108401

Data Evaluation Record

Field study

1. Chemical: Bolero

2. Formulation: Bolero 8EC

3. Citation: Studies in Halls Bayou to Test the Effects of a Pre-Emergent Herbicide, Bolero, on Aquatic Organisms by Donald Harper, et al., Harper Environmental Consulting Company, Galveston, Texas, 1979. Ref. 29 in Fish and Wildlife Safety Data to Support Registration of Bolero 10G Herbicide (acc. #241484).

4. Reviewed by: Ann Rosenkranz
Aquatic Biologist
HED/EEB

5. Date Reviewed: August 5, 1980

6. Test Type: Field Tests:
Nekton
Benthos
Phytoplankton

7. Reported Results: No definitive conclusions can be made regarding the effects of Bolero; however, the benthic community appears to be most affected by Bolero.

8. Reviewer's Conclusions: The study may be of some use in assessing the risks of Bolero to estuarine species.

(1)

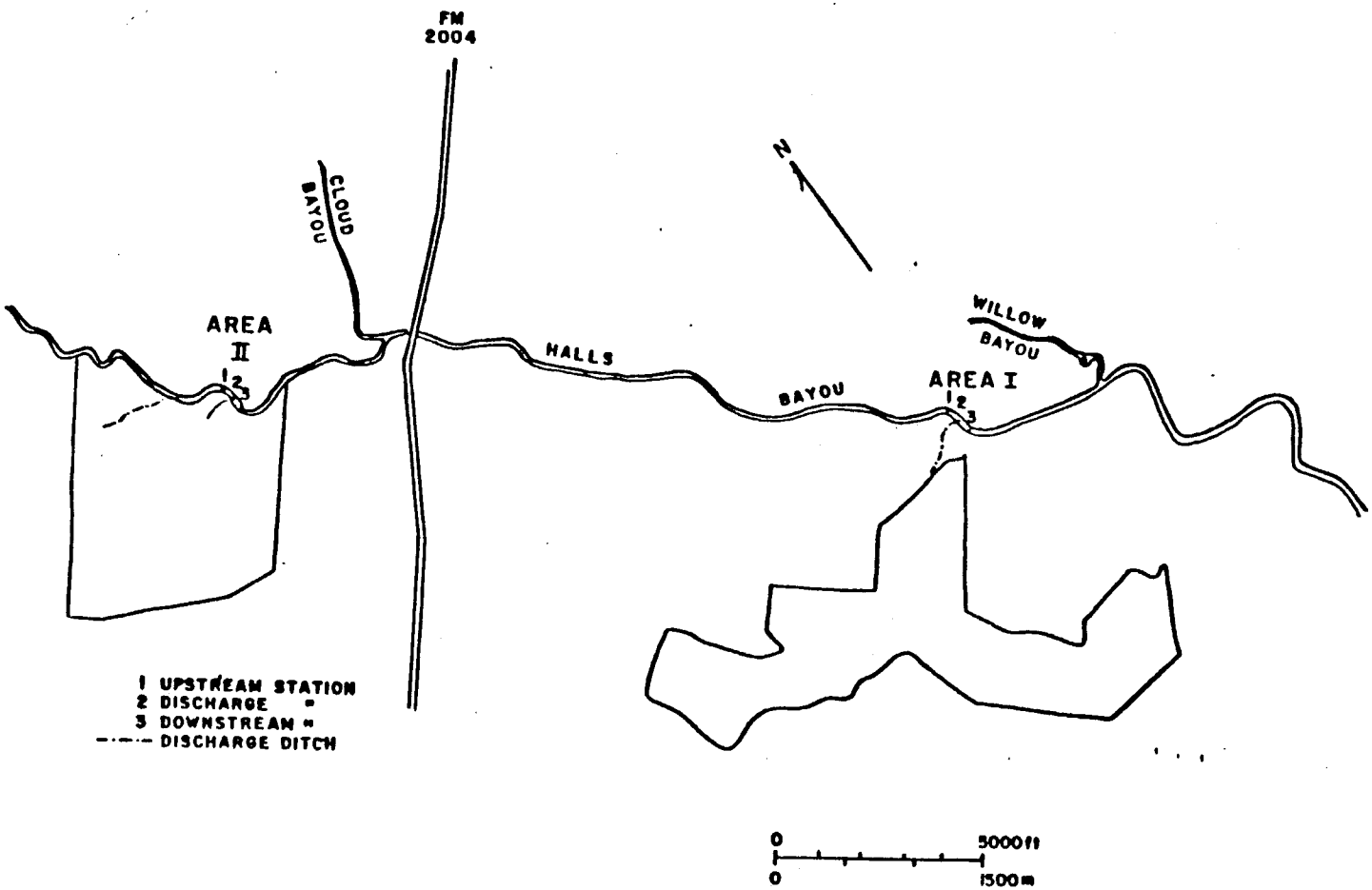
# STUDIES IN HALLS BAYOU TO TEST THE EFFECTS OF A PRE-EMERGENT HERBICIDE, BOLERO, ON AQUATIC ORGANISMS

## Introduction

The purpose of the experiments described in this report was to field test the toxicity of a rice field pre-emergent herbicide, Bolero, to aquatic biota. The usual practice in rice field preparation is to apply herbicides, flood the field, then drain the water into a local waterway. The plants and animals inhabiting the waterway are exposed to the herbicide that dissolves in the flood water. The experimental fields in this study bordered Halls Bayou, a tidally influenced, narrow stream that empties into West Bay of the Galveston Bay system. A description of the study area is provided below.

The specific groups of organisms field tested for effects caused by Bolero were the nekton (fish and swimming invertebrates such as shrimp and crabs), the benthos (small invertebrates living in and on the bottom muds) and phytoplankton (minute photosynthetic organisms that swim feebly, if at all). In addition, organisms of commercial and/or ecological significance were to be placed in cages at each site and monitored for signs of distress. The caged organisms included fish, grass shrimp, oysters and brackishwater clams.

Results of the study may have been influenced by rice farming practices and weather. Some rice farmers drained their fields before and while the experimental fields were drained and introduced unknown quantities and types of pesticides into the bayou. In addition, chemicals were aerially applied to adjacent fields while the study was in progress and unknown quantities of these chemicals fell directly on the water. Finally, heavy rainstorms occurred prior to (19, 20, 21 March and 2, 3 April) and during the study (5, 6, 18, 19, 20 April) washing unknown quantities of materials into the bayou from both farmed and unfarmed land in the watershed of the bayou. This is especially true of the rainfalls at the end of March and end of April since they initiated unscheduled flush overflows.




Map of the portion of Halls Bayou investigated during this study. Shown are the rice fields, the drainage ditches through which flush water was discharged into the Bayou, and the location of the sampling stations.

Table 8. Thiobencarb residues in flush water collected at the field outlets and in Halls Bayou (Area I, Halls Bayou).

| Date (1979) | Days after application | Thiobencarb residues (ppm)[1] in water samples from sample site[2] | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | B (Field number) | | | | Bayou sampling sites | | |
| | | B-96 | C-1 | C-14 | C-15 | E | F' | F |
| 3-20 | pretreatment | | | | | 0.000 | | 0.000 |
| 3-20 | | BOLERO APPLIED | | | | | | |
| 3-22 | | UNSCHEDULED FLUSH OVERFLOW | | | | | | |
| 3-22 | 2 | 0.142 | 0.59 | | | 0.000 | 0.011 | |
| 3-23 | 3 | | 0.315 | | | 0.000 | 0.000 | |
| 4-6 | | SCHEDULED FLUSH OVERFLOW | | | | | | |
| 4-6 | 17 | | 0.027 | | | 0.000 | <0.002 | 0.003 |
| 4-13 | | | | BOLERO APPLIED | | | | |
| 4-19 | | UNSCHEDULED FLUSH OVERFLOW | | | | | | |
| 4-20 | 32 | 0.005 | 0.007 | | | 0.006 | 0.006 | |
| 4-20 | 7 | | | 0.091 | 0.097 | | 0.005 | |

[1] Figures indicate mean values from duplicate sample analysis. Limit of detection 0.002 ppm.

