

**Evaluation of Impact from Storm Water Discharges Associated with  
Road Construction Activities on Receiving Streams**

A Thesis

Presented to

the Faculty of the

Department of Civil and Environmental Engineering

University of Houston

In Partial Fulfillment

of the Requirements for the Degree

Master of Science

in Civil Engineering

by

Kriton Theodoridis

August 1998

**Evaluation of Impact from Storm Water Discharges Associated with  
Road Construction Activities on Receiving Streams**

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## **Abstract**

In order to justify the costs involved in installing and maintaining Storm Water Pollution Prevention Plans in road construction activities, a tidal tributary (Cow Bayou), that served as a final receptor of runoff from a road construction, was monitored for a period of 1-1/2 years. Physical and biological data were collected. A benthic bioassessment method and a Microtox sediment testing technique were used. Reduction in total taxa up to 50% in the downstream sampling station at the beginning of the construction suggests that the current measures provide little protection during the early stages of construction. A reduction in total taxa up to 55% in the downstream sampling station during or right after a major rainfall event suggests that these plans provide little relief from the immediate effects of rainstorm. The Microtox basic solid phase test showed little sensitivity to the type of sediments and pollution levels encountered in this research.

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## **Chapter 1 Introduction**

### *Background*

Urbanization has generated numerous impacts to our natural environment. Maybe the most profound impact of urbanization is the effect on surface waters. Site clearing and grading have removed trees that intercepted rainfall and leveled natural depressions that served as basins for temporarily storing water. These practices have resulted in increases in flooding, bank erosion, and pollutant transport to surface waters (Schueler, 1987).

Flooding has been addressed extensively during the long history of civilization whereas the issue of erosion has received little attention. The increase in urbanization over the last century resulted in an increase in erosion and pollutant transport while at the same time deterioration of surface waters in or near urbanized environments was documented. A relationship however, between erosion from human activities and water quality deterioration of nearby watersheds was not recognized until recently. In 1965, the Water Quality Act recognized the importance of stormwater runoff and authorized the federal government to provide funds in the development of projects that would control discharges that carry, among other things, stormwater (Public Law 89-234).

Perhaps the largest study in identifying the adverse effects of urban runoff was the Nationwide Urban Runoff Program (NURP) study. This study concluded that stormwater contributed to the transport of pollutants and sediments to our surface

waters. One major activity that involves heavy clearing and paving and thus contributes extensively to stormwater runoff is road construction.

In earlier days, erosion from road construction was not such a serious problem mainly because of the narrow and shallow excavations involved. Today however, highway construction involves excavation of far greater widths and much deeper disturbances making it a high risk activity for soil erosion (Israelsen et al., 1980). The damage that can occur from the transport of the sediment and its deposition in surface water can be devastating. The areas that benthic organisms occupy and fish spawn in can be lost. Suspended solids can reduce the amount of light penetration thus affecting the photosynthesis process and disturbing the whole food cycle (Barrett et al., 1995a). These are some of the many adverse effects that can be generated from uncontrollable soil erosion from disturbed sites such as the ones found in road construction activities today.

As a result the United States Environmental Protection Agency (U.S EPA) has requested all road construction activities that result in the disturbance of over one acre of land area to provide strict Storm Water Pollution Prevention Plans (SW3Ps) as a protective measure to all surface waters.

### ***Objectives***

This research project has two objectives: One is to identify whether any impact from stormwater runoff generated by road construction activities, as those are conducted today, can be detected in receiving aquatic ecosystems. The other is to examine the utility of two different approaches in detecting and quantifying this impact and comment on their effectiveness as engineering tools for future use in similar studies. The results of this study and those of other studies currently underway at the University of Houston will be incorporated in the creation of a model to predict rainfall, erosion, transport and possible impact from future construction activities.

### ***Scope and Limitations***

The findings of this research solely reflect the effects of stormwater runoff from highway construction sites into receiving watersheds. They do not apply to stormwater discharges from agricultural sources, commercial forestry, or other land-uses. Further they apply to construction activities that are covered under the NPDES permit and have SW3Ps implemented in their sites.

The aquatic ecosystem examined in this study is a small, shallow tidal tributary that flows through the heavily urbanized area of Clear Lake and is representative of the tributaries frequently found in the Texas coastal region. The substrate of this bayou has a very high silt-clay content, also representative of this region. The findings of this research therefore are more conclusive on aquatic ecosystems similar to the one



in this study with similar soil compositions, salinity contents, and mildly sloped land surroundings.

### ***Organization***

The remainder of this thesis is organized as follows:

(1) Chapter 2 contains a review of the literature associated with this research subject.

(2) Chapter 3 contains the methods used during this research.

(3) Chapter 4 contains all the results from the 1-1/2 years long monitoring study as well as results from additional laboratory experiments.

(4) Chapter 5 contains a summary of the important findings, conclusions and recommendations.

(5) The appendices contain the raw data from the field and the subsequent laboratory

analysis as well as experimental data from the Microtox basic solid phase test standardizing procedures.

## **Chapter 2 Literature Review**

In 1965, the Water Quality Act, under section 62 (Public Law 89-234), after recognizing the significance of stormwater runoff, authorized the Federal government to provide grants in an effort to assist “the development of any project which will demonstrate a new or improved method of controlling the discharge, into any water, of untreated or inadequately treated sewage or other waste from sewerage, which carry stormwater or both stormwater and sewage or other waste ...”. Although it was clear that a national awareness was being created for the state of the nation’s surface waters, the lack of research and information about the effect of nonpoint sources on the water quality of receiving water bodies led Congress to delete Federal funding for the treatment of separate stormwater discharges, in the Clean Water Act of 1977 (Public Law 95-217). There was simply not enough information to justify such an investment in physical control systems, as they were proposed at the time.

In response to this decision by Congress, the United States Environmental Protection Agency (U.S. EPA) conducted the Nationwide Urban Runoff Program (NURP) study. The purpose of this program was to provide the State and local agencies with the ability to evaluate whether urban runoff is contributing to local water quality problems and to provide the means to develop management plans and controls that would be suitable with the needs and finances of each region (U. S. EPA 1983b).

The study determined that stormwater runoff was indeed a mechanism for the transport of pollutants such as heavy metals, organics, coliforms, nutrients, oxygen demanding substances and suspended solids.

Having similar intentions as the NURP study, the Environmental Protection Division of the Georgia Department of Natural Resources, conducted a study to determine the impact of nonpoint sources on water quality and aquatic communities of targeted streams. The nonpoint source activities considered in the study, included urban development, agriculture, and commercial forestry. General water quality, organic compounds, and metals were monitored for the specific streams and compared to control streams. In addition, this study investigated the stream biology of targeted streams and monitored the surrounding land activities.

Findings on the urban development activities suggest that there was a considerable difference between all the studied parameters for the targeted streams compared to the control ones. Stormwater runoff was considered the major cause of water quality deterioration, and there was evidence that the physical effects of increased stormflow included the selective removal of organisms from their habitat. In the case of agricultural activities, it was found that macroinvertebrate diversity indices were significantly less diverse than those in the corresponding control streams. In other words the communities were generally dominated by those species that were most tolerant to increased sediment and organic loads (Georgia Department of Natural Resources, 1985).

Based on these and other studies there seems to be an agreement that stormwater runoff is responsible for transporting pollutants such as metals, organics, nutrients, and suspended solids. In addition, there seems to be a clear connection between nonpoint sources of pollution and the health of aquatic ecosystems. More specifically, reduction in diversity and selective removal of macroinvertebrate species has been recorded.

Although studies of nonpoint sources of pollution have been performed in great extent, there have been limited number of thorough studies that focused exclusively on individual activities such as road construction and their impact to receiving watersheds. Two such studies, by Reed (1976) on a Harrisburg, Pennsylvania stream and by Burton et al., (1976) on a watershed on Tallahassee, Florida, have established a direct correlation between road construction activities to increases in suspended solids of receiving streams.

However one of the first studies that examined not only the physical and chemical but also the biological impact of discharges from motorway construction in an urban environment was conducted in Essex, Great Britain (Extence, 1978). Increases in suspended solids were observed as well as severe impairment and reduction in numbers and richness of taxa in the benthic community, however no significant increases in dissolved phosphorous or nitrogen were measured. At the same time the Virginia Commonwealth University in conjunction with the Virginia Department of

Highways and Transport (Reed, 1977) conducted a similar study. The objective of that research was to establish a relationship between the silt and sediment produced by road construction activities and the macrobenthic and fish populations. Several different creeks and tributaries were examined over a period of two years. The study concluded that drift was a major physical response of the benthic macroinvertebrate community. Reductions of approximately 23% were recorded in the number of species and of up to 66% in the number of organisms. In addition, it was observed that fish vacated the areas that were heavily stressed by increased siltation, but repopulated them within 12 months from the end of the construction.

All these studies were performed before Storm Water Pollution Prevention Plans (SW3Ps) were required and when minimal erosion-control measures, not directly related to construction enhancement, were in effect. It was concluded from these studies that nutrient contributions from highway construction were negligible and that impact could be directly related to the solids flow. Such impact includes reduction in richness and numbers of the benthic macroinvertebrate population.

As a response from information provided by studies like these, Section 402(p) of the Clean Water Act, that requires stormwater discharges, associated with industrial activity, to waters of the United States to be authorized by a National Pollutant Discharge Elimination System (NPDES) general permit, was now modified by the EPA to include stormwater discharges from construction activities provided that

these activities result in disturbing over one acre of land area, as of the latest Federal Register.

In these general permits, the most important element is the development and implementation of Storm Water Pollution Prevention Plans (SW3Ps). These plans include a site description, which identifies sources of pollution, and the implementation of appropriate measures to control and reduce the pollutants. The four classes of controls that should be developed and implemented by the permittees are the following: (1) erosion and sediment controls; (2) storm water management; (3) a specified set of other controls; and (4) any applicable procedures and requirements of State and local sediment and erosion plans or management plans.

Although these plans and their goals are very detailed, there have been no provisions for monitoring their effectiveness. In addition to the lack of information pertaining to the effectiveness of these plans in reducing erosion and protecting the health of the surface waters, the cost of implementation can reach up to 2% of the total engineering costs for the individual project. In response to these facts, the Texas Department of Transportation has conducted research that would support and justify the continuation of such practices.

In one such study conducted in the Edwards aquifer recharge zone in Texas, a field monitoring program was established to monitor changes that could be attributed to highway construction activities (Barrett et al., 1995a). Significant increases of

suspended solids, turbidity, iron, and zinc were identified between upstream and downstream of the highway construction. It should be noted that SW3Ps were used as part of the NPDES permit in this project. In another study, the effectiveness of individual sediment controls was evaluated (Barret et al., 1995b). According to that study both the sediment control fences and rock filter dams surveyed, demonstrated little to no reduction in total suspended solids and turbidity.

Based on these two studies, SW3Ps appear ineffective in reducing both suspended solids and turbidity in stormwater runoff from construction activities. However there is limited work done in evaluating the effectiveness of SW3Ps in preserving the biological health of receiving streams located close to construction sites.

When undertaking such a study there is an inherent problem of selecting a way to detect and quantify the amount of impact on watersheds from stormwater runoff associated with a construction site.

In the field studies conducted in roadway construction sites prior to the use of SW3Ps, bioassessment techniques were used to assess impact by identifying changes in the aquatic ecosystems, in particularly the benthic communities. These kinds of techniques can provide information related to the extent of the impact on watersheds but cannot be used as screening tools to predict any future impact on those watersheds. Alternatives such as toxicity tests, might prove to be viable tools in detecting changes in the ecosystem, which can be attributed to construction

activities, and thus provide early warning signs on possible future impact. The need for incorporating both bioassessment techniques and toxicity tests is even recognized by the authors of the U.S.EPA Rapid Bioassessment Protocols (RBPs), who caution that such "...rapid bioassessment protocols, are best used for detecting aquatic life impairments and assessing their relative severity. Once an impairment is detected, however, additional chemical and biological (toxicity) testing is usually necessary ..." (Plafkin et al. 1989).

One such type of toxicity testing would include the direct measurement of the contaminant concentrations in the sediments of the watersheds receiving the construction runoff in order to establish the potential toxic effects of those contaminants to the stream biota. A direct measurement of the contaminant concentrations in the sediment however is not necessarily useful for predicting impact, because such a direct measurement does not reflect the bioavailable fraction of the sediment contaminants. Further, if only the concentrations for the whole sediment were measured, the potential toxic effects to organisms such as the benthic macroinvertebrates would not be properly considered, because benthic organisms feed on mostly fine grain materials (Landrum & Robbins, 1990). In this case a method for defining the fraction of the contaminants available for biological accumulation must be performed for the specific contaminants present in each site.

However this last approach is too demanding because all contaminants must be identified and their bioavailability calculated. In the 1970s a joint research program



performed by the USEPA and the US Army Corps of Engineers to regulate contaminated sediments associated with navigational dredging concluded "...that concentrations of chemicals in sediments were not reliable indicators of water quality," and that sediment toxicity should be calculated by direct measurement using toxicity tests (Lee & Lee, 1995).

Direct measurement of sediment toxicity is non-trivial even with new techniques such as the Microtox<sup>®</sup> assay, a popular alternative to earlier methods. In a pilot study at the Carolinian Province, which was part of the USEPA/NOAA Environmental Monitoring and Assessment Program (EMAP) for estuaries, the Microtox solid-phase bioassay was one component used to evaluate toxicity of sediments. The results of the pilot studies suggested that Microtox was a promising indicator of sediment toxicity in estuarine environments and led to the continuation of the use of the Microtox solid-phase assay in the fully implemented program (Ringwood et al. 1995).

