.

<u>Burrow</u>	<u>Depth in mud</u>	<u>Distance between entrances</u>	<u>Diameter of tunnel</u>
1	2.2	7.0	1.0
2	3.0	12	1.0
3	2.2	7.0	1.0
4	2.0	8.0	1.0
5	2.0	5.5	1.5
6	2.9	11	1.0
7	3.0	6.5	1.3
8	3.0	10	1.5
9	2.5	9.0	1.4
10	3.0	8.5	1.0

The time that fourth-stage lobsters remained in the water column before settling was different in the four aquaria. Aquaria with the rocky substrates presented the fastest settlement while the flat sand with no rocks presented the slowest one - almost an absence of it; two weeks after settlement had been completed on the other three substrates, lobsters in the aquaria with a flat sandy bottom were still in the water column. Settlement to the muddy bottom presented an intermediate rate between the ones for the flat sand and the rocky substrates. Total settlement was accomplished after 60 hr on the muddy substrate, 36 hr on the sand with scattered rocks and pebbles, 32 hr on the rock garden, and total settlement was not accomplished on the flat sandy substrate. Figure V. 3 shows the number of lobsters in the water column through time for each aquarium.

To test for significant differences on the rate of settlement on each type of substrate, three time periods were selected: 24, 48 and 64 hr. Table V. 5 presents the total number of lobsters in the water column and on the substrate at the selected time periods. At 24 hr, a Chi-square value of 35.45 ($P < .001$) shows that there is a significant difference in the number of lobsters settled on each of the four substrates. A significant difference between the number settled on sand and mud ($P < .05$), mud and rock garden ($P < .05$), rock garden and scattered pebbles ($P < .05$) is determined using Fisher's exact probability test. Thus, at 24 hr every substrate is different in the number of lobsters settled.

For the 48-hr time period, a Chi-square value of 62.47 ($P < .001$) shows also a significant difference in the number of lobsters settled on each substrate. After 36 hr, however, all the lobsters were settled in both aquaria with rocky substrates. After this time, differences were still apparent in the

Figure V. 3. Number of lobsters in the water column over different substrates. Means $\pm$ standard deviation of five trials.

Table V. 5: Total number of lobsters in the water column and on the substrate at the selected time periods of 24, 48, 64 hr for each of the four aquaria with different substrates.

		Aquarium # - Substrate				Σ
Time Period		#1 - Sand	#2 - Mud	#3 - Scattered Rocks & Pebbles	#4 - Rock Garden	
24 hr	W*	20	12	5	0	37
	B	10	18	25	30	83
	Σ	30	30	30	30	120
48 hr	W	22	5	0	0	27
	B	8	25	30	30	93
	Σ	30	30	30	30	120
64 hr	W	19	0	0	0	19
	B	11	30	30	30	101
	Σ	30	30	30	30	120

* W = water column
B = bottom

other pair comparisons (mud against sand, mud against rock garden, mud against scattered rocks and pebbles). These differences were significant (Fisher's Exact Probability Test, $P < .05$).

At the 64-hr time period all the lobsters except for the ones in the sand aquarium had settled. A Chi-square value of 67.75 and a Fisher's Exact Probability Test ($P < .05$) determined a significant difference between sand and each of the other three substrates.

These data show that the order of preference for settlement was the rock garden substrate, followed by the sand with scattered rocks and pebbles, followed by the mud substrate. The flat sand substrate did not seem appropriate at all for the lobsters to settle on.

3. Discussion

Larval lobsters apparently have the ability to postpone settlement for at least two weeks, as demonstrated in the aquarium with a flat sandy bottom. It has been shown that the purely planktonic three-stage existence of lobsters may include a fourth stage under unfavorable conditions (Templeman, J. Biol. Bd. Canada 3:349-354, 1936). If these young animals were to perceive a pollutant, such as drilling mud, as inhospitable, they would presumably be able to avoid prolonged contact. On the other hand, if drilling mud (or a similarly toxic "ground" cover) were not detected as hazardous, the larvae could well settle and be exposed to long-term sublethal effects. Obviously, experiments need to be done to show how the young lobsters react, as well as to determine lethal and sublethal effects (see Section VI.).