[2] Location B represents the field outlets. Location E was 500' upstream and F' was 500' downstream of the site where the field water entered the bayou (Fields B-96, C-1). Location F was 500' downstream of the bayou entry site for Fields C-14 and C-15.

Table 9. Thiobencarb residues in flush water collected at the field outlets and in Halls Bayou, Area II.

| Date (1979) | Days after application | Thiobencarb residues (ppm) in water samples 1/ from sample site 2/ | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | B (Field number) | | | | | Bayou sampling sites | |
| | | B-12 | B-13 | B-14 | B-15 | B-16 | E | F |
| 3-20 | Pre-treatment | | | | | | 0.002 | 0.003 |
| 3-24 | | ----- BOLERO APPLIED ----- | | | | | | |
| 4-4 | | | | | | SCHE-DULED FLUSH OVER-FLOW | | |
| 4-4 | 11 | | | | | 0.160 | | < 0.002 |
| 4-5 | | | SCHE-DULED FLUSH OVER-FLOW | | SCHE-DULED FLUSH OVER-FLOW | | | |
| 4-5 | 12 | | 0.203 | | 0.021 | 0.139 | 0.041 | 0.033 |
| 4-6 | 13 | | | | 0.179 | | 0.048 | 0.041 |
| 4-7 | | | | SCHE-DULED FLUSH OVER-FLOW | | | | |
| 4-7 | 14 | | | 0.253 | 0.177 | | 0.083 | 0.064 |
| 4-19 | | BOLERO AP-PLIED | | | | | | |
| 4-19 | | -- UNSCHEDULED FLUSH OVERFLOW -- | | | | | | |
| 4-19 | 0 | 8.900 | | | | | | |
| 4-20 | 1 | 0.300 | | | | | 0.000 | 0.000 |
| 4-21 | 2 | 0.410 | | | | | 0.000 | 0.000 |
| 4-21 | 28 | | 0.039 | 0.031 | 0.024 | | | |

1/ Figures indicate mean values from duplicate sample analysis. Limit of detection 0.002 ppm.

2/ Location B was the field outlet, location E 500' upstream and location F 500' downstream of the drainage water discharge site into the bayou.

Halls Bayou is a narrow waterway, 10 miles long, that meanders through part of the Texas coastal plain. Its mouth is a relatively short distance from the Gulf of Mexico, and, therefore, the bayou is tidally influenced.

The field collections consisted of collecting representative samples of the organisms that occur naturally in Halls Bayou. This aspect of the study began prior to the start of discharge from the rice fields and continued until after a second flush. The total span of the field study was from 2 February through 25 April 1979. The purpose of the field studies was to determine if either the species present or numbers of individuals (populations) were altered in response to the introduction of water containing Bolero.

The specific groups of organisms sampled were the fish and nektonic invertebrates caught in seine, trawls and gill nets, the benthic invertebrates collected by grab sampler, and the phytoplankton collected by water samplers. Each of the above-mentioned biological units is discussed in a separate section.

## I. Nekton

### Test Procedure

### Materials and Methods

Nektonic macroinvertebrate and fish populations of Halls Bayou were sampled with gillnets, bag seines and otter trawls. Experimental gillnets, consisting of five 7.6 x 1.8 m (25 x 6 ft) panels of 2.5 cm (1 in), 3.8 cm (1.5 in), 5.1 cm (2 in), 6.4 cm, (2.5 in) and 7.6 cm (3 in) square mesh, sampled those highly mobile organisms which avoided seines and trawls. Gillnets were set off the bottom across the width of the bayou adjacent to upstream (Station E) and downstream (Station F) cages in Areas I and II. Twelve-hour sets (1800 to 0600) were made in both areas on 5-6 (1 day prior to discharge), 6-7 (peak flow), 11-12 (7-days post-discharge), and 21-22 April (1 day after second discharge). Gillnet catches were sorted to species and enumerated in the field.

Nearshore fish and macroinvertebrate assemblages of Areas I and II were sampled with a 7.6 m (25 ft) long, 1.8 m (6 ft) deep seine having a 1.8 x 1.8 x 1.8 m bag. Wings of the seine contained 6.4 mm (1/4 in) square mesh while the bag was composed of 3.2 mm (1.8 in) square mesh. Seine samples were composed of all organisms captured while towing the net through a $90^{o}$ arc. Three replicate seine samples were taken 30.5 m (100 ft) above and below the point of flush in both areas. Seining operations were commenced at 0600 in each area.

Trawl samples of demersal macroinvertebrates and fishes were collected at Area I only; submerged obstructions prevented trawling in Area II. The midchannel was sampled with a 3.3 m (10 ft) otter trawl having 2.5 cm (1 in) square mesh throughout the body and cod end. A 6.4 mm (1/4 in) square mesh

liner inside the cod end enabled capture of small specimens escaping through larger meshes of the trawl. Trawls were deployed in midstream areas adjacent to upstream and downstream bioassay cages and were towed upstream for 3 minutes. Trawling was begun at 0600 and was concurrent with seining operations.

Gillnet, seine and trawl samples were frozen to preserve the samples prior to either sorting or subsequent tissue analysis. Hydrological data were taken immediately prior to biological sampling and monitoring bioassay cages. Measurements were recorded from upstream and downstream stations in both areas for 96 hours after peak flush and every 12 hours thereafter. Hydrological parameters measured concurrently with biological sampling included water temperature, salinity and dissolved oxygen content. Water temperature and salinity were measured to the nearest 0.1 C and 0.01 ppt, respectively, with a Beckman Instruments Model RS5-3 salinometer. Dissolved oxygen content was measured to the nearest 0.1 ppm with a Yellow Springs Instrument Company Model 54A Oxygen Meter. Hydrological measurements were taken at nearshore and midchannel areas immediately adjacent to bioassay cages positioned 150 m (500 ft) upstream and downstream from the point of flush in Areas I and II. Surface, middepth and bottom readings were taken at 1-m intervals.

## Statistical Analysis

None was performed.

## Results and Discussion

### I. Abiotic Characteristics

Water temperatures during 4-12 April ranged from 17.6°C to 22.5°C and averaged near 20.5°C. Differences between study sites were slight and did not fluctuate more than 1° or 2°C. Upstream and downstream stations in each area had similar temperatures. There was no thermal stratification at any site.

Area I had a higher salinity than Area II during the early phase of monitoring from 4-6 April, which was caused by a weak salt wedge penetrating the bottom waters of Area I. The peak salinities in Area I during this period were 3.47 ppt upstream in the midchannel and 3.42 ppt downstream. Salinity decreased to less than 2.00 ppt upstream and downstream in Area I and less than 0.5 ppt in Area II after the heavy rains and flushing in early April. After 8 April, the salinity in both areas remained near 1.30 ppt.

Dissolved oxygen content remained near 5.0 ppm although low levels were recorded in Area I 5-6 April. These values of 1.6-3.3 ppm were associated with the salt wedge. D.O. levels in Area I frequently exceeded those in Area II by 1.0 ppm; this was probably due to a large amount of decaying leaf mulch along the bottom of Area II.

The sediments were virtually identical at all stations. It was a thin layer of oxidized, light brown, very fine silt over a layer of reduced, black mud.

II. Biotic Characteristics

A. Gillnet Data

Species Collected - Area I

Blue crab
Spotted gar
Longnose gar
Gizzard shad
Threadfin shad
Carp
Smallmouth buffalo
Channel catfish
Sea catfish
Freshwater drum
Black drum
Striped mullet

Species Collected - Area II

Blue crab
Spotted gar
Longnose gar
Gizzard shad
Carp
River carpsucker
Smallmouth buffalo
Channel catfish
Warmouth
Longear
White crappie
Freshwater drum
Striped mullet

Although Area II yielded a larger total catch (111 specimens) and total number of species (13) than were collected at Area I (80 specimens belonging to 12 species), species composition in both areas was relatively similar (9 taxa held in common). The spotted and longnose gars, gizzard shad and blue crab accounted for over 61% of the combined catch from both areas.