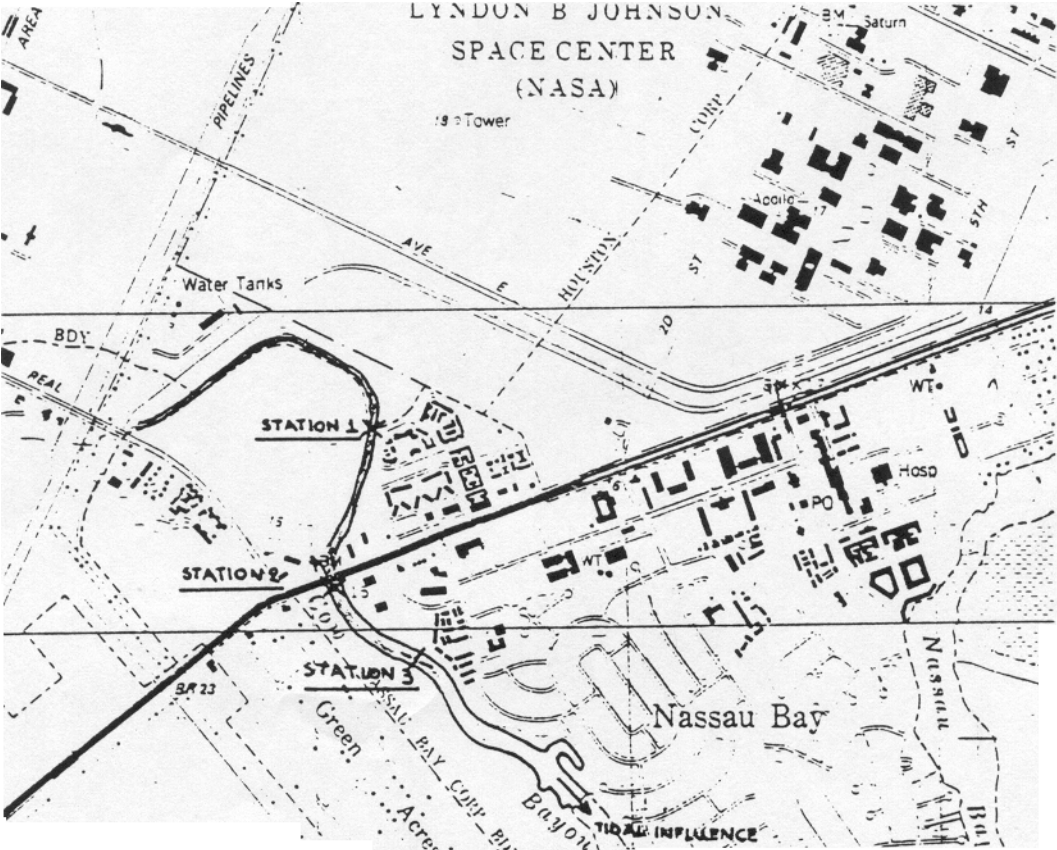
The present research attempts to answer some of the questions pertaining to the amount of impact from construction activities, as those are conducted today, to receiving watersheds. An attempt is made to identify whether any impact which can be associated to the increase in the solids flow downstream of a road construction, something which has been established on a previous study (Barrett et al. 1995a), can be detected by sampling upstream and downstream of a construction activity. Different approaches in detecting, quantifying and possibly predicting impact are

going to be utilized. Both traditional bioassessment techniques as well as Microtox sediment toxicity tests will be used. The two methods will be compared and their effectiveness as engineering tools for monitoring the health of the aquatic ecosystems which are stressed by construction activities will be evaluated.

# Chapter 3 Methodology

## General Approach

Upon agreement by the Texas Department of Transportation (TxDOT) and the University of Houston’s research team, a test site was selected at Clear Lake, Texas on NASA Road 1. The test site was a 2.368 mile construction site, with the western end located 0.36 miles east of FM 270 and its eastern end, 0.63 miles east of Space Center Blvd. A more detailed map of the general area, with the NASA Road 1 and Lyndon B. Johnson Space Center indicated, is shown in Figure 1.



**Figure 1: USGS map of the construction site and the sampling locations along Cow Bayou in Clear Lake, Texas.**

The construction site area on NASA Road 1 was approximately 52 acres and it involved a disturbance of 35 acres. Because this project disturbed more than one acre, a general permit under the NPDES system was required and all the necessary SW3Ps were placed on the site. The construction project involved the widening of NASA Road 1 and included mainly activities such as excavation, utilities relocation, grading, and paving.

The final receptors of the drainage from this project were two watersheds. There were several drainage outfalls to Clear Lake and only one to Cow Bayou. Because Cow Bayou had a single drainage input from the entire western end of the construction project, and because it was a rather small stream with no localized construction alterations present, it was selected as our target stream. Figures 2 and 3 are images of NASA Road 1 and the drainage outfall on Cow Bayou. Figure 2 is a picture taken while standing on the bridge overpass along NASA Road 1, looking west into Cow Bayou. Figure 3 is a picture taken while in Cow Bayou, looking east towards the drainage outfalls.

#### ***Selection of sampling locations and development of sampling techniques***

The initial idea of this pollution impact assessment evaluation was to monitor any changes on the health of the stream from upstream to downstream of the construction drainage outfall. Three sampling stations were selected. The first sampling station



**Figure 2: Drainage outfalls from the construction site as seen from NASA Road 1.**



**Figure 3: Drainage outfalls from the construction site as seen from Cow Bayou.**

was selected immediately downstream of the drainage outfall. This station, which was named Station 2, was anticipated to experience the highest stresses as the majority of the sediments from the construction were expected to first deposit at that location.

The next sampling station was selected further downstream of the drainage outfall and was named Station 3. Such a station was expected to demonstrate the extent of any impairment to the stream at a distance from the source and provide information on the recovery process of the bayou.

The choice of an upstream location was to serve not only as a picture of Cow Bayou that was undisturbed by the construction but also as an indicator of the overall quality of this aquatic system. The selection of such a location would have to limit the amount of uncontrolled variables that could interfere with the comparison process. The location of Station 1 (upstream sampling station) was finally selected by assuring that no other significant sources of pollution were present between it and Station 3, it had a habitat similar to the outfall and downstream stations and it was easily accessible from the street. The latter requirement was imposed because further upstream the area surrounding Cow Bayou changed from commercial to residential and it would involve special permission from the home or apartment owners to access the bayou at that point. All three sampling stations are shown on Figure 1. Stations 1 and 3 are approximately half a mile upstream and downstream of Station 2 respectively.

The first visit to Cow Bayou took place on Thursday, July 11, 1996. The main focus of this visit was to select the exact location of the sampling stations and develop proper sampling techniques. By observation it was decided that no station should be located underneath any permanent structure, such as the bridge overpass (NASA Road 1). That is why Station 2, the drainage outfall station, was located slightly downstream of the drainage outfalls, in an area not directly covered by the bridge. The reasoning behind this decision was based in the understanding that all readings with the secchi disk should be recorded under the same light conditions. Light obstruction from any permanent structure would introduce another variable and would make the light readings not comparable among sampling stations.

On this visit, light penetration readings could not be performed close to the bayou banks because of shallow water depths. Wading to the middle of the Bayou proved to be an impossible task as the analyst began sinking in the clayish substrate. Therefore incorporating wading in the regular field trips would require for one person to have a safety rope tied around his/her waist and another person to hold the rope from the shore. This idea was judged as impractical, inefficient, and even dangerous. For these reasons a floating device (canoe) was used in all future trips.

It was decided during the first visit that all water quality sampling and in situ water measurements would have to be conducted at a precise location in the middle of the bayou for each station, using certain manmade structures as reference points. The

column support for a pipe crossing and a bridge column were selected as our references for Stations 1 and 2, respectively. The two reference locations are shown on Figures 4 and 5. Figure 4 shows a picture of the column support used as our sampling reference for Station 1 and the overhanging pipe above it, while Figure 5 depicts a picture of water quality sampling taking place along the bridge column used as our reference for Station 2. A water mark in the middle of Cow Bayou further downstream was selected as our reference for Station 3. This water mark is shown on Figure 6.

On this first visit, a LaMotte stainless steel bottom sampling dredge was tested for future benthic macroinvertebrate collection. The sampler was designed to settle in the substrate and then be removed. When this technique was used in the field, the sampler recovered a minimal amount of material. The instrument failure was blamed on the cohesive nature of the soil. In order to counteract the cohesive soil's resistance, the sampler was inserted deeper in the substrate by exerting a force downwards into the substrate. By doing so, a much greater volume of bottom sediment was recovered. The obtained sediment sample was then sieved through a No. 35 sieve (0.41 mm) while at the field and all materials retained on the sieve were collected and properly stored. A subsequent laboratory analysis however showed that this technique was not suitable for the collection of benthic macroinvertebrates as most of the sample consisted of decomposed leaves, plant seeds and small twigs. The





**Figure 4: Column support used as reference for all water measurements at Station 1.**



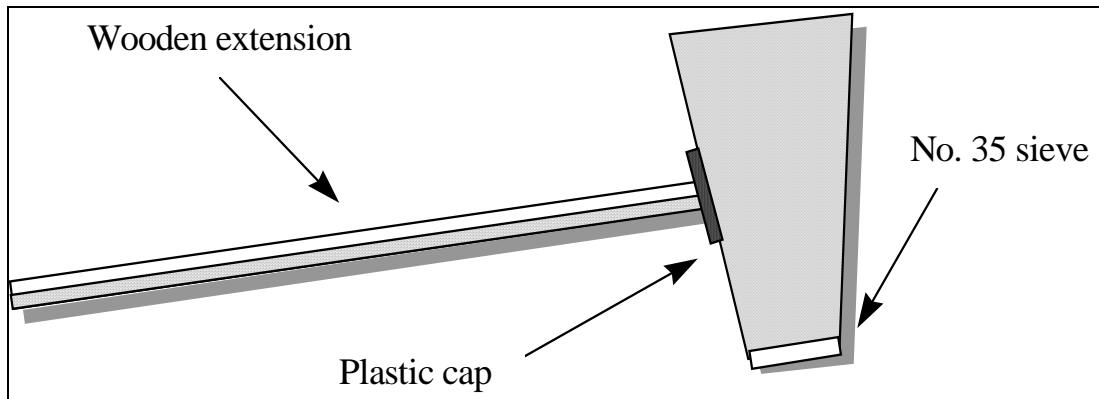
**Figure 5: Bridge column used as reference for all water measurements at Station 2.**



**Figure 6: Water mark used as reference for all water measurements at Station 3.**

act of forcing the sampler into the substrate appeared to result in the collection of sample that did not belong in the benthic category.

In order to collect benthic sample, a new device was incorporated in our next visit to accommodate for the substrate conditions present at Cow Bayou. This device is shown graphically on Figure 7. A long wooden stick was attached to a plastic cap which was opened faced at one side and closed by a No. 35 sieve (0.41 mm) on the other side. This design allowed the operator to reach in small water depths and scoop



**Figure 7: A graphical presentation of the apparatus used for benthic macroinvertebrate collection at Cow Bayou.**

away the surface of the substrate. The collected material would then have to be sieved on the site, emptied into a container and returned to the lab for further

analysis. This technique would prove to be very successful as a great number of benthic macroinvertebrates was collected during the first try and unwanted material was reduced considerably.

### ***Field trip procedures***

Regular sampling began on August of 1996 and finished on December of 1997, almost a year and a half later. A canoe was used for both shallow and mid stream measurements and sampling. The field trips were performed according to the analysts' schedules (once or twice every month) but also immediately after certain large rainfall events. The post-storm visits were conducted to identify any immediate effects on the bayou from the stormwater runoff.

Two people had to be physically present at every field trip. The canoe was loaded on the roof of a University of Houston's van and properly tied down. All the field instruments, samplers, containers and other apparel were then loaded in the van.

The instruments and containers used were the following:

- (1) A Portable Hach One pH Meter, Model 43800-00 by Hach company, was used for measurements of pH and temperature
- (2) A Conductivity / TDS (Total Dissolved Solids) Meter, Model 44600 by Hach company, was used for measurement of conductivity and total dissolved solids
- (3) A Dissolved Oxygen Meter, Model 50B by YSI Incorporated, was used for measurement of dissolved oxygen concentrations
- (4) A self made Secchi Disk was used for measurement of light penetration

- (5) A self made stream depth measure was used to make mid stream depth readings
- (6) Three polypropylene Clear Boxes with a volume of 4.4 L, by Rubbermaid<sup>®</sup>, were used for the storage of the benthic macroinvertebrate sample from each station
- (7) Three polypropylene containers with a volume of 0.95 L, by Anchor Hocking, were used for the storage of soil samples from each station for later analysis using Microtox<sup>®</sup>
- (8) Three 500 mL widemouth amber sample jars were used for storing water sample for Total Suspended Solids (TSS) analysis in the lab

In addition to all the instruments and containers mentioned above the following miscellaneous items were carried along at every field trip: Two paddles, two life jackets and two coolers.

After loading was completed, the team proceeded to drive to Cow Bayou. A suitable area behind a commercial establishment was used for unloading the canoe upon arrival to the bayou. This area was located between the upstream station, Station 1 and the outfall station, Station 2. The canoe was then inserted in the bayou and all the equipment loaded in. A picture showing the unloading of the canoe from the van is presented in Figure 8, while the exact area where the canoe was inserted in Cow Bayou is shown in Figure 9.

Once in the stream, the team paddled upstream to the reference location of Station 1. The canoe was tied around the column support to allow the operators to work freely in taking measurements and samples. One operator was making the measurements while the other was recording the readings on the field data sheet. A copy of this field data sheet is shown in Figure 10.

Most of the important physical and habitat parameters, as those are defined in the U.S.EPA Rapid Bioassessment Protocols (RBPs), appear in this field data sheet. Measurements of pH, temperature, dissolved oxygen, conductivity, total dissolved solids, light penetration, stream depth, and high water mark were made using the



**Figure 8: Unloading of the canoe at the field site.**



**Figure 9: Area where canoe was inserted in Cow Bayou.**

STATION No. \_\_\_\_\_ Date: \_\_\_\_ / \_\_\_\_ / \_\_\_\_.

Estimated Stream Depth:

Estimated Stream Width:

High Water Mark:

Light Penetration:

pH:

Water Temperature:

Dissolved Oxygen:

Conductivity:

TDS:

\_\_\_\_\_.

Water Odors:

Sediment Odors:

---

**Figure 10: Field data sheet used in all field trips at Cow Bayou**

instruments mentioned before, while the stream width, sediment, and water odors were field estimated. Once the field data sheet was completed for Station 1, a water sample was collected in the 500 mL amber sample jar and then the operators proceeded to move directly to the western bank of the bayou. At that location, a previously established 5 ft x 5 ft sampling area was used for benthic macroinvertebrate sampling and soil sample collection.

Such sampling areas were created for all three stations by inserting four wooden sticks in the bayou substrate. One such sampling area can be seen to the left of the water mark, used as a reference for Station 3, in Figure 6. The use of a defined sampling area for benthic macroinvertebrates is strongly recommended in most bioassessment techniques.

Once all samples and readings were completed for Station 1, the operators went on to repeat the same procedure at Stations 2 and 3 further downstream and then returned to the launching area. The time spent in the water decreased as the operators became more familiar with the canoeing and sampling techniques. At the end of this research project, the time spent in the water was approximately 2 hours.