D. Effect of Light and Shelter Availability

The results from Section V. A demonstrate the phototactic responses of larval lobsters before and after settling, and results from Sections V. B and C show substrate preferences of settling lobsters. The experiments described in this part extend these two studies by offering choices of an inhospitable substrate (sand) with and without added shelters and in different light conditions.

1. Materials and Methods

One 680-1 aquarium (130 cm x 90 cm x 60 cm) with running seawater was used. One half of the tank was illuminated with a 15-W bulb hanging 70 cm above the top of the water column; the other half was kept dark by covering it on the top and sides with black cloth. The bottom of the tank was divided into four quadrants. Shelters were built with black and transparent pieces of plastic tubing 15 mm long, and 7 mm in diameter. The pieces of tubing were glued to pieces of wood 3 cm high and 1.5 cm long. The transparent shelters with their wooden bases were glued to the bottom of one of the quadrants in the illuminated section of the tank; the black shelters with their wooden bases were glued to the bottom of one of the quadrants in the dark section. Sand was spread between the shelters. The other two quadrants (one in each section) were provided with a one-inch layer of sand, so as to have one quadrant with flat sand and one quadrant with shelters in each of the illuminated and dark sections.

Eighty-five fourth-stage lobsters were introduced into the center of the tank. After a period of 7 days, when no lobsters were visible in the water column, plexiglass dividers were placed between the quadrants to prevent any

movement of the lobsters from one quadrant to another during substrate sampling. Most of the water was drained and the number of lobsters living in each quadrant was counted. The differences between quadrants were analyzed statistically using a Chi-square test of significance.

2. Results

After five days, no lobsters were present in the water column. When all the lobsters were settled, they were allowed two more days to become firmly established before sampling of the substrate was started. Of the 85 fourth-stage lobsters released, 55 were recovered. Fifty-one of these were living in the dark quadrant containing shelters; some were found walking between the shelters, but the great majority (38) were inside the shelters. The remaining four lobsters recovered were found walking in the dark quadrant containing no shelters. There were no lobsters recovered from the lighted quadrants.

A Chi-square of 100.9 shows a significant difference between the number of lobsters found in the dark sheltered quadrant and the number found in the dark quadrant with no shelters. There is also a significant difference with the other two quadrants in which no lobsters were found.

3. Discussion

Larval lobsters will settle on an undesirable substrate (sand, see Section V. C) when provided with shelter and darkness. Whether they will settle on a polluted surface if other conditions are favorable will be tested by experimentation. If they do, obviously they will be exposed to potentially dangerous effects from the pollutant.

VI. EFFECTS OF DRILLING MUD ON POST-LARVAL LOBSTERS: FEEDING, MOLTING AND SURVIVAL

Shelley Gibbons and Jelle Atema

Post-larval lobsters are the first non-pelagic stage, depending solely on the substrate for shelter and food. Alterations in post-larval behavior obviously may affect future generations of lobsters in an exposed area even if the effects are not readily apparent at the time of dumping. This study explores the behavior, especially feeding, and molt cycle of post-larval lobsters (Homarus americanus) over three months of constant exposure to low levels of drilling muds.

Materials and Methods

One hundred and twenty post-larval lobsters were placed individually into 7.5 cm x 22.5 cm x 5 cm plastic boxes with oyster chips for substrate and a small rock for shelter. The larvae were divided into three groups of 40: one group received a flow-through concentration of 10-ppm drilling mud, a second group received 10-ppm "clean" mud, collected from nearby waters and filtered to a consistency similar to the drilling mud, and the third received plain seawater. One drilling mud (DM-2 -- see Section I, Materials and Methods) has been tested so far, with another sample scheduled to be tested before termination of this experiment on 14 Nov 79. Each group of boxes was placed in a flow-through seawater system topped by a head tank (Fig. VI. 1). The head tanks had a constant inflow of seawater of 9.6 l/min which supplied each box with a flow of 200-240 ml/min. Standpipes in each box drained into a gutter system

Figure VI. 1. Flow-through dosing system for post-larval lobsters.