In area I the total abundance [combined catches at Stations E (upstream) and F (downstream)] increased during and after the initial discharge from the fields (April 7 and 12, respectively). The total abundance on April 22, during a second discharge, was less than that of April 12 but greater than that of April 7. Although catches at downstream Station F were equal to or greater than those at upstream Station E on April 6, 7, and 12, the 2 stations were similar with regard to changes in abundance through time.

In Area II, the total abundance declined during the discharges of April 7 and April 22. The largest total abundance occurred on April 12, 7 days after peak flow. This latter catch was the only one where the number caught downstream exceeded the number caught upstream.

Total Abundance - Area I

| Date | 6 Apr | | 7 Apr | | 12 Apr | | 22 Apr | |
|---|---|---|---|---|---|---|---|---|
| Station | E | F | E | F | E | F | E | F |
| Total catch per Station | 0 | 6 | 6 | 11 | 18 | 17 | 13 | 9 |

Total Abundance - Area II

| Date | 6 Apr | | 7 Apr | | 12 Apr | | 22 Apr | |
|---|---|---|---|---|---|---|---|---|
| Station | E | F | E | F | E | F | E | F |
| Total catch per Station | 23 | 5 | 8 | 2 | 30 | 38 | 5 | 0 |

B. Seine Data

Major Species - Area I

Gulf menhaden
Grass shrimp
Mysis shrimp

Major Species - Area II

Gulf menhaden
Mysis shrimp

A total of 37 taxa was seined in the 2 areas, 35 in Area I and 25 in Area II. Twenty-two species were common to the 2 areas.

Mean Abundance of Macroinvertebrates and Fishes from Replicate Seine Samples - Area I

| Date | 29 Mar | | 6 Apr | | 7 Apr | | 12 Apr | | 22 Apr | |
|---|---|---|---|---|---|---|---|---|---|---|
| Station | E | F | E | F | E | F | E | F | E | F |
| Grand Mean Per Station | 1361 | 458.3 | 110.3 | 470.7 | 158.4 | 229.3 | 186.3 | 342.0 | 40.6 | 341.0 |

Mean Abundance of Macroinvertebrates and Fishes from Replicate Seine Samples - Area II

| Date | 29 Mar | | 6 Apr | | 7 Apr | | 12 Apr | | 22 Apr | |
|---|---|---|---|---|---|---|---|---|---|---|
| Station | E | F | E | F | E | F | E | F | E | F |
| Grand Mean Per Station | 514.0 | 1978.3 | 297.7 | 747.3 | 1744.3 | 863.3 | 309.3 | 423.7 | 48.3 | 35.3 |

In Area I, the highest combined mean abundance was recorded on March 29, when no fields were being drained. The lowest catches were recorded during the discharges on April 7 and April 22. At all sampling times but March 29, the mean catch for the downstream station was greater than that for the upstream station.

In Area II, the highest combined mean abundance was recorded on April 7 and the lowest was recorded on April 22, the days of peak flow and second discharge, respectively. The combined abundance on March 29, when there was no discharge, was similar to that of April 7. The catches at the upstream station exceeded that of the downstream station during discharge on April 7 and April 22.

C. Trawl Data

Species Collected

| | |
|---|---|
| Blue crab | Carp |
| Grass shrimp | Blue catfish |
| River shrimp | Channel catfish |
| Mysis shrimp | Atlantic croaker |
| Spotted gar | Clown goby |
| Longnose gar | Southern flounder |
| Gulf menhaden | Hogchoker |
| Bay anchovy | |

Total Abundance - Area I

| Date | 6 Apr | | 7 Apr | | 12 Apr | | 22 Apr | |
|---|---|---|---|---|---|---|---|---|
| Station | E | F | E | F | E | F | E | F |
| Total catch per Station | 718 | 709 | 674 | 236 | 413 | 587 | 73 | 90 |

A total of 3500 specimens was caught during the study period. The Atlantic croaker and the blue crab were the dominant species, comprising over 84% and 12%, respectively, of the total catch. The highest total catch occurred on April 6, 1 day prior to peak flow, and the lowest catch occurred on April 22. The total abundance of this latter catch was in turn 6 times less than that of the catch of April 7, the date of the first discharge.

Species composition at both stations was similar. Eleven species were caught downstream and thirteen upstream, with nine species held in common. Trawl catches between the two sites were similar, except on April 7, when the catch downstream was significantly less than the catch upstream.

Data from gillnet, seine and trawl samples taken in Areas I and II revealed few faunaldifferences between upstream (control) and downstream (affected) sites which could be associated with the discharge of Bolero. Gillnet data from Area I did not exhibit the declines in downstream nekton populations normally associated with the discharge of a suspected pollutant, unlike the data from Area II. However, the data from both areas indicate that the effects of flushing water from the fields have an impact on not only downstream sites but also upsteam sites.

Although the seine data indicate that nekton abundance decreased once discharging was started, more organisms were collected downstream than upstream. Likewise, trawl data reveal little difference between upsteam and downstream sites.

It is very likely that the changes in abundance and species composition were influenced by environmental factors such as tidal cycles, salinity changes and the concurrent release of other pesticides from neighboring areas into Halls Bayou and cannot be soley attributed to Bolero.

Reviewer's Evaluation

Test Procedure

Materials and Methods

Although the basic sampling techniques are valid, they are not sufficient to assess changes in species composition and population dynamics in Halls Bayou. The introduction to the nekton study states that preliminary sampling was conducted at least one month prior to the first flushing of the fields. Supposedly this data would determine species composition and population density prior to introduction of a potential toxicant into the ecosystem, but these data are not included in this report.

A preferred method would have been to conduct the preliminary sampling the previous April in order to determine typical species composition, density, distribution and migrations for this time period. If this was not possible, then a better method to estimate population, at least for the fish species, would have been to use an accepted capture-recapture method.

Statistical Analysis

Instead of using total abundance as a measure of pesticide impact, a species diversity index was calculated for upstream and downstream sites on each sampling date in both areas, since changes in species diversity are indicative of environmental changes. A common index is the Shannon-Weiner index:

$H = -\sum p_i \ln p_i$, where pi is the proportion of total sample belonging to ith species.

Additionally, an index of similarity between Areas I and II was calculated for each sampling method. The formula is: $S = 2c/a+b$, where a = number of species in Area I, b = number of species in Area II, c = species occurring in both areas. The results are as follow:

//

## Gillnet - Species Diversity Index (H) and Index of Similarity (S)

| | 6 Apr | | 7 Apr | | 12 Apr | | 22 Apr | |
|---|---|---|---|---|---|---|---|---|
| | E | F | E | F | E | F | E | F |
| Area I | | | | | | | | |
| H | 0 | 1.24 | 1.33 | 1.03 | 1.23 | 1.76 | 1.52 | 0.64 |
| S | 0 | | 0.25 | | 0.83 | | 0.29 | |
| Area II | | | | | | | | |
| H | 1.87 | 0.50 | 1.56 | 0.69 | 1.82 | 2.04 | 0.95 | 0 |
| S | 0.36 | | 0.57 | | 0.67 | | 0 | |

Index of similarity between Areas I and II = 0.72.

## Seine - Species Diversity Index

| | 29 Mar | | 6 Apr | | 7 Apr | | 12 Apr | | 22 Apr | |
|---|---|---|---|---|---|---|---|---|---|---|
| | E | F | E | F | E | F | E | F | E | F |
| Area I | | | | | | | | | | |
| H | 0.83 | 0.91 | 2.30 | 2.10 | 0.79 | 1.28 | 0.64 | 0.87 | 2.07 | 0.67 |
| S | 0.81 | | 0.80 | | 0.74 | | 0.58 | | 0.71 | |
| Area II | | | | | | | | | | |
| H | 0.79 | 0.70 | 0.37 | 0.22 | 0.22 | 0.15 | 0.19 | 0.29 | 1.54 | 1.53 |
| S | 0.67 | | 0.59 | | 0.59 | | 0.77 | | 0.58 | |

Index of similarity between Areas I and II = 0.73

## Trawl-Species Diversity Index

| | 6 Apr | | 7 Apr | | 12 Apr | | 22 Apr | |
|---|---|---|---|---|---|---|---|---|
| | E | F | E | F | E | F | E | F |
| Area I | | | | | | | | |
| H | 0.48 | 0.07 | 0.57 | 0.72 | 0.56 | 0.43 | 1.09 | 0.87 |
| S | 0.46 | | 0.67 | | 0.62 | | 0.83 | |

Index of similarity between Stations E and F = 0.75

## Results and Discussion

This study was part of the EUP that was granted to Chevron in early 1979. However, some discrepancies were noted between this report and the one entitled "Final Report of the Field Study" by J.E. Lauck with regard to rainfall and residue data.