To preserve the samples, ice was purchased and placed in the cooler containing all the samples. Upon arrival at the University, all sample containers were unloaded and placed in a refrigerator at 4°C. By the end of this research study the entire time spent on a field trip was reduced to approximately 3 hours and 30 minutes.

Once all the samples were stored in the laboratory, a Suspended Solids (SS) analysis was performed on all water samples. The benthic macroinvertebrate analysis on the sieved benthic sample was usually the second to follow with Microtox analysis on the soil samples performed last.

### *Suspended Solids Analysis*

The Suspended Solids (SS) analysis was performed within 48 hours from initial storage. The water sample was removed from the refrigerator and 500 mL were poured in a commercial blender. The sample was mixed at a high speed for exactly two minutes and then poured in a calibrated beaker of greater volume than the sample itself. The sample was manually stirred in the beaker and immediately 25 mL were poured on a sample cell provided by the Hach Company.

A Hach DR/2000 spectrophotometer was used for the analysis. In using this instrument, the sample should be compared to a blank sample. The spectrophotometer analyzed the amount of light of a predetermined wavelength that passed through the blank sample and would calibrate that amount to zero. The

instrument would then read the light passing through the actual sample and using calibration curves built in the spectrophotometer was able to provide a value for the sample itself.

The program number entered in the Hach DR/2000 spectrophotometer for the calculation of Suspended Solids was 6-3-0 and the wavelength that was measured was 810 nm. The blank sample was simply 25 mL of deionized water (DI water) poured in a sample cell. All Suspended Solids analysis results were recorded in a laboratory notebook in units of mg/L.

#### ***Benthic Macroinvertebrate Analysis***

Benthic macroinvertebrate samples are usually stored in some type of ethanol or formaldehyde mixtures to ensure preservation of the sample. In this study no preservative mixtures were used. The organisms were stored and analyzed while still alive.

The reason why this approach was selected and followed in all analyses thereafter, had to do with the initial inexperience of the researcher in benthic macroinvertebrate identification. The researcher's selection to observe and record the benthic species while alive, was based at the observation that the individuals of certain species, exhibited identical behavior when alive and also retained their natural coloration. This property made recognition of organisms of the same species much easier.

At the beginning of this analysis, a small subsample was scooped from the sample container and placed into a standard petri dish. Deionized water (DI water) was added to fill up the bottom surface of the dish. The subsample was uniformly spread over the dish, to ensure that approximately the same amount of material would be observed at every point in the dish. Such a uniform distribution of sample material was a means of standardizing the whole procedure.

The petri dish was then placed over a transparency with enumerated square grids, each 1 cm x 1 cm wide, enveloped by a subscribed circle, the size of the dish. The circle in the transparency was separated into four quadrants. Both the transparency and the petri dish containing the sample were then placed under an SZ40 model Olympus microscope.

The researcher would then proceed to look at the enumerated square grids (starting from grid 1,2,3... etc.). Whenever a benthic macroinvertebrate was found, it was picked up using forceps and removed to another petri dish, also covered by DI water. Each time this procedure was done, the type of organism (by an assigned number) and the quadrant that it was found in were recorded in a laboratory notebook. Whenever a quadrant was completed the researcher would then move to the next one, until close to 100 macroinvertebrates were recovered.

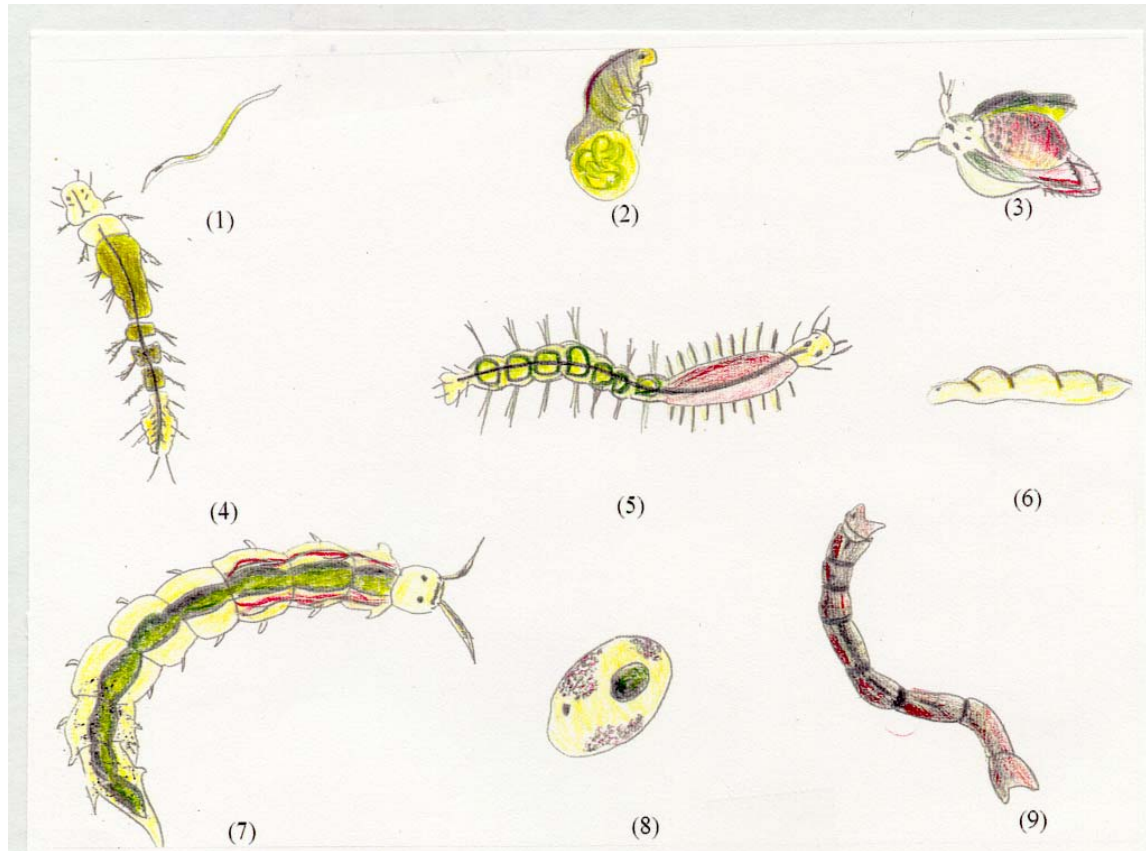
If all four quadrants within the subscribed circle were examined and 100 organisms could not be recovered, the subsample was wasted and another scoop was placed in its place. The same procedure was then repeated making sure that the introduction of the new subsample was recorded. If 100 organisms were recovered, the researcher would proceed to finish the enumeration of the quadrant were the 100th organism was found in and complete the analysis.

At the beginning of this research project, benthic macroinvertebrate analysis was very time consuming. Approximately 9 hours were required for the operator to enumerate 100 organisms from a single sample. The researcher would have to carefully sketch every new organism encountered in the sample and assign a number. Therefore during the analysis of samples from the first field trips, a considerable amount of time was spent in this characterization (sketching) procedure. Figure 11 shows the characterization procedure that was involved in the initial stage of this study. Different benthic macroinvertebrates, numbered 1 through 9 are depicted through sketches created by the researcher.

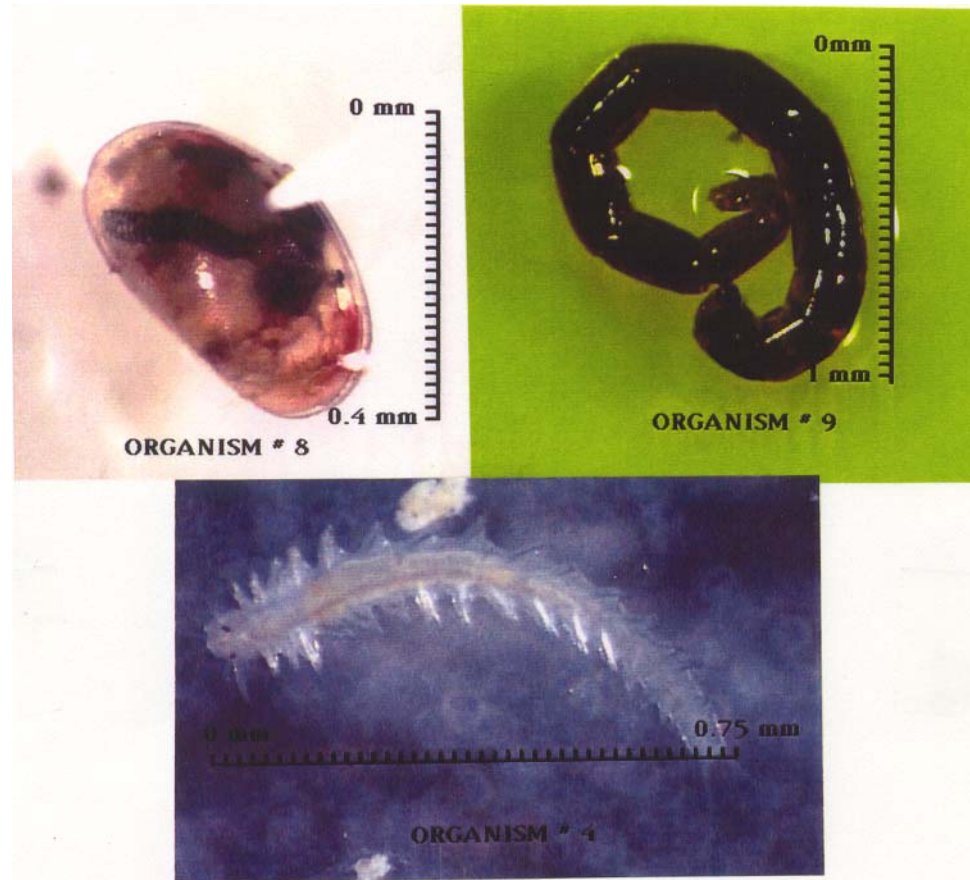
Further along in this research, the operator became increasingly more knowledgeable and was able to recall easier the species' numbers assigned in previous analyses. Additionally, fewer new species were found as the research progressed. Those species found early on this project (and assigned a small number) were the dominant species for Cow Bayou, whereas the ones found at the later parts of this research

(and assigned a large number) would be species rarely found again during this project.

A SONY CCD\_IRIS/RGB color video camera, connected to a SONY camera adaptor, a SONY HR-TRINITRON PVM1353MD model color video monitor, a VAS archiving system and a SONY UP-5600MD model color video printer, was introduced in the middle of this research project. This equipment eliminated the time spent sketching new species. Most of the species were now photographed, given the same number as before and then stored in a large database. Figure 12 illustrates the type of photographs taken using this system by showing some representative species. It is interesting to compare some of the photographs with the initial sketches shown in Figure 11. In doing so one might see that the idea of first sketching the organisms and assigning them a number, was a good one, as the sketches seem to be a good representation of the actual organisms.



**Figure 11: Sketches and assigned numbers for the first nine species found during benthic macroinvertebrate analysis**



**Figure 12: Photographs of representative species, taken using the SONY system during benthic macroinvertebrate analysis.**

All of the improvements mentioned above, contributed towards a considerable reduction in time spent during analysis. One sample was now completed within 3 hours instead of 9 hours. This reduction in analysis time had an effect on the storage period for the benthic samples. The maximum time the samples were kept in the refrigerator was reduced from approximately a week (during the beginning of this study) to about 2-3 days.

### ***Microtox Analysis***

The last of the analyses performed in the laboratory after each field trip was the Microtox analysis on the soil samples. The Microtox assays were done in accordance to the basic solid phase test protocols, as those are established by Azur Environmental, using a Microtox 500 analyzer. The materials, reagents, diluents and reconstitution solutions were all purchased from Azur Environmental, Carlsbad, California.

The reagent used is composed of naturally occurring *Photobacterium phosphoreum*, a non pathogenic marine organism. The metabolic process of this bacterium is intrinsically tied to its respiration and the end product of its respiration includes the production of light. The Microtox 500 analyzer is able to detect changes in the light outputs that can be attributed to the introduction of a toxicant in the system. By testing a series of nine dilutions from the sample, the analyzer is able to produce a value of effective concentration which reduces light production by the *Photobacterium phosphoreum* by 50 % relative to controls used (EC50).



When preparing the sample for the basic solid phase test, the soil should be first thoroughly mixed to ensure that both soil particles and toxicants are homogenized throughout the sample. A manual mixing of about 10 minutes is recommended. Then a slurry of soil and solid phase diluent is created to run for Microtox. The highest concentration of soil sample in the analyzer, after all procedures are followed should be 0.099 mg/mL. However such a concentration for certain soil samples might be too lethal for the bacteria. In that case, a reduction of the highest concentration should be made to provide reasonable results. The Microtox analyzer's software is designed to accommodate changes of initial concentrations. A number of initial dilutions ratios were run for the soil samples from Cow Bayou and it was found that an initial dilution of 1/1.5 over the recommended concentration or in other words an initial concentration of 0.066 mg/mL was suitable for most samples in this analysis.

When performing most of the toxicity assays using the Microtox 500 analyzer, reference samples are highly recommended. The reference sample should have similar particle size, organic material, moisture content, turbidity and color as that of the sample under investigation. By selecting a reference sample, natural toxicity is distinguished from human induced toxicity. However an approach of how to select reference samples is not provided in the training manuals for Microtox. It is therefore left to the individual researchers to select a way of providing a baseline toxicity for their samples. Samples from Station 1 were selected as our reference

samples for every field trip. These samples were not intended to separate natural toxicity from human induced toxicity. They were intended to provide a baseline for separating any human induced toxicity taking place at any point beyond the upstream station, from the toxicity introduced at any point before the upstream station. All Microtox runs with the soil samples from Cow Bayou were performed in triplicates in this study.

It was also assumed that the organic material, moisture content and color of the samples from all three stations were similar to each other. However particle size was initially expected to be a variable among stations. In addition the use of different initial dilution for our samples raised a concern that the turbidity of the samples was now altered. Therefore a method was developed to provide an interpretive strategy of examining the results from Microtox by using particle size and initial dilution of each sample for correction. The fact that most of the soil samples were not analyzed until a week after collection led to development of a method of evaluating the effect of storage period to measurable toxicity by Microtox.

### ***Particle Size and Initial Dilution Experiments***

The development of a method to demonstrate the effects of particle size and initial dilution in the EC50 values produced by the Microtox basic solid phase test is described in this section. It was initially hypothesized that particle size has an effect on the effective concentration values (EC50) produced by the Microtox test. A set

of samples, with identical natural toxicity but different particle sizes was run through the Microtox analyzer to support this hypothesis.