The system has three head tanks: two 45 cm x 50 cm conical tanks and one 37.5 cm x 45 cm square tank. The inflows for the conical tanks enter from the bottom through two 90° turns facing the bottom of the tank. This creates a swirling action which hinders sedimentation. The square tank inflow enters from the top. All three tanks have an inflow of 9.6 l/min. and each one supplies 40 boxes, each with an inflow of 200-240 ml/min. Standpipes in each box drain into a gutter system which empties into a holding tank before entering an acrylic fiber filter box. This filter removes the drilling mud from the water before it reaches the drain.

which emptied into a holding tank before entering an acrylic-fiber filter box. This fiber removed drilling mud from the water before reaching the drain and was changed twice a day to insure thorough filtering.

The lobsters were allowed to acclimate for three days at which time the few dead animals were removed. During this time and during the experiment each lobster was fed five brine shrimp twice daily. Muds were introduced into the head tanks by syringe pump at a constant rate to deliver 10 ppm. The muds were injected into a jet stream of water within the head tanks, breaking up any coagulations which result when the mud enters seawater. Swirling action created by the conical shape of the head tanks and the downward jet stream of water kept the muds in suspension in the head tanks. Settling did occur in the distribution system and individual boxes. To counteract this, the distribution system was cleaned once a week and the boxes every three days.

The following parameters were compared among the three groups: 1) time to alert to the presence of food (alert), 2) time to find food (hit), 3) amount eaten, 4) time between molts, 5) size after each molt, 6) location (in or out of shelter, recorded three times daily). All molt shells not eaten by the lobsters were removed and frozen individually for future chemical analysis.

Results

We observed no statistically significant differences between exposed lobsters and the controls for any of the parameters (Table VI.1). There were no observable differences between "clean" mud controls and the seawater controls. However, drilling mud-exposed animals tend to be found in their shelters barricaded with oyster chips more often than the controls. This did not appear to alter feeding or molting.

Table VI..1. Effect of drilling mud on feeding, molting and shelter occupancy of post-larval lobsters. Values given for drilling mud (DM), coastal ocean mud (CM) and plain seawater (C) are expressed as means $\pm$ standard deviation.

	DM (n = 40)	CM (n = 37)	C (n = 38)
Alert (min)	8.9 $\pm$ 2.5	9.2 $\pm$ 2	9.0 $\pm$ 2
Hit (min)	28 $\pm$ 5.7	24 $\pm$ 4.2	27.3 $\pm$ 4.9
Shrimp eaten per day	10 $\pm$ 0	10 $\pm$ 0	10 $\pm$ 0
Molt 5 $\rightarrow$ 6 (days)	10 $\pm$ 2	9.3 $\pm$ 4	11 $\pm$ 3.7
6 $\rightarrow$ 7	15 $\pm$ 3	14.8 $\pm$ 2.2	15.2 $\pm$ 4.2
7 $\rightarrow$ 8	16.6 $\pm$ 2.6	17.9 $\pm$ 4	18 $\pm$ 3.7
Time found in shelter (%)	84 $\pm$ 8	35 $\pm$ 4	26 $\pm$ 7
No. lobsters that died during experiment	3	2	1

Discussion

In general no deleterious effects of this drilling mud (DM-2) were observed. The clean-mud controls showed no behavioral differences from the seawater controls. After the dramatic effects observed in Dec 78 with DM-1 on adult lobsters (Section I), the results with this drilling mud are surprising. They point out the tremendous variability in drilling muds and pose many questions pertinent to the development of efficient, effective, and financially feasible dumping regulations. It seems possible that certain drilling muds, such as DM-2, may be safely (and economically) dumped off drilling rigs. Others, such as DM-1, would severely damage lobster populations in the exposed area and very likely be expensive to treat or dispose of safely. Hence a clear idea of what constitutes a toxic mud is necessary before reasonable, effective dumping regulations can be established. Only through further biological testing coupled with chemical analysis of each tested mud can one hope to understand drilling mud and deal with its disposal.