Chevron's comments on this report indicate the levels of thiobencarb in Halls Bayou at the cage sites from 4 April to 12 April. They are (in ppm):

| | Area I | | Area II | |
|---|---|---|---|---|
| | Site E | Site F | Site E | Site F |
| 4 Apr. | - | - | - | 0.002 |
| 5 Apr. | 0.002 | 0.002 | 0.04 | 0.033 |
| 6 Apr. | 0.000 | - | 0.048 | 0.041 |
| 7 Apr. | 0.084 | 0.012 | 0.083 | 0.064 |
| 8 Apr. | 0.031 | 0.051 | - | 0.033 |
| 9 Apr. | 0.044 | 0.010 | - | 0.010 |
| 10 Apr. | - | - | - | 0.011 |
| 11 Apr. | 0.009 | 0.011 | - | 0.014 |
| 12 Apr. | - | - | - | 0.021 |

A comparison of the above residue values with those in Tables 8 and 9 (from "Final Report of EUP Field Study") indicates that the results for Area I do not agree. Also, the "Final Report" gives the dates of significant rainfall as 22, 23, 24, 25 March, 2, 3, 4 April and 18, 19, 20 April. The rainfalls in March and the end of April caused unscheduled flush overflow. The Harper report varies slightly in the dates given for the early April rainfall, 5 and 6 April, in addition to the 3 earlier days. This may be significant since it indicates that it rained during the scheduled flush overflow. It is important to clarify these discrepancies.

### A. Gillnet Data

The index of similarity indicates that species composition was similar for both areas. However, the species caught on each sampling day were not always the same for the upstream and downstream stations. In Area I, nothing was caught upstream on 6 April prior to discharge, whereas 4 species were caught downstream. Species composition was most similar for these 2 stations on 12 April, 7 days following the initial discharge and very different on 7 April and 22 April, the days following discharge. In Area II, species composition was most similar for the upstream and downstream stations on 12 April and more different on 6 April. In this area, no specimens were caughtdownstream on 22 April.

When the indices of diversity for the 2 areas are examined, it is evident that the downstream site is most diverse on 12 April. The downstream sites are less diverse than the upstream sites at all other times in Area II and on 7 April and 22 April in Area I.

The report only examined the data in terms of total numbers caught and did not find any trends that could be related to the discharges from the Bolero-treated fields. However, the differences between the upstream and the downstream stations in each area on 7 April and 22 April, the days immediately following discharge, as compared to the days either prior to or sometime after discharge indicates that something is affecting the nekton community.

Undeniably, the natural environmental fluctuations affected these results. One aspect that was quite often referred to in the report was the salinity differences in early April (through 8 April). A stepwise multiple regression was done of the gillnet data for both the upstream and downstream stations in Area I to learn if species diversity was affected by temperature, salinity, and/or dissolved oxygen content. Since there were not enough data points, a good fit could not be obtained. Salinity may be a factor in the changes in diversity upstream, but not downstream. Some other factor is operating to cause the shifts in species composition on the days immediately following the discharges of flushwater from the Bolero-treated fields.

B. Seine Data

The mean abundance data were incorrectly reported. The correct values are:

Area I

| 29 Mar | | 6 Apr | | 7 Apr | | 12 Apr | | 22 Apr | |
|---|---|---|---|---|---|---|---|---|---|
| E | F | E | F | E | F | E | F | E | F |
| 1361 | 1791.6 | 110.3 | 469.1 | 158.4 | 229.3 | 186.3 | 342 | 40.6 | 958.6 |

Area II

| 29 Mar | | 6 Apr | | 7 Apr | | 12 Apr | | 22 Apr | |
|---|---|---|---|---|---|---|---|---|---|
| E | F | E | F | E | F | E | F | E | F |
| 503.8 | 1978.3 | 297.7 | 747.3 | 1744.3 | 863.3 | 309.3 | 423.7 | 48.3 | 54.2 |

Unfortunately, the theories regarding shifts in species composition that were discussed in the section on gillnet data do not apply to these data since the shifts do not follow any specific pattern.

C. Trawl Data

As for the seine data, the shifts in species composition cannot be correlated with any specific environmental factors.

Although the gillnet data indicate that Bolero may be affecting that segment of the nekton population, the seine and trawl data do not corroborate it. However, the influence of Bolero along with fluctuating ambient conditions are all operating. More and better designed testing is needed to resolve the problem.

Conclusions

1. Category: Supplemental

2. Rationale: Poorly designed study that does not allow for proper assessment of role of Bolero in affecting nekton populations. However, there is some information in the study that may be useful in a risk assessment.

3. Repairability: Study should be run again, but with better experimental design.

## Benthos Study

### Test Procedures

### Materials and Methods

The experimental design was identical at both study areas. The primary station was located in the middle of the bayou directly off the mouth of the discharge canal where Bolero concentrations should have been highest. One station was located 150 m (500 ft) upstream and one was 150 m downstream from the primary station. The boat was anchored, fore and aft if necessary, to maintain station location.

At each station, six replicate benthic samples were collected using a 232 $cm^2$ (6 in x 6 in) Ekman grab. The surficial sediments were removed from one sample, placed in an aluminum foil package, and stored in a cooler. The other mud samples were washed on a 0.5 mm (U.S. #35) mesh sieve and fixed in 5% formalin. Abiotic data recorded included the temperature, salinity and dissolved oxygen (D.O.) of samples of the surface and bottom of the water column, and the temperature of the sediments. The characteristics of the sediments were also recorded.

Each station was sampled five times during the study. The first collection was made on 3 April, the day prior to flooding the fields. The second and third collections occurred on 6 and 7 April during early and peak discharge flows, respectively. The fourth collection was made on 12 April, a week after discharge began. The last collection occurred on 25 April, a week after a second discharge to drain excess rain water off the fields.

On 3 April, the field investigators were led to a site considerably upstream from the Area II location by Chevron personnel, and the "Area II" data collected on this date are not comparable with subsequent data from Area II.

The sediment samples were frozen at the Texas A&M Marine Laboratory and later transferred to Chevron personnel for analysis. The preserved samples were washed with fresh water on a 0.5 mm mesh sieve to remove formalin and any remaining sediments. The material retained on the sieve was preserved in rose Bengal stained 70% ethanol. After 24 hours had elapsed, the samples were examined microscopically and all stained organisms were removed and placed in 70% ethanol. The organisms in 3 of the 5 replicate samples were subsequently identified to lowest possible taxon and counted.

### Statistical Analysis

N/A

## Results and Discussion

Table 1 - Summary of benthic data collected in Halls Bayou during April 1979. The 9 most abundant and frequently occurring species are listed in order of total abundance. Total number of individuals collected each sampling date are listed. *-largest population during study, @-data not collected at the Area II site and are included for comparison only.