The soil sample used for this experiment was collected at the Clear Lake area. This was not soil from Cow Bayou but rather regular top soil. Soil from Cow Bayou was not used in order to minimize the presence of excess organic content and the effects of coloration. Particle sizes however, were similar to those present in Cow Bayou. The sample was oven dried and a dry sieve analysis was performed on a Tyler portable sieve shaker. Intermediate sieves (No. 50 and No. 100) with greater aperture sizes than 75  $\mu\text{m}$  were used to retain the flaky soil particles that were created when the sample was oven dried, while the No. 200 sieve was used as the bottom sieve in the stack. The soil retained at the No. 200 sieve was collected in a widemouth jar and properly labeled. The same was done for the soil that passed the No. 200 sieve and collected at the pan. The 75  $\mu\text{m}$  particle diameter is considered by most classification systems as the separation point between clay-silt and sand. Based on this fact and the findings from the Carolinian Province study, where the presence of clay-silt in the soil samples had a considerable effect on the EC50 values (Ringwood et al. 1997), the 75  $\mu\text{m}$  diameter was selected as the criteria in separating small particles from larger ones.

A series of mixtures for the two samples was generated. These mixtures ranged from 0% by weight for the soil passing the No. 200 sieve to 100%, by increments of 10%. All these soil mixtures were then run through a Malvern Instruments

Mastersizer to further check the effectiveness of dry sieving. From the results produced by the Mastersizer, the 0%, 10%, 50%, 70% and 100% mixtures were selected as a good distribution of particle sizes to demonstrate any effect of particle size on the Microtox EC50 values.

These soil mixtures were expected to provide information on the effect of particle size on EC50 values. However the effect of cloudiness (turbidity) on the bioassay readings could not be accomplished solely on the results of this experiment. Therefore dilutions, such as the ones performed in the samples from Cow Bayou, were performed for the five different mixtures selected for Microtox analysis. By doing so, the particle sizes would remain the same while turbidity was expected to change because of the dilution process. The same dilution as the one used when running the Microtox analysis on the soil samples from Cow Bayou (1/1.5 initial dilution) as well as 1/3 and zero initial dilution were selected. All fifteen samples (five mixtures of three different dilutions) were first run through the Microtox analyzer using the basic solid phase test described in the Microtox analysis section of this chapter. All Microtox runs using the previously mentioned soil mixtures, were performed in duplicates in this study because the necessary mixture volumes were hard to predict and the sample preparation was extremely time consuming.

### ***Storage Time Experiments***

Because of the delay in the analysis of many of the soil samples collected during this study using the Microtox basic solid-phase test, an experimental procedure was

developed to assess the effects of prolonged storage of soil samples on the Microtox's EC50 values. A soil sample collected from Cow Bayou was first homogenized by a procedure described earlier in the Microtox Analysis section and then separated in five different glass containers. Four of the five containers were tightly sealed and then stored at a constant 4°C temperature. The sample in the fifth container was tested the same day of collection in triplicates using the Microtox analyzer. The rest of the samples were analyzed subsequently. Test runs were performed the day after, 1 week, 2 weeks and 1 month following the date of collection. All Microtox runs for this experiment were done in triplicates.

## **Chapter 4 Discussion of Results**

### *Water Quality Summary*

Light penetration was measured in the field by use of a Secchi disk. The water in Cow Bayou has a natural high turbidity which resulted in measurements of small light extinction depths. Light penetration recorded in the field ranged from a low value of 0.75 ft to a high value of 1.92 ft. The pH measurements came very close to what was expected for a natural stream such as Cow Bayou. The highest pH value recorded was 8.65 whereas the lowest recorded value was 5. The average value of pH for the entire study was about 7.

As mentioned in the Methodology chapter dissolved oxygen was recorded at every field trip. In some of the field trips however the measured dissolved oxygen approached the theoretical saturation value. In cases where saturation levels of dissolved oxygen were measured the instrument was recalibrated prior to the next sampling date. Dissolved oxygen ranged in magnitude between 2.71 mg/L and 9.17 mg/L.

Conductivity was also measured in the field in order to monitor for changes in salinity. Since Cow Bayou was an estuarine environment, salinity was expected to fluctuate. Low salinity values were usually recorded with the exception of the December 18, 1996 and the September 15, 1997 field trips. In September 15, 1997

salinity appeared at its highest, measuring 11.9 ppt whereas the lowest value recorded was 0.1 ppt.

Another important physical parameter measured in the field was that of water temperature. Water temperature is essential in identifying seasonal effects on the benthic macroinvertebrate community. Water temperature was the lowest on December 18, 1996 measuring just 8 °C. On the summer months however water temperature was considerably increased reaching up to 32 °C on July 31, 1997.

### ***Bioassessment Results***

The benthic samples were evaluated using two different metrics: Total taxa and diversity. These metrics were selected upon the advice of the Texas Natural Resource Conservation Commission (TNRCC) field operations division. These exact metrics were used by TNRCC in the assessment of Patrick Bayou, another estuarine environment in the same geographical region as Cow Bayou. Total polychaete taxa and Sheldon's evenness index were also calculated and the results appear in the Appendix section of this thesis.

Total taxa is a biotic indices. It reflects the health of the community through a measurement of the variety of taxa present. In general it increases with increasing water quality. Total taxa is defined as the total number of species present at each

sampling station. The organisms of the same species are grouped together and the total number of species is calculated.

A diversity index indicates the state of the community by combining abundance and richness. According to Norris and Georges (1993) diversity indices “... decrease with decreasing water quality. Also low diversity supposedly indicates a stressed community that tends to be unstable. The most widely used diversity indices are those derived from information theory such as the Shannon-Wiener (H') index”.

When calculating diversity in this study, the Shannon - Wiener index (H') was used as it was done in the Patrick Bayou assessment study. This metric as shown by Shannon (1948) and Wiener (1948) is calculated as follows:

$$\text{Shannon - Wiener (H') index} = - \sum_{i=1}^s (p_i)(\log p_i), \quad (1)$$

where  $s$  is the number of different taxa, and

$p$  is the proportion of total in the  $i$ th taxa.

A maximum value is reached for this index when all the species are distributed evenly. The theoretical maximum of H' is LOG ( $s$ ). Biologically this is thought to be the most desirable situation (Norris and Georges, 1993). Because this index of heterogeneity is based upon information theory, larger information content equals



greater uncertainty. Therefore a community with only 1 taxa has no uncertainty in it and  $H' = 0$ . According to what base one uses for the logarithmic calculation, the units of  $H'$  will vary. When the base is 2, e, and 10, the units of  $H'$  are “bits”, “nits”, and “decits” respectively.

In this study when calculating diversity, the natural logarithm was used as the logarithmic base and diversity index appears in values of “nits”. Tables 1, 2 and 3 show the calculated results for both metrics for each sampling station respectively.

The standard deviation as well as the mean of all field trips, with the exception of the November 8, 1996 field trip, is calculated for every sampling station for both metrics. There were no reportable data for the November 8, 1996 field trip. The standard deviation is calculated as follows:

$$\text{standard deviation} = \left[ \sum (x - x_{\text{mean}})^2 / (n-1) \right]^{1/2}, \quad (2)$$

where  $n$  is the number of measurements,

$x$  is the value of any measurement, and

$x_{\text{mean}}$  is the mean value of all the measurements.

**Table 1: Summary of the two metrics calculated at Station 1**

Sampling Date	Total Taxa	Diversity
Aug. 30, 1996	8	0.8
Sept. 10, 1996	8	0.9
Oct. 4, 1996	9	1.1
Nov. 8, 1996	No data	No data
Dec. 18, 1996	5	1.4
Jan. 24, 1997	6	0.9
Feb. 14, 1997	9	1.4
March 7, 1997	8	1.3
March 19, 1997	11	1.9
April 18, 1997	8	0.9
May 13, 1997	9	1.4
June 6, 1997	7	1.3
July 10, 1997	10	0.9
July 31, 1997	11	1.7
Sept. 5, 1997	8	1.3
Oct. 16, 1997	9	1.7
Dec. 5, 1997	7	1.4
<b>Mean</b>	<b>8.3</b>	<b>1.3</b>
<b>Standard Deviation</b>	<b>1.6</b>	<b>0.3</b>

**Table 2: Summary of the two metrics calculated at Station 2**

Sampling Date	Total Taxa	Diversity
Aug. 30, 1996	6	0.46
Sept. 10, 1996	5	0.82
Oct. 4, 1996	7	1.15
Nov. 8, 1996	No data	No data
Dec. 18, 1996	9	1.69
Jan. 24, 1997	8	1.36
Feb. 14, 1997	7	1.05
March 7, 1997	6	1.29
March 19, 1997	8	1.46
April 18, 1997	10	1.57
May 13, 1997	12	1.67
June 6, 1997	10	1.83
July 10, 1997	8	1.07
July 31, 1997	7	1.38
Sept. 5, 1997	8	1.43
Oct. 16, 1997	6	0.87
Dec. 5, 1997	10	1.03
<b>Mean</b>	<b>7.9</b>	<b>1.3</b>
<b>Standard Deviation</b>	<b>1.8</b>	<b>0.4</b>

**Table 3: Summary of the two metrics calculated at Station 3**

Sampling Date	Total Taxa	Diversity
Aug. 30, 1996	4	0.56
Sept. 10, 1996	6	0.64
Oct. 4, 1996	6	0.91
Nov. 8, 1996	No data	No data
Dec. 18, 1996	9	1.42
Jan. 24, 1997	11	1.63
Feb. 14, 1997	4	1.19
March 7, 1997	7	1.49
March 19, 1997	9	1.69
April 18, 1997	14	1.95
May 13, 1997	9	1.79
June 6, 1997	9	1.85
July 10, 1997	7	1.41
July 31, 1997	11	1.93
Sept. 5, 1997	7	0.99
Oct. 16, 1997	7	1.04
Dec. 5, 1997	10	1.76
<b>Mean</b>	<b>8.1</b>	<b>1.4</b>
<b>Standard Deviation</b>	<b>2.6</b>	<b>0.4</b>

Natural variability is frequently encountered in single biological indicators such as total taxa and diversity. In such a case knowledge of the variability of repeated measurements is needed. Otherwise it is ambiguous whether any measured difference in the metrics of two sampling stations is natural variability between the two stations as mentioned before or real difference in the benthic communities. Because of the initial inexperience of the researcher with sampling theory for biological data, replicates samples of the same station at the same time were not collected.

To compensate for the lack of replicates in our sampling procedure, samples collected from Station 1 will be used as a guidance for identifying any natural

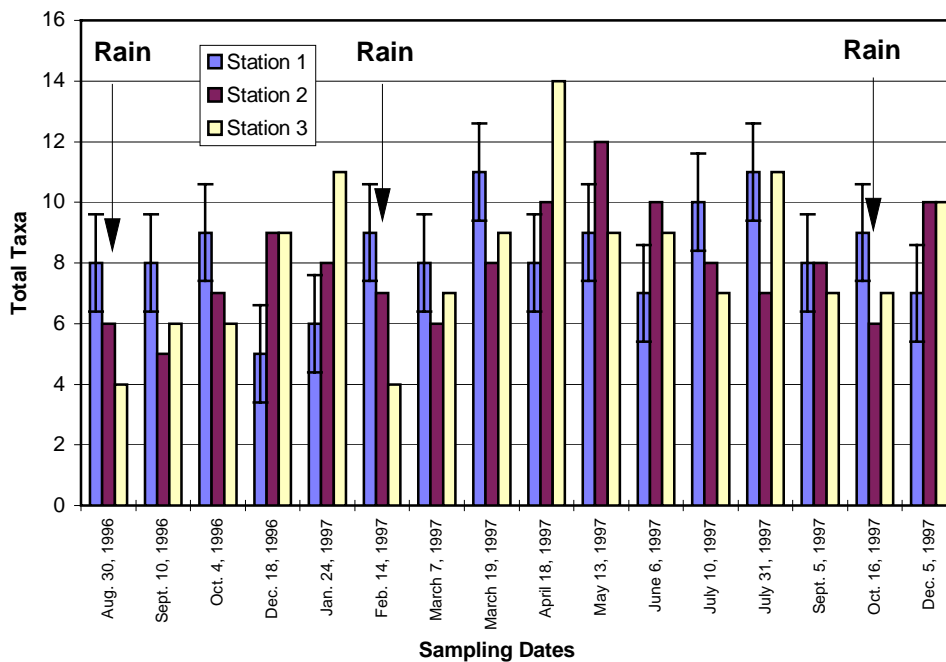
variability in the benthic community downstream of the construction. For a real difference in the downstream benthic community to exist when comparing to the upstream one, any metric value from the downstream benthic community will have to be greater or smaller than the corresponding value of the metric on the upstream station plus or minus the calculated standard deviation for Station 1 respectively.

A total of 54 different organisms were found during this study. Upon consulting with biologist Dr. Cynthia L. Howard from the biological sciences department at the University of Houston, Clear Lake campus, two organisms (organism No. 21 and No. 41) were excluded from the analysis because they were not benthic. At any field trip the maximum number of benthic macroinvertebrates encountered was 14 and therefore the calculated metrics on each day are based on a number of organisms less or equal to 14. The most commonly found benthic macroinvertebrates in this study appear at the Appendix section of this thesis.

A better visualization of possible trends of the benthic community that might indicate worsening or improvement with time from upstream to downstream of the construction site can be achieved by graphically representing the indices at each sampling station for all sampling dates.

Figure 13 displays the value of total taxa calculated at each of the three stations during this study. There are three graphical bars present for every sampling date.

All field trips are shown in the x axis of the graph. The height of the left bar in any sampling day indicates the value recorded for the total taxa at Station 1 for that date. The standard deviation from the analysis performed on Station 1 for all field trips is shown as error bars. The height of the middle bar represents the value of total taxa for Station 2 while the height of the right bar in each sampling date is the corresponding value of total taxa at Station 3. The arrows that appear on the August 30, 1996, February 14, 1997 and October 16, 1997 field trips indicate that sampling took place during or right after a major rainfall event. As mentioned in the literature review chapter there is an interest on identifying the immediate effects of rainfall on the benthic communities downstream of a construction activity.