VII. NEUROPHYSIOLOGICAL ANALYSIS OF THE EFFECTS OF DRILLING MUD ON THE ACTIVITY OF CHEMORECEPTORS

Charles D. Derby and Jelle Atema

Chemoreception is probably the dominant sensory modality with which lobsters and most other benthic marine organisms relate to the environment, using it for obtaining food or mates and avoiding danger. Direct pollutant effects on the chemoreception process can be measured by a neurophysiological test such as the one described here. Leg chemoreceptors are of particular interest since these are heavily exposed in cases where a pollutant is part of the substrate. Leg receptors are essential in feeding behavior.

Materials and Methods

The effects of short-term exposure of drilling mud (DM) on the physiology of chemoreceptors were examined by recording neurophysiological responses from chemoreceptors on the walking legs of lobsters. Legs from three groups of lobsters exposed to DM-2 were used: non DM-exposed; light DM-exposed; and heavy DM-exposed (see Section I, Materials and Methods).

Each leg was removed from an animal and dissected to expose the sensory nerve bundle. The last two leg segments, which remain intact, were inserted through a rubber dam in a plexiglas chamber so that the leg section was isolated from the exposed nerve (Fig. VII. 1). The nerve was bathed in lobster Ringer's solution, and a constant stream of filtered seawater (10 ml/min) flowed over the distal leg region. Chemical stimuli (50 μ m) were injected into this flow, and extracellular responses were recorded from chemoreceptive neurons that had been separated from the sensory nerve bundles.

Figure VII. 1. Diagram of leg olfactometer. Seawater (A) flows over the distal segment of the dactyl. Stimulus is introduced at (B) into the flowing seawater, so that it is swept past the dactyl chemosensory hairs. A rubber dam (C) and plexiglass plate (D) prevent the stimulus from exciting the exposed nerves (E), which are bathed in Homarus Ringer (F). A hook electrode picks up one of the nerve bundles (E) for recording against a reference electrode (G).

Two chemical stimuli known to be important feeding attractants were used: mussel extract, and 10^{-5} M L-glutamate. At least two responses were recorded for each stimulus tested, with a thorough rinsing period following the termination of each response. Responses to artificial seawater and a 10-ppm drilling mud solution were also recorded. Following this first set of recordings, the seawater flow over the leg was replaced by 10-ppm DM in seawater, flowing at the same rate as before. After 3 min of exposure to the 10-ppm DM, the previous stimuli were again tested to make a second set of recordings. The 10-ppm DM flow was then replaced by the seawater flow, and a third set of responses was recorded after 3 min of rinsing. Responses were tested again with a 10-ppm DM background flow for a final fourth set of recordings. Each series of four sets was made from only one nerve bundle per leg to avoid effects of previous exposures on subsequent tests. Responses were recorded from 7 legs of previously non DM-exposed lobsters and from 7 legs of DM-exposed lobsters. Of the exposed animals, 4 received heavy exposure and 3 light exposure. Magnetic tape recordings of the responses were transcribed onto a Brush chart recorder from which the number of action potentials in a response and the action-potential temporal patterns could be analyzed.

Results

Injection of DM itself into the seawater background flow caused a response on a non DM-exposed leg in one case, indicating that this type of DM can be sensed by leg chemoreceptors.

Short-term DM exposure clearly caused interference in 4 of the 7 legs of previously non DM-exposed lobsters; i.e., the responses to mussel extract (Fig.

VII. 2) and L-glutamate (Fig. VII. 3) were consistently lower when introduced in the presence of DM background than in the seawater background. The degree of interference was variable. No interference was observed for the previously light and heavy DM-exposed lobsters.

Discussion

It is clear from this study that brief exposure to low concentrations of drilling mud can interfere with the normal activity of chemoreceptors on the walking legs of lobsters. The interference mechanism was not determined in this study. It could possibly result from a physical blockage of the receptor surface such that odorant molecules cannot reach the receptor sites on the chemoreceptor membrane surface. Another possibility could be that there is a change in the membrane resting potential (hyperpolarization).

Interference was observed in 4 of 7 of the lobsters not previously exposed to drilling mud, but not in the previously exposed animals. This could be only a problem of the sample size. However, it is possible that there are only a few receptors that are affected by this type and concentration of drilling mud. Those receptors that are susceptible are already interfered with in previously exposed lobsters and cannot therefore be located by the means used in this procedure.