AREA I

| Organism | 3 | 6 | 7 | 12 | 25 | TOTAL |
|---|---|---|---|---|---|---|
| Streblospio benedicti | 426 | 563* | 548 | 353 | 301 | 2191 |
| Hypaniola floridus | 273 | 280 | 371* | 239 | 204 | 1367 |
| Tubificidae | 51 | 55 | 73* | 69 | 61 | 309 |
| Chironomidae D | 32* | 22 | 11 | 10 | 10 | 85 |
| Chironomidae E | 10 | 7 | 3 | 10 | 23* | 53 |
| Chironomidae F | 4 | 1 | 4 | 7 | 22* | 38 |
| Chaoborus | 2 | | | 1 | 9* | 12 |
| Chironomidae B | | 1 | 5* | 2 | | 8 |
| Others | 5 | 3 | 4 | 10 | 5 | 27 |
| TOTAL INDIVIDUALS | 803 | 932 | 1019 | 701 | 635 | 4090 |
| TOTAL SPECIES | 11 | 11 | 12 | 13 | 12 | |

AREA II

| Organism | 3@ | 6 | 7 | 12 | 25 | TOTAL |
|---|---|---|---|---|---|---|
| Tubificidae | 362 | 200 | 183 | 192 | 335* | 910 |
| Hypaniola floridus | 125 | 149 | 164 | 172* | 123 | 608 |
| Streblospio benedicti | 4 | 48* | 39 | 42 | 9 | 138 |
| Chironomidae D | 10 | 43* | 23 | 16 | 2 | 84 |
| Chironomidae E | 15 | 30* | 19 | 17 | 25 | 91 |
| Chironomidae F | 12 | 2 | 3 | 12 | 14* | 31 |
| Chaoborus | 6 | | 2 | 1 | 8* | 11 |
| Tendipidae | | 1 | 1 | 1 | 2* | 5 |
| Chironomidae B | | | | | 1* | 1 |
| Others | 8 | 2 | | 4 | 4 | 10 |
| TOTAL INDIVIDUALS | 542 | 475 | 434 | 457 | 523 | 1889 |
| TOTAL SPECIES | 10 | 10 | 9 | 11 | 13 | |

Twenty one species or other lower taxa of benthic invertebrates were collected during the study. Annelids, principally two polychaetes, Streblospio benedicti and Hypaniola floridus, and tubificid oligochaetes overwhelmingly dominated the populations. Insect larvae comprised the only other relatively abundant group.

The total number of species collected in Area I was nearly constant and does not reflect any environmental stress. Comparison of the numbers of species collected at each station in Area I through time reveals no pattern that can be interpreted as having been caused by any factor. The total number of species collected in Area II increased slightly after the experimental discharge, but this does not suggest an environmental stress. The numbers of species collected at each station in Area II through time were more stable than at Area I, and there was a relative uniformity between stations. These data do not indicate that Bolero induced a toxic response in the benthic populations.

Of the 21 species or taxa collected during the study, only 6 were common to all collections at both areas (Table 1). At both sites, these six species accounted for 99% of all individuals collected during the study.

The changes in total population density at the two study areas were different. In Area I, there was an apparent rapid increase in density from 3 April (pre-discharge) to 7 April (peak discharge flow) followed by an equally rapid decrease by 12 April. The population decline continued through 25 April (subsequent discharge). The two numerically dominant species in Area I, the polychaetes Streblospio benedicti and Hypaniola floridus, attained largest populations on 6 and 7 April, respectively, and then decreased in abundance. The population of tubificid oligochaetes, which were third in total abundance, did not vary as much during this time as the two dominants. Three of the chironomid insects were most abundant on the last day of sampling.

At Area II, the total number of individuals collected was always smaller than at Area I, and the temporal variability was much less; there was a slight increase in population density following discharge. Tubificid oligochaetes and Hypaniola, the two numerical dominants, increased in abundance after the experimental discharge of water from the fields. The third most abundant organism, Streblospio, was over an order of magnitude less abundant in Area II than Area I, even when data from the discounted 3 April collection are included.

The numbers of individuals collected at each station varied widely through the course of the study in both Areas I and II. There was no pattern suggesting that the populations at any station were depressed consistantly following discharge.

If study Area I alone is considered, the rapid decrease in population density following discharge of water from the rice field suggests that something in the water affected the organisms. The conflicting total population trends at the two study areas, however, do not support this premise, and it is probable that some other factor caused the decrease.

The temperature of both water and sediments, the dissolved oxygen, the sediment composition, and presumably the amounts of Bolero entering the water, were similar at both areas. The salinity is the only abiotic factor that was different. Hydrographic measurements, determined from water samples, indicated that the bottom water salinity at Area I was about 1 ppt through 7 April and 0.5 ppt or less thereafter, while less than 0.5 ppt at all times in Area II. Landry, however, measured salinity with a probe lowered from the boat, and recorded bottom water salinities of about 3.5 ppt at Area I through 6 April and less than 1.6 ppt thereafter (refer to report on Nekton study). His measurements at Area II showed that the salinity was less than 1.4 ppt throughout the study.

The organisms collected during this study are characteristic of low salinity to freshwater environments, and suggest that the salinity had been depressed for quite some time prior to the first collection on 3 April. When a saline regime is replaced by fresh water, the salinity of the sediment interstitial water is reduced more gradually, and benthic organisms adapted to higher salinities may survive for some time after freshwater conditions are established.

The dominant organism in Area I, Streblospio benedicti, tolerates low salinities, but near freshwater conditions are apparently lethal. Hypaniola floridus is tolerant of lower salinities than Streblospio.

The population decrease at Area I occurred soon after the salinity decreased from near 3.5 to less than 1.6 ppt. At Area II, where the salinity was relatively stable (less than 1.4 ppt), the populations did not change appreciably. These data strongly suggest that the decrease in population density following discharge from the field was caused by a coincident salinity decrease.

Reviewer's Evaluation

Test Procedure

Materials and Methods

The study follows accepted field procedures. One complaint is that the surface area of each sampling site should have been given since population density is defined as the number of organisms of each species per unit area. Another complaint is that the time of day for each collection should have been indicated since the area is tidally influenced.

Statistical Analysis

N/A

## Results and Discussion

Changes in relative abundance of the dominant species, the polychaetes Streblospio benedicti, Hypaniola floridus and the Tubificidae species, are a better indicator of stress than absolute numbers. The relative abundance of each species in relation to the total number of specimens at each sampling time was determined for the upstream, downstream and point of discharge collection sites for each sampling time. The results are listed in Table 2. The relative abundances for the 3 species at each site were plotted against sampling time in order to see if any changes in population could be correlated with the flushing schedule of the fields and/or recorded salinity changes.

When the data are examined by the above methods, it is noted that for Area I Streblospio is generally most abundant and the Tubificidae the least abundant at each site throughout the study. The report states that the polychaetes decreased in abundance after April 7. This may be true in terms of absolute numbers, but in terms of relative abundance, the populations of Streblospio at the discharge site and Hypaniola at the upstream site increased sharply after April 12. The relative abundances of the Tubificidae were generally stable at each site. For Area II, Hypaniola decreased in abundance and the Tubificidae increased at the 3 sites after April 12.

Although changes in salinity may be partially responsible for the changes in relative abundance of the 3 species through time, other factors, including tidal cycles and the presence of Bolero in the water, are responsible for either the unexpected differences in relative abundance of each species at each site on any given day (Table 2) or the response of the Tubificidae in Area II. Although the upstream sites were supposed to be control sites, on several occasions (April 12 and 25) species abundances were lower when compared to the other two sites. Additionally, responses at the sites of discharge were not always the lowest (April 7 - day of peak flow).

Although low salinity may be responsible for the low abundance of Streblospio in Area II, it cannot account for the responses of Hypaniola (which one would have expected to increase as in Area I) and the Tubificidae (indicator species for stressed environments). Something other than solely salinity is causing the decreases in relative abundance of the two latter species - possibly Bolero.

Therefore, the influence of Bolero cannot be discounted, particularly in Area II. However, since the study was conducted in an estuary, which is a dynamic ecosystem, many other factors are also operating. The data do not allow for any definitive conclusions.

## Conclusions

1. Category: Supplemental.
2. Rationale: The data are inconclusive as to the effect of Bolero on the benthic organisms, although the presence of Bolero in the water cannot be discounted as causing the observed changes in population.
3. Repairability: N/A.