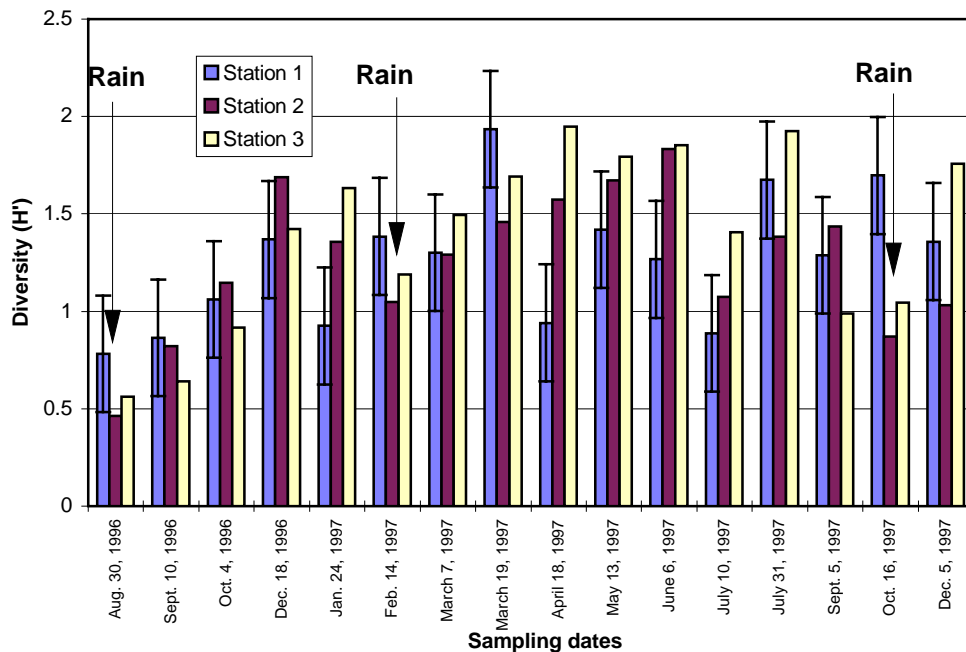


**Figure 13: Total taxa at every station**

The data in Figure 13 indicates that for the first three field trips a detectable difference in total taxa between the upstream and downstream of the construction activity exists. The downstream station (Station 3) exhibits a reduction in total taxa in the magnitude of 25% to 50% when compared to the upstream sampling station (Station 1) in the first three field trips. This difference appears to be significant and not simply natural variability of the benthic macroinvertebrate data. After the third field trip however this pattern fluctuates. On samples from the December 18, 1996 field trip for example, the downstream station's benthic community appears to have a greater number of taxa than the upstream station's community. On a later field trip, like the one on February 14, 1997, this trend is reversed again.

In Figure 14 the same graphical format as in the Figure 13 is used. There are three graphical bars present for every sampling date. The height of each bar in any sampling day indicates the value recorded for diversity ( $H'$ ) at each sampling station for that date. The height of the bar on the left represents the value of diversity ( $H'$ ) calculated for Station 1 whereas the height of the middle and right bar represent the value of the index for Stations 2 and 3 respectively. The standard deviation from the analysis performed on Station 1 for all field trips is shown as error bars. The arrows shown in Figure 14 point to dates when sampling took place during or right after a major rainfall event.

The diversity ( $H'$ ) data in Figure 14 indicate that for the first three field trips a difference between the upstream and downstream of the construction activity exists. However the value of diversity ( $H'$ ) for all three field trips falls within the standard deviation observed in the upstream station which leads the author to believe that this is not a significant change between the two stations but merely a natural variability frequently encountered in biological data. After the third field trip however this pattern is again absent.



**Figure 14: Diversity ( $H'$ ) at every station**

Regression analysis was performed to determine the relationship of the calculated metrics and the different water quality parameters collected in the field. Total taxa

and diversity ( $H'$ ) for every field trip and every sampling station was plotted against the values of water temperature, light extinction depth, pH, and dissolved oxygen recorded for those same dates and sampling stations. For each of these plots linear regressions were run and the percentage of the variability in the metrics ( $R^2$ ) that can be expressed in each of the water quality parameters was calculated. In Tables 4 and 5 the  $R^2$  values are presented for the series of linear regressions performed for total taxa and diversity ( $H'$ ) respectively. The value of  $R^2$  is calculated as follows:

$$R^2 = 1 - \left[ \frac{\sum (y - y_r)^2}{\sum (y - y_{\text{mean}})^2} \right], \quad (3)$$

where  $y$  is the variable in the y axis,

$y_r$  is an estimate of  $y$  for any given  $x$ , and

$y_{\text{mean}}$  is the mean value of all  $y$  points.

**Table 4.  $R^2$  values for regressions performed for total taxa with water quality parameters**

Total Taxa	Temperature	Light Penetration	pH	Dissolved Oxygen
Station 1	0.1616	0.1074	0.0654	0.2877
Station 2	0.0083	0.0001	0.0778	0.7187
Station 3	0.0026	0.1339	0.061	0.0727



**Table 5. R<sup>2</sup> values for regressions performed for diversity (H') with water quality parameters**

Diversity (H')	Temperature	Light Penetration	pH	Dissolved Oxygen
Station 1	0.0828	0.002	0.0381	0.0796
Station 2	0.0084	0.0647	0.1971	0.5486
Station 3	0.0059	0.0275	0.0642	0.2769

The coefficient of linear correlation (R<sup>2</sup>) is practically zero for all parameters and all sampling stations, which indicates that there is no correlation between each one of these parameters and any of the metrics. The only exception to this observation appears to be the relation of dissolved oxygen with both metrics for Station 2. Whenever dissolved oxygen increases at Station 2, total taxa and diversity increase and the coefficient of linear correlation appears significantly increased comparing to the rest of the observations.

In this study, there is interest in recognizing and possibly quantifying any immediate effects of rainfall on the benthic community located downstream of a construction site. In Figure 13, the sampling dates with a recorded rainfall event during or prior to a sample collection are clearly marked with an arrow. For those dates total taxa is reduced for the benthic community located downstream of the construction activity when compared to the upstream community. Total taxa is only 22% lower in the downstream station compared to the upstream one for the October 16, 1997 field trip. However, a significant 55% reduction is observed for the February 14, 1997 field trip.

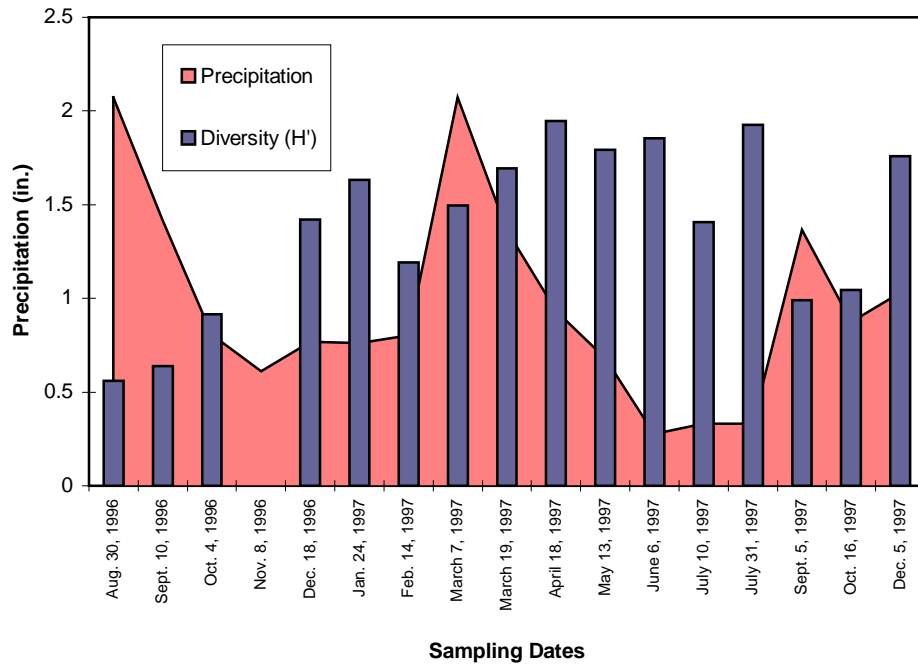
Recognizable immediate effects of rainfall on the benthic community downstream of the construction site are not observed when looking at the diversity index ( $H'$ ). In Figure 14, for those sampling dates with a recorded rainfall event during or prior to sample collection, there is no consistent pattern of reduction of diversity ( $H'$ ) in the downstream sampling station. The only considerable reduction in diversity ( $H'$ ) occurs at the October 16, 1997 field trip where diversity ( $H'$ ) appears 39% lower in the downstream station compared to the upstream one.

Although looking at these three individual rainfall events might shed some light on the immediate effects of rainfall, a precise relation between precipitation and the health of the benthic community directly downstream of a construction site cannot be determined solely on these observations. The development of such a relation would require very frequent monitoring of the stream with simultaneous daily precipitation measurements at the test site. Because of the time and labor constraints in this project such elaborate sampling was not performed. In addition there was no site specific rainfall data collected in any of the field trips.

In an attempt to determine if a relation between precipitation and health of the benthic community downstream of the construction site is detectable, historical rainfall data were analyzed.

Total monthly precipitation amounts from Houston Intercontinental and Galveston were obtained from the National Climatic Data Center (NCDC). Because recorded precipitation readings for the area close to Cow Bayou could not be found, the arithmetic mean for the two stations was used as the estimate of precipitation at Cow Bayou. Cow Bayou is located approximately halfway between the two rainfall gages and using the distance weighted mean is a common practice in hydrology for estimating rainfall at missing stations. The monthly total precipitation was then plotted with each metric calculated during the monthly collections from the downstream station. The goal here is not to provide a quantifiable relation between rainfall and the benthic community but rather to find any consistent pattern between the two variables.

Figure 15 depicts precipitation and diversity ( $H'$ ) at the downstream sampling station (Station 3) for the months of sampling. Diversity ( $H'$ ) is shown in solid bar lines. The height of these lines is the magnitude of diversity ( $H'$ ) for Station 3. Precipitation appears as a solid area behind the solid bar lines. The x axis presents all field trips that took place during this study. Since this representation is not intended to produce a quantifiable relationship between the two, the total monthly precipitation has no arithmetic values assigned to it.



**Figure 15: Total monthly precipitation and diversity (H') for Station 3**

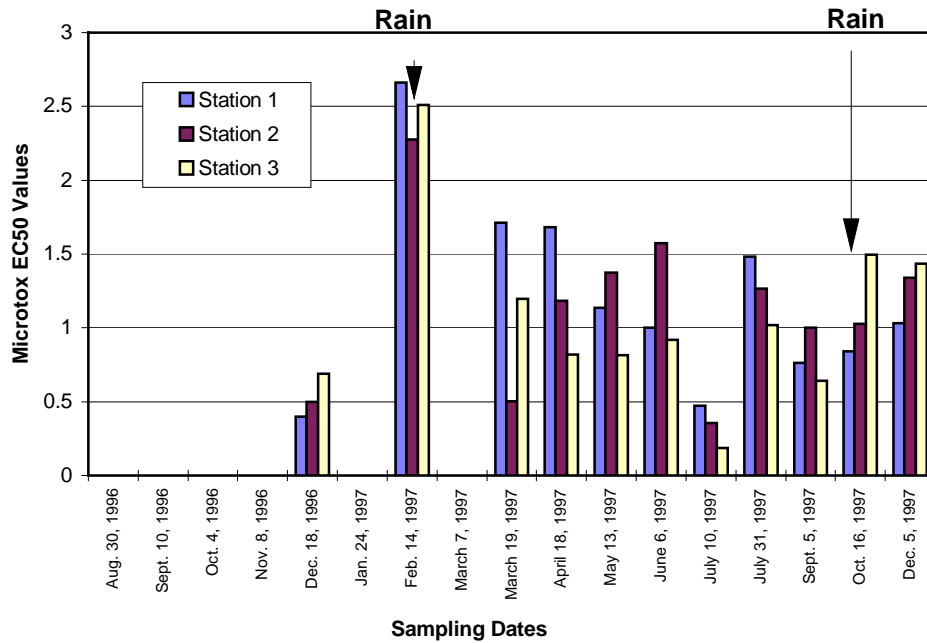
The two variables depicted in Figure 15 exhibit a weak inverse correlation. Diversity (H') appears to decrease with increasing rainfall amounts and to increase at low rainfall periods. Although other variables also enter this relationship, such as seasonality, the data suggest some evidence exists to support the hypothesis that precipitation has an effect on the integrity of the benthic community directly downstream of the construction activity. The effect of precipitation on total taxa and for the other two sampling stations is not as clear.

### ***Microtox Sediment Test Results***

Results from the Microtox basic solid phase test for all soil samples collected during this study are plotted for all sampling stations at every sampling date on Figure 16. They are three graphical bars present for every sampling date in Figure 16. The height of the left bar indicates the EC50 value recorded from the Microtox test on the sediment sample at Station 1 for the particular sampling date. The height of the middle bar represents the EC50 value for Station 2 while the height of the right bar in each sampling date is the corresponding EC50 value recorded from the Microtox test on the sediment sample from Station 3. A high EC50 value indicates low toxicity whereas a low EC50 value indicates increased toxicity. The arrows that appear on the February 14, 1997 and October 16, 1997 field trips indicate that sampling took place during or right after a major rainfall event.

The first soil samples, collected on December 18, 1996, exhibit high toxicity for all three stations whereas every sample collected between February 14, 1997 and September 5, 1997 showed consistently a more toxic sediment composition at the downstream sampling station when compared to the upstream one. EC50 values for the downstream sampling station were in the range of 0.2 to 2.5 for that period of time. When some of these values are compared to the upstream station for the same period of time, they appear up to 50% smaller in magnitude than the corresponding EC50 values in Station 1. In other words, sediments in Station 3 appear up to 50%

more toxic than those in Station 1 for that period of time. The outfall sampling station



**Figure 16: Microtox EC50 values at every station**

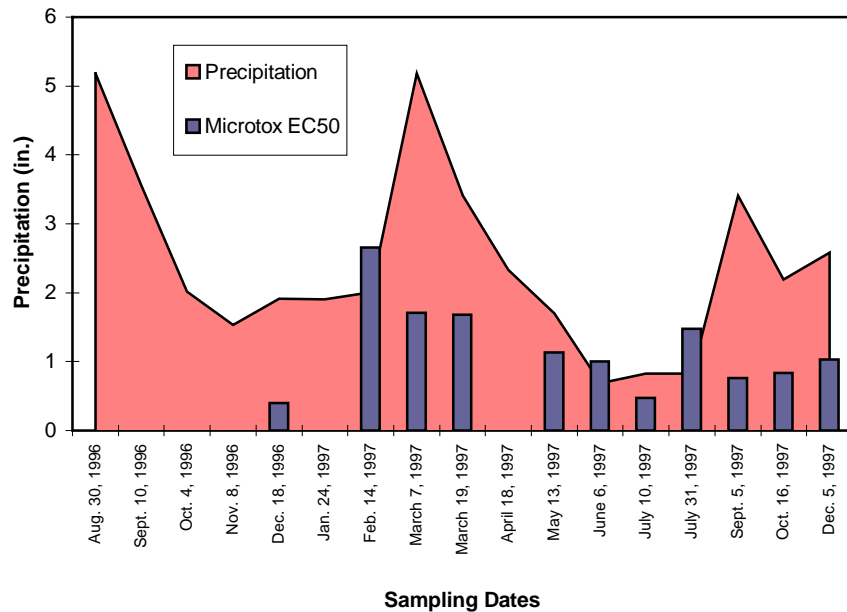
(Station 2) exhibited a more irregular pattern by appearing both more toxic and less toxic than the upstream station during the same time period.