Receptors on walking legs are used in sensing food items. Without chemosensory input, the localization and handling of food is less efficient. Therefore, if many receptors are blocked, it may affect the response to feeding stimuli. Behavioral experiments are necessary to examine if drilling mud has such effects on lobster feeding (Section I).

Figures VII. 2 and VII. 3.

Neural responses of leg chemoreceptors from non DM-exposed lobsters. Responses from the same nerve bundle were recorded for various stimuli introduced into different backgrounds. Arrow indicates time of injection of stimulus. Parenthetical DM and SW indicate background flow into which stimuli were introduced.

- Figure VII. 2. A. Mussel extract with filtered seawater background
 B. 10-ppm drilling mud with filtered seawater background
 C. Mussel extract with 10-ppm drilling mud background
 D. Mussel extract with filtered seawater background
 E. Mussel extract with 10-ppm drilling mud background

- Figure VII. 3. A. 10^{-5} M L-glu with filtered seawater background
 B. 10-ppm drilling mud with filtered seawater background
 C. 10^{-5} M L-glu with 10-ppm drilling mud background
 D. 10^{-5} M L-glu with filtered seawater background
 E. 10^{-5} L-glu with 10-ppm drilling mud background

A. Mussel (SW)

B. DM (SW)

C. Mussel (DM)

D. Mussel (SW)

E. Mussel (DM)

Figure VII. 2

A. L-glu (SW)

B. DM (SW)

C. L-glu (DM)

D. L-glu (SW)

E. L-glu (DM)

Figure VII. 3

VIII. "BOXING MATCHES" AS A BIOASSAY FOR POLLUTANT EFFECTS

Thomas J. Trott, Elisa B. Karnofsky and Jelle Atema

The importance of aggressive behavior for the maintenance of territories has been demonstrated for many species in a variety of social contexts. Although yet to be substantiated by field studies, laboratory observations suggest that the American lobster, Homarus americanus, may establish "territories" within an animal's area of general activity. With the exception of one study (Scrivener, 1971, Fish. Res. Bd. Can. Tech. Rep. 235), the aggressive behavior of mature lobsters is known only from casual observations. To reach some understanding of lobster social behavior, which could be used to measure effects of pollutants, we began to study aggressive interactions in more detail during the spring and summer of 1979.

Materials and Methods

Submature lobsters (6 males, 3 females) were held in individual 115-liter tanks supplied with flow-through seawater at ambient temperatures. The animals were allowed two weeks to acclimate to laboratory conditions before the experiment began. Boxing matches were 15 min long and held in 680-liter aquaria with a pebble substrate and flow-through seawater. Two lobsters were placed in the fighting tank with a divider between them two hours before the match began. After raising the divider, events occurring during aggressive interactions were recorded by two observers. (Aggressive units are described in Table IV. 1). After each match the animals were returned to their respective tanks.

Three groups of 3 animals (2 male groups and 1 female group) were formed. Fights were conducted only between animals belonging to a group. A dominance

order was established within each group by a series of 3 fights. Lobsters held either an alpha, beta or gamma position depending upon the number of matches they won or lost. All animals fought twice a week. After this baseline, dominance and aggressive behavior were then followed over several months for all animals.

Results and Discussion

Our observations of boxing matches have indicated several interesting features about their aggressive behavior. Damage to an opponent during fights between animals of the same sex never occurred. Many behavior units relating to claw posture appear to be visual displays. It may at first seem unusual to interpret claw postures as visual displays since lobsters are active most at night. However, the coloration of the underside of each claw is quite visible to us under low intensity light. Lobsters are probably at least as sensitive to low light as we are. For many claw postures the undersurface of the claws is presented to an onlooking opponent. This case is especially true for the "hands-up" posture when a beaten animal attempts to place outstretched claws on either side of the opponents rostrum while walking forward. In contrast, in matches held between male and female lobsters, females hold their claws down and folded in front of them. Since the underside of the claws is not visible in this position, a female may appear less aggressive. The evolution of visual displays in the aggressive behavior of lobsters might explain the low incidence of damage during fighting. Obviously, if drilling mud has any effect upon the neuromuscular system, such interference might result in the abolishment of some claw displays. The implications for competition are self-evident and will be tested later.