Table 2

Area I

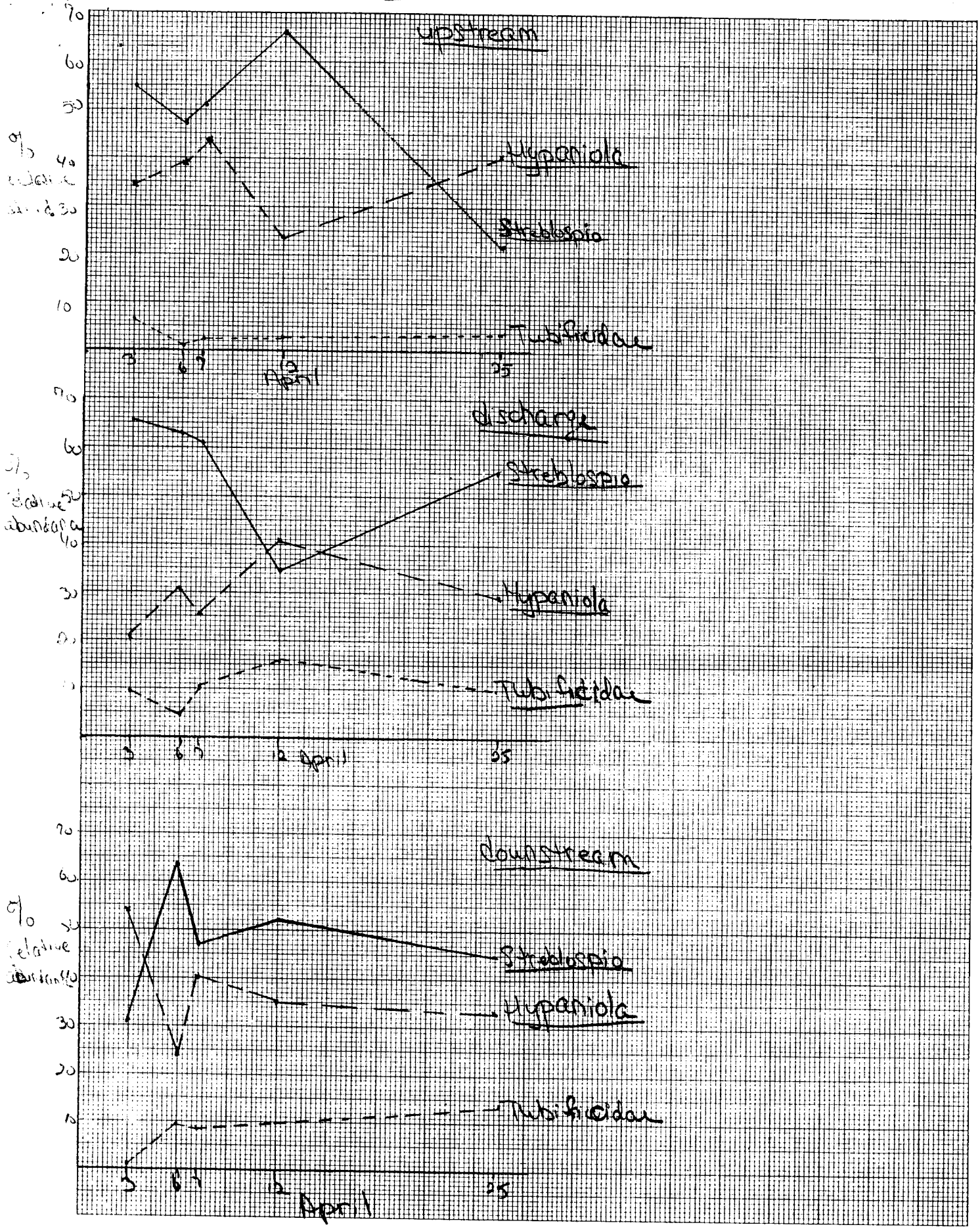
| Species | Upstream | Discharge | Downstream | Date |
|---|---|---|---|---|
| Streblospio benedicti | 55.0 | 65.6 | 31.4 | 3 Apr |
| | 47.5 | 63.2 | 63.7 | 6 Apr |
| | 51.3 | 61.3 | 47.2 | 7 Apr |
| | 66.1 | 34.7 | 52.0 | 12 Apr |
| | 22.1 | 55.2 | 44.8 | 25 Apr |
| Hypaniola floridus | 34.7 | 20.7 | 54.1 | 3 Apr |
| | 39.0 | 30.6 | 24.3 | 6 Apr |
| | 44.0 | 25.5 | 40.2 | 7 Apr |
| | 23.8 | 41.1 | 35.0 | 12 Apr |
| | 40.7 | 29.3 | 33.6 | 25 Apr |
| Tubificidae A | 6.5 | 9.5 | 1.3 | 3 Apr |
| | 1.1 | 4.8 | 9.8 | 6 Apr |
| | 2.6 | 10.2 | 8.9 | 7 Apr |
| | 3.0 | 15.8 | 9.7 | 12 Apr |
| | 4.4 | 9.9 | 13.8 | 25 Apr |

Area II

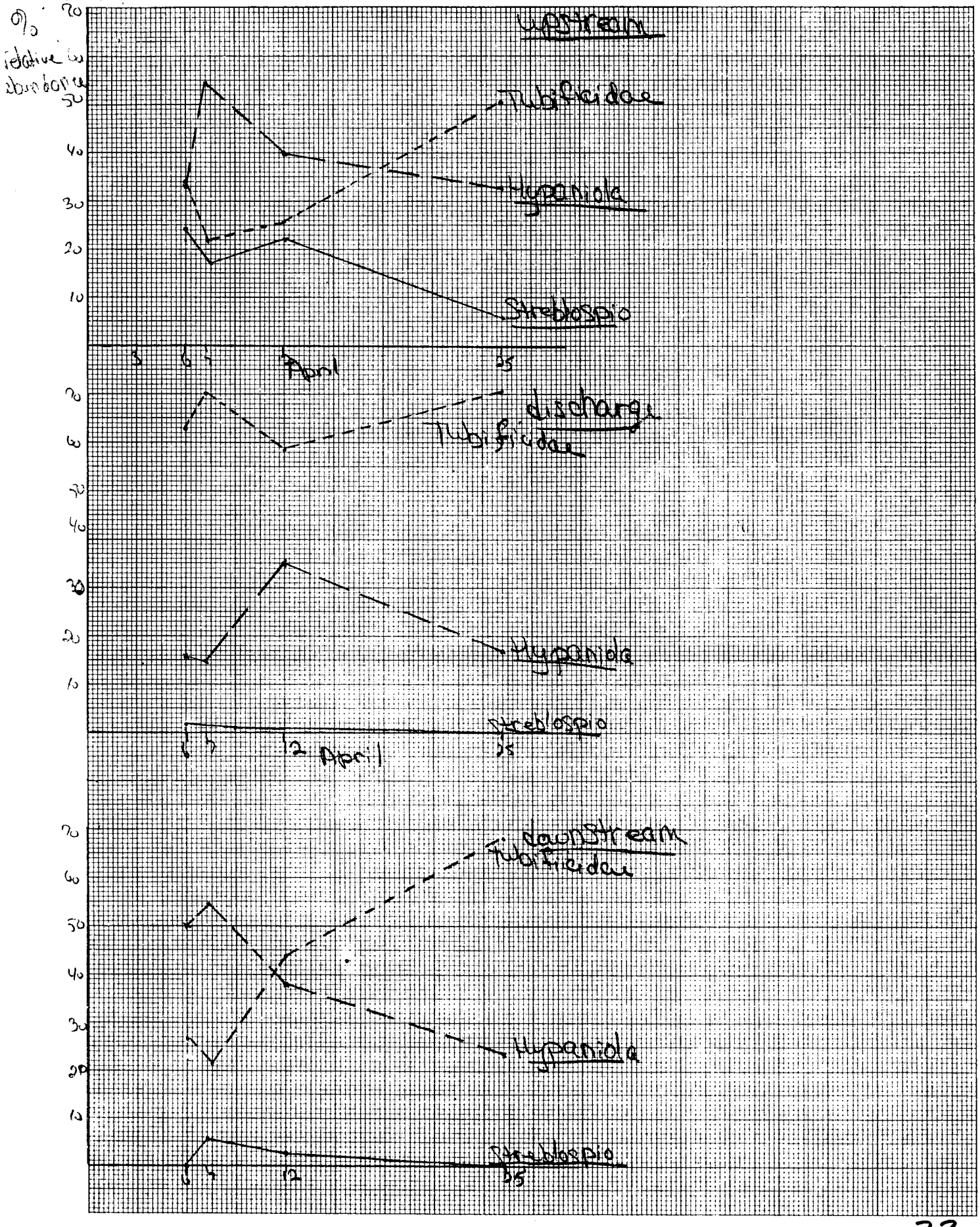
| Species | Upstream | Discharge | Downstream | Date |
|---|---|---|---|---|
| Streblospio benedicti | 24.3 | 1.8 | 0 | 6 Apr |
| | 17.3 | 1.1 | 5.5 | 7 Apr |
| | 22.3 | 0.7 | 2.8 | 12 Apr |
| | 6.0 | 0 | 0 | 25 Apr |
| Hypaniola floridus | 33.0 | 15.7 | 50.0 | 6 Apr |
| | 54.3 | 14.8 | 54.5 | 7 Apr |
| | 39.8 | 34.7 | 38.2 | 12 Apr |
| | 32.9 | 16.9 | 23.4 | 25 Apr |
| Tubificidae A | 33.6* | 63.3 | 26.7* | 6 Apr |
| | 21.8* | 70.3 | 21.8 | 7 Apr |
| | 25.9 | 58.5 | 43.8 | 12 Apr |
| | 50.3 | 70.5 | 68.3 | 25 Apr |