As with the benthic results it is also important to observe any effects of certain rainfall events and total precipitation on the sediment toxicity. Again when looking at the immediate effect of rainfall events on sediment toxicity one may want to

observe for any changes in the EC50 values among sampling stations for those sampling dates that large rainfall events were recorded during or shortly prior to the time of sampling. In Figure 16 the sampling dates with a recorded rainfall event are marked with an arrow. Only two sampling dates when soil samples were collected and a rainfall event was recorded are present in our study and do not seem to provide any conclusive information on the immediate effect of rainfall on the sediment toxicity.

Toxicity tests on sediment samples collected during the February 14, 1997 field trip show slightly more toxic sediments on the downstream sampling station comparing to the upstream one. This observation however is reversed on the sediment samples collected on the October 16, 1997 field trip. On that date the downstream station appears less toxic than the upstream one.

The monthly total precipitation was also plotted with the Microtox results. Figure 17 shows precipitation and Microtox EC50 values at the downstream station (Station 3) for the months of sampling. Microtox EC50 results are shown in solid bar lines. The height of these lines is the magnitude of EC50 values for Station 3. Precipitation appears as a solid area behind the solid bar lines. The x axis presents all field trips that took place during this study. Since this representation is not intended to produce a quantifiable relationship between the two, the total monthly precipitation has no arithmetic values assigned to it.



**Figure 17: Total monthly precipitation and Microtox EC50 value for Station 3**

The plot on Figure 17 indicates that sediment toxicity downstream of the construction activity doesn't exhibit any relation to precipitation as was initially expected. Increased precipitation is not accompanied with a decrease in EC50 values (increase in toxicity) while at the same time during the months of least precipitation toxicity appears at its highest.

***Comparison of benthic bioassessment and Microtox sediment toxicity tests***

An essential part of this research was to investigate the utility of both the traditional bioassessment technique and that of the Microtox sediment toxicity test as engineering tools for monitoring the health of aquatic ecosystems which are stressed



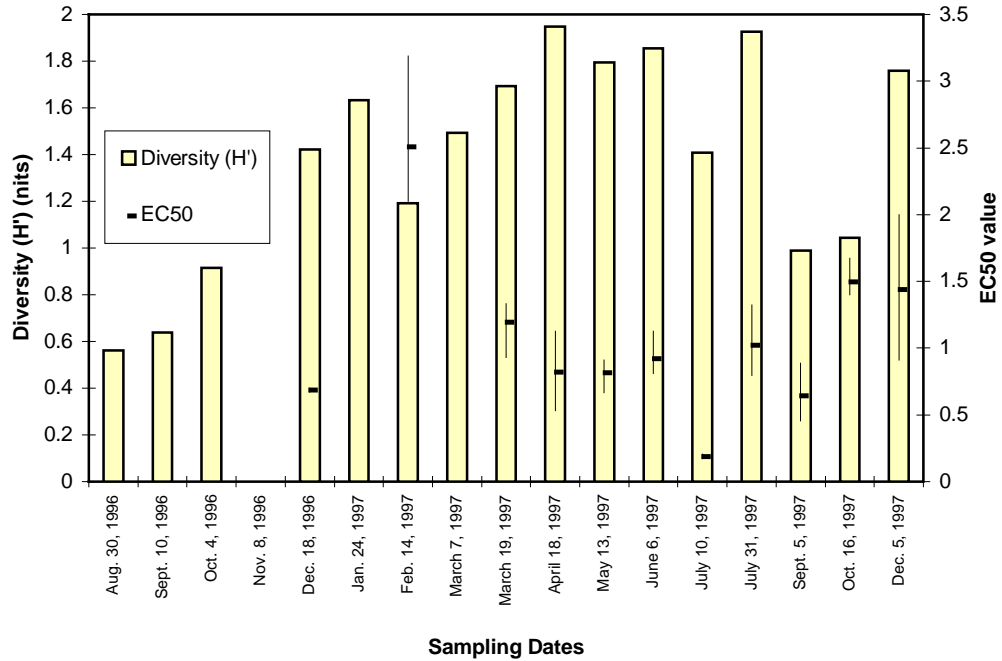
by construction activities. Each technique was examined individually at the previous two sections, however a comparison of the two is performed here.

One way to compare the two techniques is to examine whether they follow the same trends for each sampling station. For example when an improvement in the benthic community is observed at any station does that also correspond to an actual reduction in sediment toxicity for that station and vice versa. In order to find that, the Microtox EC50 values were plotted together with total taxa and diversity for Station 3.

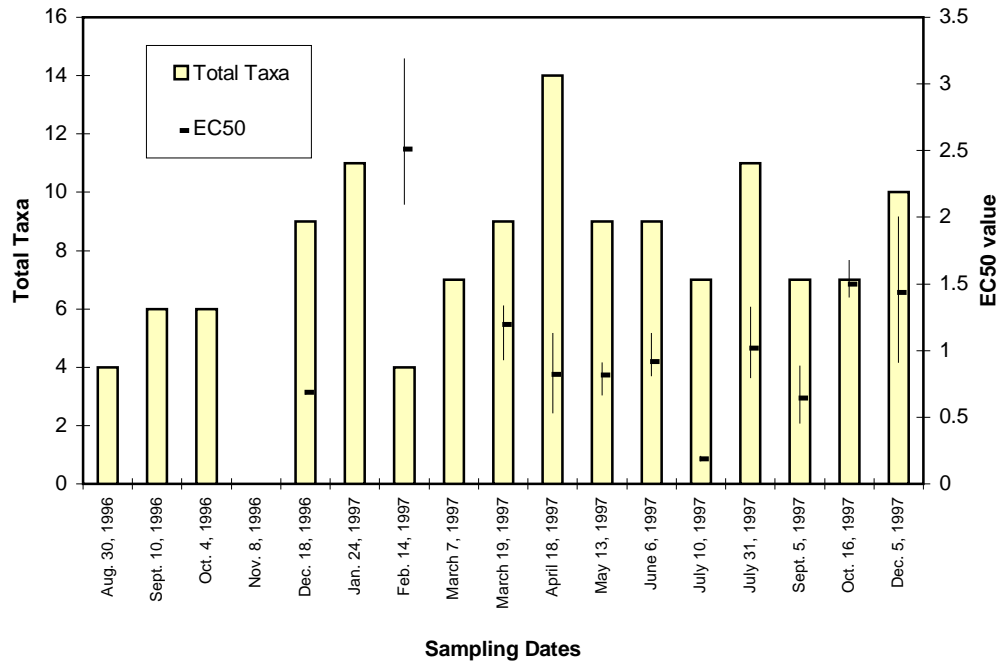
The graph on Figure 18 represents Diversity (H') index with bar lines for every field trip performed. The height of each line indicates the magnitude of Diversity (H') in units of "nits" for Station 3 and the arithmetic values can be found on the left hand axis. At the same time the average Microtox EC50 value at Station 3 for the same sampling date appears as a point in the graph and the corresponding magnitude can be found on the right hand axis. The lowest and highest value of EC50 from the triplicate tests performed in the sediment sample of Station 3 from that date are also shown as error bars.

The graph on Figure 19 uses the same format as the plot in Figure 18. The only exception is that the bar lines on every field trip date, now represent the magnitude

of total taxa for that date. Again the arithmetic values appear at the left hand axis for total taxa as they did for Diversity ( $H'$ ) in Figure 18.



**Figure 18: Diversity ( $H'$ ) and Microtox EC50 values for Station 3**



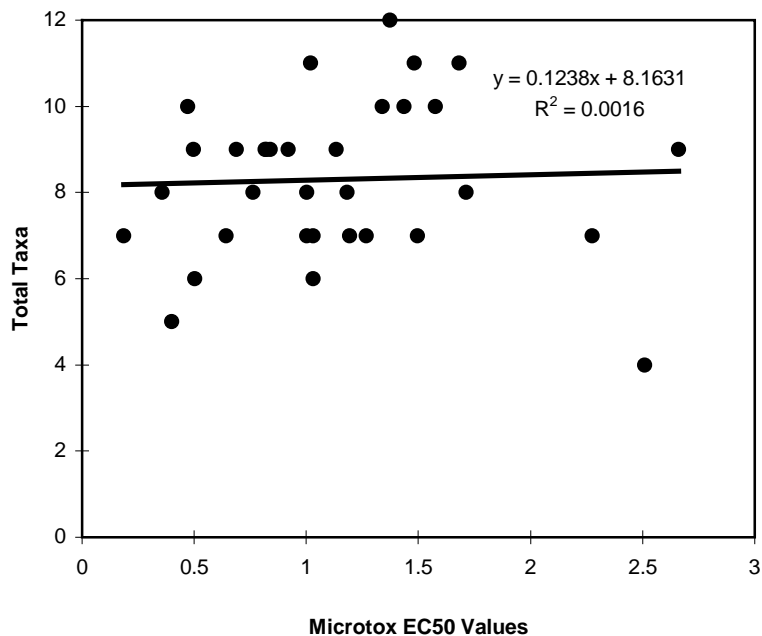
**Figure 19: Total Taxa and Microtox EC50 values for Station 3**

The two plots do not indicate that sediment toxicity has any relation to the two metrics downstream of the construction activity. For example on the February 14, 1997 field trip, total taxa exhibited the lowest value recorded during this study whereas toxicity appeared at a record low for the entire study. The same behavior was observed for the rest of the metrics and the rest of the sampling stations.

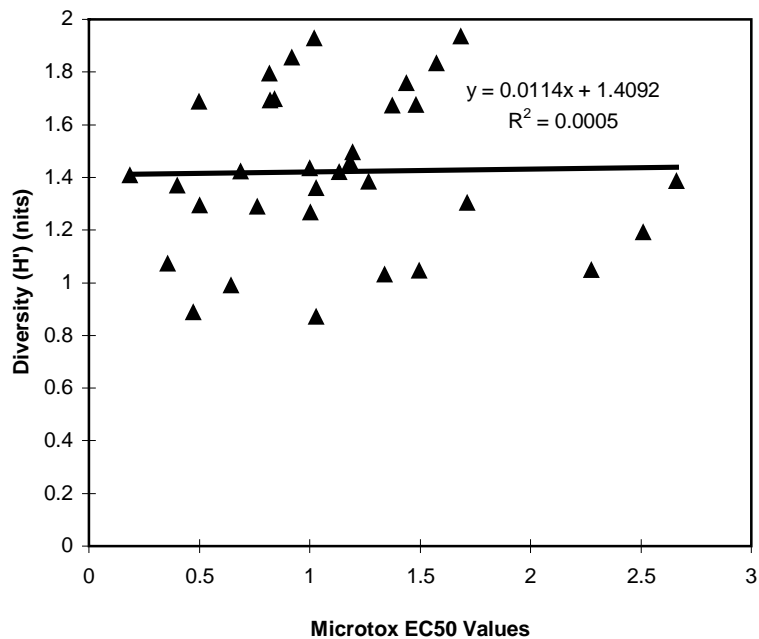
In order to further support this lack of correlation we can perform a regression analysis for the relationship of the Microtox EC50 values and the calculated metrics and then measure the percentage of the variability in the metrics that can be expressed in the EC50 values. In other words we can calculate the  $R^2$  value using

Equation 3, for a series of linear regressions where the y variable is a metric value and the x is the Microtox EC50 values.

Figure 20 and Figure 21 are plots of total taxa and diversity against Microtox EC50 values respectively. The graph in Figure 20 shows the total taxa measurements made for all benthic samples collected in this study plotted against the Microtox EC50 values from all sediment samples collected from the same sampling stations and the same sampling dates as the benthic samples that generated the total taxa values. The y axis represents total taxa whereas the x axis represents the Microtox EC50 values. The same graphical format is used in Figure 21. In this plot however, diversity (H')



**Figure 20: Total taxa versus Microtox EC50 values for all stations**



**Figure 21: Diversity (H') versus Microtox EC50 values for all stations**

measurements were plotted against Microtox EC50 values and the y axis represents Diversity (H') in values of “nits” instead of total taxa.

Linear regressions were made for both plots as shown by the straight lines and the  $R^2$  values were calculated and tabulated in Table 6.

**Table 6:  $R^2$  values for regressions performed for all metrics with Microtox results**

	Total Taxa	Total Polychaete Taxa	Diversity (H')
$R^2$ Values	0.0016	0.0156	0.0005

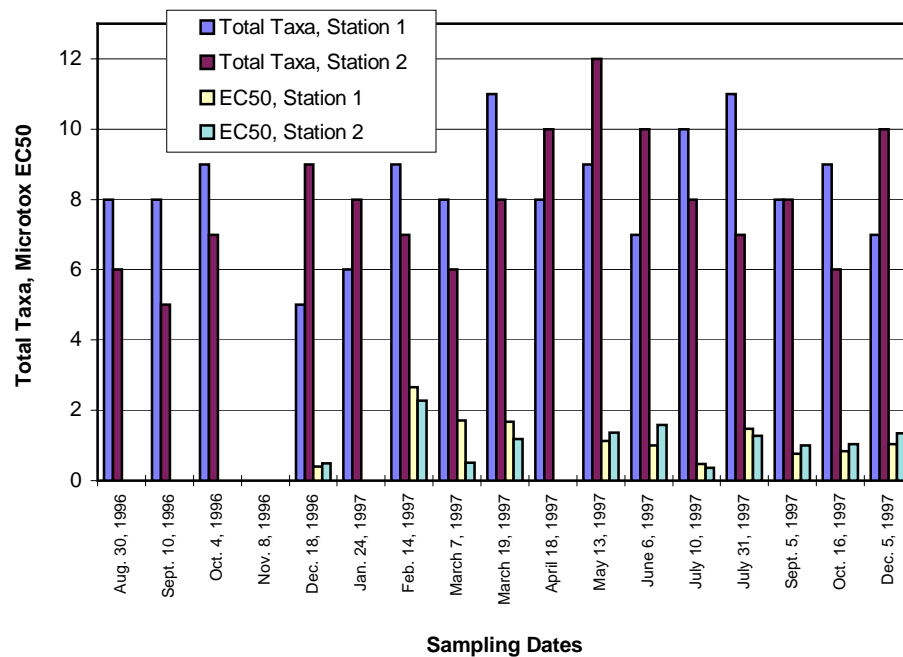
The calculated slopes of the regression lines are practically zero (no trend) and the coefficient of linear correlation ( $R^2$ ) is also practically zero, which supports the conclusion that there is no detectable pattern between the two methods when looking at single observations.

However, is there an absence of detectable pattern when comparing trends among stations? In other words, when the downstream station's benthic population appears affected compared to the upstream station's does the station's sediment also appear more toxic in comparison to the upstream station?

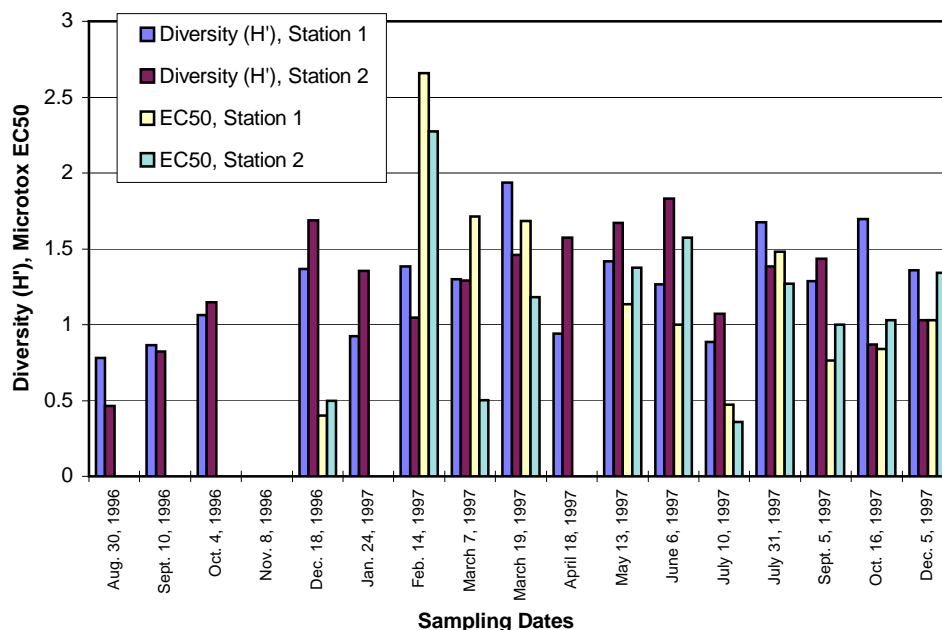
In order to make such a comparison a series of plots of the Microtox EC50 values and the metrics from the benthic bioassessment were created and compared. In Figure 22, total taxa and Microtox EC50 values are plotted together for each sampling date. The x axis shows the dates when samples were collected during this study. With the exception of the sampling days occurring before December 18, 1996, when only benthic samples were collected, all other sampling dates have four column bars. The left most bar, for each sampling date, represents the total taxa for Station 1 and the height of this bar, which can be measured from the left hand axis, is the magnitude of this metric at Station 1 for that day. The bar to its right represents total taxa for Station 2 and once more the height provides the magnitude of the metric at Station 2. The other two bar lines reflect the magnitudes of the Microtox EC50 values measured from the sediment samples for that particular field

trip. The third bar from the left shows the magnitude of EC50 for Station's 1 sediment sample whereas the last bar reflects the same measurement for Station's 2 sample.

Figure 23 has the exact same format as Figure 22. The only difference here is that the two left bars represent diversity (H') instead of total taxa. Again the comparison remains between Station's 1 and Station's 2 samples.



**Figure 22: Total taxa and Microtox EC50 values for Stations 1 and 2**



**Figure 23: Diversity (H') and Microtox EC50 values for Stations 1 and 2**

It appears that when comparing two stations using either technique there is almost always an agreement. When the outfall station's (Station 2) sediment is more toxic than the upstream one, the benthic community appears more affected by exhibiting decrease in total taxa or diversity (H'). This relation also holds true if a comparison is made between the upstream and downstream stations for both these metrics. However, the magnitude of change in benthic metrics and Microtox EC50 values between the two stations is not comparable to each.

*Effect of particle sizes on the Microtox basic solid phase test results*



In the process of developing a method for demonstrating the effects of particle sizes of sediment samples on the effective concentration values (EC50) produced by the Microtox basic solid phase test, several techniques were used.

Soil sample was collected from the Clear Lake area and was first wet sieved through the No. 200 sieve (75  $\mu\text{m}$ ). When the sieved sample was run through a particle size analyzer (Mastersizer) it was found that many large particles appeared in the subsample that passed the No. 200 sieve while a large number of smaller particles was left trapped in the filter cake formed during the sieving process. This result indicated that wet sieving was an inappropriate procedure as it didn't provide good separation of particles with sizes smaller than 75  $\mu\text{m}$  from those greater in size.

Dry sieving was then considered as an alternative for providing the type of separation needed to perform these experiments. After oven drying, the Clear Lake soil sample was manually sieved. As with wet sieving, the sample was then run through the Mastersizer to detect the accuracy of the process. It was concluded that although the procedure was largely improved it still failed to provide a good separation. The subsample that was retained in the No. 200 sieve should only consist of particles with size greater than 75  $\mu\text{m}$ , but when run through the Mastersizer it showed that at least 40% by volume of its particles were smaller in size than 75  $\mu\text{m}$ .

Finally a mechanical sieve shaker was used on the dried soil sample as discussed in the methodology chapter. The particle size analyzer showed improvement over the manual sieving process. Total separation of the two particle sizes (those greater in size than 75 $\mu\text{m}$  from those smaller in size) however, was never accomplished. After mechanically sieving five times the subsample that was retained in the No. 200 sieve, the particle size analyzer was still able to detect that 25%-30% by volume of the sample's particles had sizes smaller than 75  $\mu\text{m}$ . On the other end, the subsample that passed through the No. 200 sieve and would theoretically consist in its entirety of particles with sizes smaller than 75  $\mu\text{m}$ , was detected by the Mastersizer as having only about 80% by volume of its particles smaller in size than 75  $\mu\text{m}$ . In other words, with a standard procedure such as mechanical dry sieving

and with repeatable sieving efforts a clear separation between particles with sizes greater and less than 75  $\mu\text{m}$  was never achieved.

Several mixtures by weight of the dried sample that was manually and mechanically sieved were run through the Mastersizer and the results are presented in Figure 24. The graph in Figure 24 shows a series of soil mixtures composed from 0% to 100% by weight of particles passing the No. 200 sieve (Aperture 75  $\mu\text{m}$ ) in increments of 10%, plotted against the readings from the particle size analyzer (Mastersizer) for both the mechanical and manual sieving process. The corresponding measurements from the particle size analyzer appear in percent by volume smaller in size than 75  $\mu\text{m}$ .

The plot on Figure 24 shows a considerable improvement in the mechanical sieving procedure when compared to the manual sieving one for up to 70% by weight of sample passing the No. 200 sieve. Above 70% by weight, the manual sieving with the less vigorous shaking seems to provide a better separation of the bigger particles from the smaller ones when compared to the mechanical sieving process. When comparing the two procedures it is assumed that the closer numerically the sample compositions expressed in a weight and volume percentage are, the better the results. In other words the ideal procedure would plot on a 45 degree angle in Figure 24.

In a similar set of experiments that were performed in the Carolinian Province study, for a different Microtox sediment testing procedure however, artificial clay and sand mixtures were used. Although the selection of such soil mixtures insured a clear

**Figure 24: Mixtures by percent weight vs. Mastersizer readings by**

**percent volume for the mechanically and manually dry sieved samples.**

separation of the two sizes it failed to properly represent the soil composition of natural soils. All results in the Carolinian Province study expressed particle size in a percent by weight smaller than 75  $\mu\text{m}$  (or percent of silt-clay by weight).

In this study it was decided that natural soil from the same region as the test site (Cow Bayou) would more accurately reflect the particle size distribution of sediments regularly collected. Results from this experiment express particle size in a percent by volume smaller than 75  $\mu\text{m}$  (less than No. 200 sieve) as measured using the particle size analyzer. As long as all the sediments collected in the field are run through the particle size analyzer and all have at least 25%-30% of their particles by volume smaller than 75  $\mu\text{m}$ , this might prove to be a better representation of particle size. Because the test site appeared to have a highly silty substrate, all collected samples were expected to have at least 25%-30% of their particles by volume smaller than 75  $\mu\text{m}$ . However there would still remain an untested region between 80% - 100% by volume smaller in size than 75  $\mu\text{m}$  for which this procedure would not compensate.

Once mechanical sieving was selected as the procedure for separating the different soil sizes, the samples to be run through the Microtox were prepared. As discussed in the methodology chapter, samples composed of particles with 0%, 10%, 50%,

70% and 100% by weight smaller than 75  $\mu\text{m}$  were selected to demonstrate the effect of particle size on the EC50 values. The same sample mixtures were also prepared for running through the Mastersizer and obtaining their percent by volume smaller than 75  $\mu\text{m}$  composition.

In total fifteen samples were run using the Microtox basic solid phase test. The five soil mixtures mentioned before were tested using no initial dilution, 1/1.5 initial dilution and 1/3 initial dilution. Each sample was tested in duplicates. The results of those tests as well as those from the Mastersizer were combined to generate Figure 25. The graph in Figure 25 shows the five soil mixtures composed of particles with 0%, 10%, 50%, 70% and 100% by weight smaller than 75  $\mu\text{m}$  in three initial dilutions, plotted using the readings from the particle size analyzer (Mastersizer) in the x axis and the corresponding values (EC50) from the Microtox test in the y axis. The measurements from the particle size analyzer appear in percent by volume smaller in size than 75  $\mu\text{m}$ . Because all samples were run in duplicates, the results are averaged for each sample and the upper and lower values are shown as error bars.

**Figure 25: Effect of particle size on Microtox EC50 values.**

Several observations can be made from the results of this experiment:

(1) If for a soil sample its composition of particles with sizes less than 75  $\mu\text{m}$  exceeds the value of approximately 55% by volume, the EC50 value as that is

generated through the Microtox basic solid phase test is reduced to a value of less than about 3. This observation is true for all three dilutions.

(2) The results from the soil samples having a 1/1.5 initial dilution, which was frequently used when testing most of the soil samples from Cow Bayou, show small variation in their EC50 values when compared to the samples having no initial dilution. The EC50 values are almost identical for the two different dilution sets for all soil mixtures except the 100% by weight mixture ( $\cong$  82.65% by volume). In that particular soil mixture the EC50 value is slightly increased (sample appears less toxic) when compared to the no initial dilution case.

(3) For all five soil mixtures, the highest dilution (1/3) appears to give the highest EC50 values (samples appear least toxic) on the Microtox basic solid phase test.

#### ***Effect of sample storage time on the Microtox Basic Solid Phase Test results***

As described in the methodology chapter, a soil sample from Cow Bayou was well homogenized prior to testing and was then separated in five containers each to be run through the Microtox analyzer using the basic solid phase test. One of the subsamples was run the same day of its collection while the others were run the second day after collection, a week after, two weeks and a month later. The results are shown in Figure 26 where the EC50 values produced by the basic solid phase test are plotted against the time of storage for each sample. Because all samples were run



**Figure 26: Effect of sample storage time on EC50 values.**

in triplicates, the results are averaged for each sample and the upper and lower values are shown as error bars.

It can be seen from Figure 26 that toxicity is slightly reduced with storage time. The samples that were run after the first week exhibit higher EC50 values, therefore appear less toxic, than the samples that were run the first two days of the experiment. However the difference can be considered minimal because the lowest average EC50 value occurring at Day 2 of storage is 0.45 while the highest average EC50 value occurring at Day 14 of storage is only 0.66, a difference of less than 0.21.

## **Chapter 5 Summary and Conclusions**

### *Summary*

The Federal Register requires all construction activities that disturb over one acre of land area to be authorized by a National Pollutant Discharge Elimination System (NPDES) general permit. Such a permit mandates the usage of Storm Water Pollution Prevention Plans (SW3Ps). The effectiveness of these plans has not yet been determined in protecting the health of the surface waters. In order to justify the usage and continuation of such practice a road construction site in the Houston area was selected together with a target stream that was determined as one of the final receptors of the drainage from this project. Monthly monitoring and sampling of the stream was undertaken during construction. Two techniques were to be used in detecting the presence of impact on the aquatic ecosystem. A traditional benthic bioassessment technique was accompanied by a newly developed sediment testing technique, the Microtox basic solid phase test. Samples from three stations were collected at every field trip. One station was located upstream of the drainage outfall, one was located at the outfall and the third was a small distance further downstream. A total of 17 sets of samples were collected in a period of 1-1/2 years.

### *Conclusions*

At the beginning of this field monitoring study, construction on NASA Road 1 was the heaviest right at or just near Cow Bayou. The most land disturbance observed in the area adjacent to the test site during this study was at that period of time. Slowly

thereafter, the construction moved along NASA Road 1 further to the east. By the end of sampling, no equipment or workers could be located near the test site. Official completion of the part of the project that drained in Cow Bayou was not claimed until April 1998, almost four months after the last sample collection. Completion of the whole project and removal of all SW3Ps was announced on August 1998.

For the first three field trips a significant difference in the structure of the benthic community upstream and downstream of the construction was detected. There was 25% - 50% less taxa in the downstream sampling station (Station 3) comparing to the upstream station (Station 1) for those three measurements. Diversity showed no apparent reduction for the same period of time. It might be concluded that the presence of construction near the test site during the early stages of this study had the most adverse effect to the benthic population downstream of the construction site.

After the first three field trips however and for every field trip thereafter, both total taxa and diversity (H') indices showed variability in their results. The downstream station showed increase as well as reduction in taxa and diversity when compared to the upstream station.

The relation between rainfall and the structure of the downstream benthic community was investigated. It was found that a large rainfall event has an immediate effect on the total taxa of the benthic community located downstream of the construction. There was 22% - 55% less taxa in the downstream sampling station (Station 3) comparing to the upstream station (Station 1) for those days when large rainfall occurred during or shortly prior to sample collection.

The Microtox sediment testing reported increased toxicity in the sediment of the downstream sampling station when compared to the upstream station. Unlike the total taxa and diversity ( $H'$ ) data, this trend lasted the entire study period. In contrast with the findings from the bioassessment study, sediment toxicity results were independent of precipitation, a somehow unexpected result.