*A and B

Area I


upstream
Hyperniola
Streblospio
Tubificidae
5
6 7
12 April
25
% relative abundance
discharge
Streblospio
Hyperniola
Tubificidae
5
6 7
12 April
25
% relative abundance
downstream
Streblospio
Hyperniola
Tubificidae
5
6 7
12 April
25

# Area II


%
relative abundance
upstream
Tubificidae
Hyperiidae
Streblospio
5
6
April
25
discharge
Tubificidae
Hyperiidae
Streblospio
12 April
25
downstream
Tubificidae
Hyperiidae
Streblospio
12
25

## Phytoplankton Study

### Test Procedures

### Materials and Methods

In most cases a pair of 1-liter water samples were taken at each collecting site. The samples were collected directly from the surface area with the sample container (1-liter glass jars). One sample ("fixed" sample) was preserved with 1% formalin and the other ("live" sample) was untreated in order that an assessment of live organisms could be made. The preservation usually ruptures many of the unarmored flagellates and dinoflagellates. The "live" samples, which were analyzed within 12 to 24 hours after collection, were maintained in an ice chest or cool room until analyzed.

At approximately 1500 hours on 3 April 1979, "live" and "fixed" samples were taken from the reservoir water supply just before its entrance into a treated rice field in Area II. A few minutes later a second set of "live" and "fixed" samples were taken in about the middle of the treated field during flooding. A second series of phytoplankton samples were taken between 0645 and 0700 hours on 6 April 1979. This series was taken during the first discharge from the treated field that was sampled on 3 April 1979. On this latter occasion a pair of water samples (one "fixed" and one "live") were taken from each of three sites: the reservoir supply, approximate middle of the treated field and the discharge point. On 6 April, three additional "live" samples were taken between 0705 and 0715 hours from Halls Bayou at Area II. The bayou depth in this region was about 3 m (9 ft) deep, and the samples were all taken in the approximate middle of the bayou about 1.3 m (4 ft) below the surface. These three samples were taken with a Kemmerer water sampler. One "live" sample was collected at the downstream site, another at the upstream site, and the third opposite the discharge point.

The phytoplankton in the "live" and "fixed" samples were identified to genus and counted. The counts obtained for each genus found in the sample were used to calculate the number of individuals per ml of water. The population density calculated for each genus was rounded to the nearest 100 individuals.

### Statistical Analysis

N/A.

## Results and Discussion

Table 1 - Phytoplankton Population (no./ml) of Reservoirs, Area II, Halls Bayou at 1500 hours, 3 Apr 79, and 0645-0700 hours, 6 Apr 79. ("Live" Samples)

| Class | Organism | 3 Apr | 6 Apr |
|---|---|---|---|
| Bacillariophyceae (Diatoms) | Caloneis | 1900 | |
| | Navicula | 2100 | |
| | Nitzschia | 500 | 300 |
| | Surirella | 200 | |
| | Thalassiosira | * | |
| | Diatoma | | 500 |
| | Fragilaria | | 200 |
| Chlorophyceae (Green algae) | Chlamydomonas | 4500 | 900 |
| | Pteromonas | 2400 | |
| | Stichococcus | * | |
| Chrysophyceae (Golden-brown algae) | Chromulina | 2200 | |
| | Epichrysis | | 2900 |
| Dinophyceae (Dinoflagellates) | Cysts | 200 | |
| | Gymnodinium | | 1000 |
| | Glenodinium | | 600 |
| Myxophyceae (Blue-green algae) | Merismopedia | * | |
| Xanthophyceae (Yellow-green algae) | Gloeobotrys | 2000 | 4300 |
| Total Number Per Ml | | 16,000 | 10,700 |

*Only found in fixed sample which was disregarded.

Table 2 - Phytoplankton Population (no./ml) of Treated Rice Field, Area II, Halls Bayou, at 1500 hours, 3 Apr 79, and 0645-0700 hours, 6 Apr 79. ("Live" Samples)

| Class | Organism | 3 Apr | 6 Apr |
|---|---|---|---|
| Bacillariophyceae (Diatoms) | Caloneis | 500 | * |
| | Navicula | 1000 | 900 |
| | Nitzschia | 700 | * |
| | Thalassiosira | 1900 | |
| Chlorophyceae (Green algae) | Chlamydomonas | * | 800 |
| | Stichococcus | 200 | * |
| | Chlorococcum | | * |
| | Scenedesmus | | 2900 |
| Dinophyceae (Dinoflagellates) | Gymnodinium | 100 | 700 |
| | Gyrodinium | 100 | |
| | Peridinium | 200 | |
| | Prorocentrum | 100 | |
| Myxophyceae (Blue-green algae) | Merismopedia | 200 | |
| | Microcystis | 900 | 2000 |
| | Raphidiopsis | 100 | 200 |
| Xanthophyceae (Yellow-green algae) | Gloeobotrys | 800 | 1100 |
| Total Number per Ml | | 6800 | 9400 |

*Only found in fixed sample which was disregarded.

Table 3 - Phytoplankton Population (no./ml) of Discharge Ditch from Treated Rice Field, Area II, Halls Bayou, 0645-0700 hours, 6 Apr 79. ("Live" Sample)

| Class | Organism | "Live" |
|---|---|---|
| Bacillariophyceae (Diatoms) | Chaetoceros | 3100 |
| | Navicula | 500 |
| | Pinnularia | * |
| | Thalassiosira | 200 |
| Chlorophyta (Green algae) | Chlamydomonas | 1000 |
| | Chlorococcum | * |
| | Palmella | * |
| Chrysophyceae (Golden-brown algae) | Rhizochrysis | * |
| Dinophyceae (Dinoflagellates) | Gymnodinium | 500 |
| Euglenophyceae (Euglenoids) | Euglena | 700 |
| Xanthophyceae (Yellow-green algae) | Gloeobotrys | 2900 |
| Total Number per Ml | | 8900 |

*Only found in fixed sample which was disregarded.

Table 4 - Phytoplankton population (no./ml) in Area II region of Halls Bayou. "Live" samples taken from middle portion, approximately 4 feet below surface, 0705-0715 hours, 6 Apr 79. (Live Sample)

| Class | Organism | "Upstream" | "Discharge" | "Downstream" |
|---|---|---|---|---|
| Bacillariophyceae (Diatoms) | Chaetoceros | 3400 | 2400 | 1400 |
| | Navicula | 400 | 1000 | 200 |
| | Nitzschia | | 200 | 300 |
| | Thalassiosira | 1200 | 1900 | |
| Chlorophyceae (Green algae) | Chlamydomonas | 1000 | 1700 | 2200 |
| | Scenedesmus | | 700 | |
| | Stichococcus | 1500 | 500 | 1000 |
| Chrysophyceae (Golden-brown algae) | Chrysacapsa | 200 | | |
| | Ochrymonas | 1700 | 2400 | 2900 |
| | Rhizochrysis | 500 | 1700 | 1400 |
| | Synura | | | 1700 |
| Dinophyceae (Dinoflagellates) | Gymnodinium | 1000 | 1600 | 1200 |
| Euglenophyceae (Euglenoids) | Euglena | 200 | 1400 | 500 |
| Xanthophyceae (Yellow-green algae) | Gloeobotrys | 5800 | 5500 | 4100 |
| Total Number per Ml | | 16,900 | 21,000 | 16,900 |

The results of the phytoplankton analysis are presented in Tables 1 through 4. Five to six classes of phytoplankton were encountered in each of the samples. The classes included a wide range of organisms, consisting of dinoflagellates, euglenoids and algae such as greens, blue-greens, golden-browns and yellow-greens.

As expected, the total population density was greater for the "live" samples than for the "fixed" ones. A comparison of the total population for five pairs of "live" and "fixed" samples shows the "live" samples yielded two to three times more organisms than the "fixed" ones. Moreover, greater numbers of a particular class or genus were encountered in the "live" than the "fixed" samples. With few exceptions, all forms found in "fixed" samples were also encountered in the "live" ones. Thus, the data obtained from "fixed" samples will be disregarded in evaluating the effects of Bolero on phytoplankton.

Although there was considerable variation in relative abundance of various groups, the diatoms, green algae, golden-brown algae and yellow-green algae appeared to be the dominant phytoplankters. Also, there is some suggestion that the abundance and species composition of the reservoir water supply differed on 3 April from that of 6 April. For example, diatom and green algae populations appeared to be more abundant on 3 April than 6 April. On the other hand, the density of golden-browns and yellow-greens was similar on both dates. Another group, the dinoflagellates, appeared to be more abundant in the reservoir supply on 6 April than 3 April.

An inspection of Tables 1 and 2 suggests that the total density of organisms in the reservoir supply wasabout twice as great as that encountered in the sample from the middle of the treated field during the flooding period (3 April 1979). A similar consideration of all the Tables shows no appreciable difference in the total number of individuals found in the reservoir supply, treated field or discharge ditch during the first discharge of water from the treated field (6 April 1979). The concentrations of phytoplankton per ml were 10,700, 9,400 and 8,900 in the reservoir supply, treated field and discharge ditch, respectively.

Samples taken in the middle of Halls Bayou on 6 April (Table 4) showed very similar total population densities and species composition at the three sites sampled. The total number of individuals per ml was 16,900, 21,000 and 16,900 at upstream, discharge and downstream sites, respectively. These values are roughly twice as great as those encountered in the water entering and leaving the treated field.

The significance of this study is limited by the fact that there were samll numbers of samples. Since the dispersion of phytoplankton is patchy, some species are probably overrepresented and others are underrepresented.

A comparison of the density of phytoplankton populations from the reservoir supply and the treated field during the flooding period (3 April) suggests that residence of the water in the treated field resulted in about a 50% reduction in the total population. There is a further suggestion that the reduction in green and golden-brown algae populations was primarily responsible for this difference. Possibly some reduction occurred in the yellow-green algae population. The diatoms, however, appeared to show little or no change in numbers in the treated field.

During the flushing period (6 April) the passage of the reservoir water through the treated field did not appear to alter either the total density or species composition of the phytoplankton populations. Furthermore, the data (Table 4 ) suggest that the discharge of water (6 April) from the treated field did not affect the phytoplankton population in the middle region of Halls Bayou. The population densities and species composition were similar at three stations (upstream, discharge and downstream) in Area II; and the total densities were about twice as great as those found in water entering and leaving the treated rice field.

There is some suggestion that organisms in the reservior water supply may have been affected by exposure to the treated rice field. The available data, however, suggest that discharge of water from the treated field did not affect the density and composition of the phytoplankton in Halls Bayou.

Reviewer's Evaluation

Test Procedure

Materials and Methods

A preferred method of determining phytoplankton populations is to do a plankton tow. Furthermore, the treated field and its corresponding stations in the bayou should have been compared with an untreated area in order to assess the effects of Bolero on phytoplankton.

Statistical Analysis

N/A

## Results and Discussion

Table 5 - Relative Abundances of Classes of Phytoplankton

| | Reservoir | | Treated Field | | Discharge Ditch | Halls Bayou - 6 Apr | | |
|---|---|---|---|---|---|---|---|---|
| | 3 Apr | 6 Apr | 3 Apr | 6 Apr | 6 Apr | Upst. | Disch. | Downst. |
| Bacillariophyceae | 29.4 | 9.4 | 60.3 | 9.6 | 42.6 | 29.6 | 26.2 | 11.3 |
| Chlorophyceae | 43.1 | 8.4 | 2.9 | 39.4 | 11.2 | 14.8 | 13.8 | 18.9 |
| Chrysophyceae | 13.8 | 27.1 | - | -- | | 14.3 | 19.5 | 35.6 |
| Dinophyceae | 1.3 | 14.9 | 7.4 | 7.4 | 5.6 | 5.9 | 7.6 | 7.1 |
| Myxophyceae | - | - | 17.6 | 23.4 | - | - | - | - |
| Xanthophyceae | 12.5 | 40.2 | 11.8 | 20.2 | 32.6 | 34.3 | 26.2 | 24.3 |
| Euglenophyceae | - | - | - | - | 7.9 | 1.2 | 6.7 | 3.0 |

Table 6 - Species Diversity of Each Sampling Site

| Reservoir | | Treated Field | | Discharge Ditch | Halls Bayou - 6 Apr | | |
|---|---|---|---|---|---|---|---|
| 3 Apr | 6 Apr | 3 Apr | 6 Apr | 6 Apr | Upst | Disch | Downst |
| 1.92 | 1.63 | 2.02 | 1.71 | 1.59 | 1.96 | 2.25 | 2.14 |

Calculated by Shannon-Weiner Method.

Table 5 illustrates the considerable variation in relative abundance of the several classes of phytoplankton. Even the reservoir water supply, considered to be the control site, exhibited significant variation from 3 April to 6 April. Although the report states that the densities of golden-brown (Chrysophyceae) and yellow-green (Xanthophyceae) algae were similar on both dates in the reservoir, this table indicates that they were two times to three times more abundant on the latter date.

A comparison between the reservoir and the treated field on 3 April indicates that the field had a lower total density (6800/ml) than the reservoir (16,000/ml), which was primarily attributed to fewer green and golden-brown algae. On 6 April when flushing began, the total density of the algae in the field increased 38% and that of the reservoir decreased 33%. One cause of this latter observation is probably the movement of water from the reservoir to the field. This movement may also be responsible for the related shifts in relative abundance of several classes of algae, but does not explain the increased numbers of golden-brown and yellow-green algae in the reservoir. Likewise, the 3 April observation may not be due to Bolero. If Bolero had a toxic effect on phytoplankton, the field should have had a lower density index than the reservoir. Furthermore, the data do not indicate if the differences in relative abundance of the diatoms and several other classes

of algae are due to the presence of Bolero or simply to the fact that the ricefield presents a different environment than a reservoir. The only way to tell if Bolero had any effect would be by comparing a treated field with a nearby untreated field served by the same reservoir.

The same is true regarding relative abundances, species composition and diversities for the discharge ditch and the sites in the bayou. Without data from similar untreated areas, there is no way to know if the differences between stations in the bayou are due to Bolero or to normal phytoplankton distribution patterns.

The data are inconclusive regarding the effects of Bolero on phytoplankton.

Conclusions

1. Category: Supplemental.
2. Rationale: The study provides some basic information regarding phytoplankton in a rice area, but does not aid in assessing the effects of Bolero.
3. Repairability: N/A.