Shortcomings of the Microtox toxicity test were discovered from experiments that were initially developed to understand the sensitivity of this procedure. The basic solid phase test was found to be very sensitive to the sediment composition of a soil sample. For example, when a sample's silt-clay content (expressed in percentage by volume smaller in size than  $75 \mu\text{m}$ ) exceeded 55 - 60 %, even if the sample had only small natural toxicity levels, the sample appeared highly toxic with EC50 values falling below the value of two. Observations similar to this were made by the researchers involved in the Carolinian Province EMAP study while using the traditional solid phase test. For that test whenever the silt-clay content (expressed in

percentage by weight smaller than 75  $\mu\text{m}$ ) exceeded 10 - 20 %, the sample appeared toxic with EC50 values falling below two.

Despite such shortcomings, the Microtox basic solid phase test still possesses some advantages:

- (1) The Microtox basic solid phase test is not a complicated test and does not require a trained biologist to perform.
- (2) It is a relatively fast procedure when compared to the bioassessment method.
- (3) It provides a single output which is easy to understand.

### ***Recommendations***

The amount of impact observed in the early phases of the construction activity, expressed as reduction in total taxa, is quite comparable to the impact observed in studies that were undertaken when minimal erosion-control measures were required.

Reed (1977) reports reduction in taxa between 23% to 40% as the primary response observed in the macrobenthic community directly downstream of a construction activity. This reduction is actually less severe than the one observed in this study where reduction in taxa ranges from 25% to 50%.

This result leads the author to believe that the presence of such measures provides little if any relief to the benthic communities early in the construction process. The

long term effect of such measures however cannot be concluded with confidence from the results of this study. One thing that might be concluded however is that the present measures do not seem to protect the benthic community from the immediate effects of large rainfall events.

As far as selecting a technique that might be appropriate for monitoring similar aquatic ecosystems as the one monitored in this study, the traditional bioassessment method appears more reliable when compared to the newly developed technique of testing sediment toxicity (Microtox basic solid phase test).

For studies performed in estuarine environments where the silt-clay content is quite high and where external stresses are similar to the ones experienced from a road construction activity, the Microtox toxicity test on sediments will fail to show any subtle changes in actual toxicity as the presence of a high silt-clay content will overshadow the results of these tests.

However, in a site with high silt-clay content and highly contaminated sediments, or in a site with a more sandy substrate, this test might prove more useful. Great care should be exercised when choosing a reference sample. If the reference sample has a different clay-silt content than the actual sample to be tested, the results can be grossly misleading.

### ***Future Work***

Although this research has provided evidence that there might be a detectable early change in the health of the aquatic system adjacent to the construction site more work is necessary to identify whether any long term effects from road construction activities such as the one examined in this project to receiving watersheds can be detected.

This research has shown that there is evidence of a connection between rainfall amounts and the health of the benthic community downstream of a construction activity; however additional effort should be put in quantifying the relation between the two. A future research project that would attempt to quantify such a relation would have to monitor the aquatic life more frequently while at the same time collect daily site specific rainfall data.

From conclusions made in this thesis it is apparent that when developing such a project, care should be taken that all phases of construction are monitored and recorded. The type of daily activities near the sampling site should be carefully recorded as well as the location in respect to the investigated site.



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## Appendix A: Results of Benthic Macroinvertebrate Analysis

**Table 7: Benthic species collected in Cow Bayou for the August 30, 1996 field trip**

Phylum	Class/SubClass	Order/Family	Genus	Species	Station 1	Station 2	Station 3
			<i>Acari</i>		0	0	0
<b>Annelida</b>							
Polychaeta							
			<i>Hopsonia</i>		0	0	0
			<i>Laeonereis</i>	<i>Culveri</i>	0	1	0
			<i>Mediomastus</i>		0	0	0
			<i>Nereis</i>		0	1	0
		Spionidae	<i>Streblopsio</i>	<i>Benedicti</i>	0	0	0
				other	0	0	0
Oligochaeta					0	0	2
<b>Anthropoda</b>							
Crustacea							
	Cirripedia				0	0	0
	Cladocera				0	0	0
	Copepoda				0	0	0
		Cyclopoida			0	0	0
		Harpacticoida	<i>Scottolana</i>	<i>Canadensis</i>	2	4	0
	Ostracoda				0	0	0
<b>Insecta</b>		Chironomidae			10	1	1
<b>Nematoda</b>					100	91	110
<b>Nemertea</b>					7	3	22
<b>Mollusca</b>							
Gastropoda					0	0	0
Bivalvia			<i>Rangia</i>	<i>Cuneata</i>	0	0	0
<b>Platyhelminthes</b>							
Turbellaria					0	0	0

**Table 8: Benthic species collected in Cow Bayou for the September 10, 1996**

**field trip**

Phylum	Class/SubClass	Order/Family	Genus	Species	Station 1	Station 2	Station 3
			<i>Acari</i>		1	0	1
<b>Annelida</b>							
Polychaeta							
			<i>Hopsonia</i>		0	0	0
			<i>Laeonereis</i>	<i>Culveri</i>	0	0	0
			<i>Mediomastus</i>		0	0	0
			<i>Nereis</i>		4	0	1
		Spionidae	<i>Streblopsio</i>	<i>Benedicti</i>	0	0	0
				other	0	0	0
Oligochaeta					0	3	4
<b>Anthropoda</b>							
Crustacea							
	Cirripedia				4	0	0
	Cladocera				0	0	0
	Copepoda				0	0	0
		Cyclopoida			0	0	0
		Harpacticoida	<i>Scottolana</i>	<i>Canadensis</i>	0	0	0
	Ostracoda				0	5	0
<b>Insecta</b>		Chironomidae			12	1	1
<b>Nematoda</b>					84	80	93
<b>Nemertea</b>					1	20	12
<b>Mollusca</b>							
Gastropoda					0	0	0
Bivalvia			<i>Rangia</i>	<i>Cuneata</i>	0	0	0
<b>Platyhelminthes</b>							
Turbellaria					0	0	0

**Table 9: Benthic species collected in Cow Bayou for the October 4, 1996 field trip**

Phylum	Class/SubClass	Order/Family	Genus	Species	Station 1	Station 2	Station 3
			<i>Acari</i>		0	0	0
<b>Annelida</b>							
Polychaeta							
			<i>Hopsonia</i>		0	0	1
			<i>Laeonereis</i>	<i>Culveri</i>	0	0	0
			<i>Mediomastus</i>		0	0	0
			<i>Nereis</i>		2	0	0
		Spionidae	<i>Streblopsio</i>	<i>Benedicti</i>	1	0	0
				other	0	0	0
Oligochaeta					1	1	0
<b>Anthropoda</b>							
Crustacea							
	Cirripedia				2	3	2
	Cladocera				0	0	0
	Copepoda				0	0	0
		Cyclopoida			0	0	0
		Harpacticoida	<i>Scottolana</i>	<i>Canadensis</i>	1	2	0
	Ostracoda				0	0	0
<b>Insecta</b>		Chironomidae			5	18	2
<b>Nematoda</b>					82	68	77
<b>Nemertea</b>					9	0	17
<b>Mollusca</b>							
Gastropoda					0	0	0
Bivalvia			<i>Rangia</i>	<i>Cuneata</i>	0	0	0
<b>Platyhelminthes</b>							
Turbellaria					0	0	0

**Table 10: Benthic species collected in Cow Bayou for the December 18, 1996**

**field trip**

Phylum	Class/SubClass	Order/Family	Genus	Species	Station 1	Station 2	Station 3
			<i>Acari</i>		0	0	0
<b>Annelida</b>							
Polychaeta							
			<i>Hopsonia</i>		11	36	16
			<i>Laeonereis</i>	<i>Culveri</i>	0	0	0
			<i>Mediomastus</i>		0	0	0
			<i>Nereis</i>		0	0	0
		Spionidae	<i>Streblopsio</i>	<i>Benedicti</i>	0	0	0
				other	0	0	0
Oligochaeta					0	10	2
<b>Anthropoda</b>							
Crustacea							
	Cirripedia				0	0	1
	Cladocera				0	0	0
	Copepoda				0	0	0
		Cyclopoida			0	0	0
		Harpacticoida	<i>Scottolana</i>	<i>Canadensis</i>	4	3	8
	Ostracoda				0	0	3
<b>Insecta</b>		Chironomidae			4	5	1
<b>Nematoda</b>					21	46	57
<b>Nemertea</b>					0	4	25
<b>Mollusca</b>							
Gastropoda					0	0	0
Bivalvia			<i>Rangia</i>	<i>Cuneata</i>	0	0	0
<b>Platyhelminthes</b>							
Turbellaria					0	0	0



**Table 11: Benthic species collected in Cow Bayou for the January 24, 1997**

**field trip**

Phylum	Class/SubClass	Order/Family	Genus	Species	Station 1	Station 2	Station 3
			<i>Acari</i>		0	0	0
<b>Annelida</b>							
Polychaeta							
			<i>Hopsonia</i>		2	6	8
			<i>Laeonereis</i>	<i>Culveri</i>	0	0	0
			<i>Mediomastus</i>		0	0	0
			<i>Nereis</i>		0	0	0
		Spionidae	<i>Streblopsio</i>	<i>Benedicti</i>	0	0	2
				other	0	0	0
Oligochaeta					0	0	1
<b>Anthropoda</b>							
Crustacea							
	Cirripedia				0	0	2
	Cladocera				0	0	0
	Copepoda				0	0	0
		Cyclopoida			0	0	0
		Harpacticoida	<i>Scottolana</i>	<i>Canadensis</i>	4	9	24
	Ostracoda				1	2	11
<b>Insecta</b>		Chironomidae			0	1	1
<b>Nematoda</b>					93	93	70
<b>Nemertea</b>					7	5	8
<b>Mollusca</b>							
Gastropoda					0	0	0
Bivalvia			<i>Rangia</i>	<i>Cuneata</i>	0	0	0
<b>Platyhelminthes</b>							
Turbellaria					0	2	0

**Table 12: Benthic species collected in Cow Bayou for the February 14, 1997**

**field trip**

Phylum	Class/SubClass	Order/Family	Genus	Species	Station 1	Station 2	Station 3
			<i>Acari</i>		0	0	0
<b>Annelida</b>							
Polychaeta							
			<i>Hopsonia</i>		0	1	0
			<i>Laeonereis</i>	<i>Culveri</i>	0	0	0
			<i>Mediomastus</i>		0	0	0
			<i>Nereis</i>		0	0	0
		Spionidae	<i>Streblopsio</i>	<i>Benedicti</i>	0	0	0
				other	0	0	0
Oligochaeta					0	0	0
<b>Anthropoda</b>							
Crustacea							
	Cirripedia				1	0	0
	Cladocera				0	0	0
	Copepoda				0	0	0
		Cyclopoida			0	0	0
		Harpacticoida	<i>Scottolana</i>	<i>Canadensis</i>	11	27	9
	Ostracoda				1	0	0
<b>Insecta</b>		Chironomidae			3	2	0
<b>Nematoda</b>					64	93	69
<b>Nemertea</b>					18	1	19
<b>Mollusca</b>							
Gastropoda					0	0	0
Bivalvia			<i>Rangia</i>	<i>Cuneata</i>	0	1	0
<b>Platyhelminthes</b>							
Turbellaria					0	0	0

**Table 13: Benthic species collected in Cow Bayou for the March 7, 1997 field trip**

Phylum	Class/SubClass	Order/Family	Genus	Species	Station 1	Station 2	Station 3
			<i>Acari</i>		0	0	0
<b>Annelida</b>							
Polychaeta							
			<i>Hopsonia</i>		3	1	3
			<i>Laeonereis</i>	<i>Culveri</i>	0	0	0
			<i>Mediomastus</i>		0	0	0
			<i>Nereis</i>		0	0	0
		Spionidae	<i>Streblopsio</i>	<i>Benedicti</i>	0	0	0
				other	0	0	0
Oligochaeta					0	0	0
<b>Anthropoda</b>							
Crustacea							
	Cirripedia				1	1	1
	Cladocera				0	0	0
	Copepoda				0	0	0
		Cyclopoida			0	0	0
		Harpacticoida	<i>Scottolana</i>	<i>Canadensis</i>	4	32	23
	Ostracoda				0	0	0
<b>Insecta</b>		Chironomidae			3	0	1
<b>Nematoda</b>					61	50	57
<b>Nemertea</b>					73	26	28
<b>Mollusca</b>							
Gastropoda					0	0	0
Bivalvia			<i>Rangia</i>	<i>Cuneata</i>	0	0	0
<b>Platyhelminthes</b>							
Turbellaria					0	0	0

**Table 14: Benthic species collected in Cow Bayou for the March 19, 1997 field trip**

Phylum	Class/SubClass	Order/Family	Genus	Species	Station 1	Station 2	Station 3
			<i>Acari</i>		0	0	0
<b>Annelida</b>							
Polychaeta							
			<i>Hopsonia</i>		2	0	2
			<i>Laeonereis</i>	<i>Culveri</i>	0	0	0
			<i>Mediomastus</i>		0	0	0
			<i>Nereis</i>		0	0	0
		Spionidae	<i>Streblopsio</i>	<i>Benedicti</i>	0	0	1
				other	0	0	0
Oligochaeta					1	0	0
<b>Anthropoda</b>							
Crustacea							
	Cirripedia				6	2	5
	Cladocera				0	0	0
	Copepoda				0	0	0
		Cyclopoida			0	0	0
		Harpacticoida	<i>Scottolana</i>	<i>Canadensis</i>	21	13	8
	Ostracoda				0	0	0
<b>Insecta</b>		Chironomidae			8	4	11
<b>Nematoda</b>					33	54	62
<b>Nemertea</b>					24	44	17
<b>Mollusca</b>							
Gastropoda					0	0	0
Bivalvia			<i>Rangia</i>	<i>Cuneata</i>	0	0	0
<b>Platyhelminthes</b>							
Turbellaria					3	1	0

**Table 15: Benthic species collected in Cow Bayou for the April 18, 1997 field trip**

Phylum	Class/SubClass	Order/Family	Genus	Species	Station 1	Station 2	Station 3
			<i>Acari</i>		0	0	0
<b>Annelida</b>							
Polychaeta							
			<i>Hopsonia</i>		1	0	0
			<i>Laeonereis</i>	<i>Culveri</i>	0	0	0
			<i>Mediomastus</i>		0	0	0
			<i>Nereis</i>		0	0	0
		Spionidae	<i>Streblopsio</i>	<i>Benedicti</i>	0	0	0
				other	0	0	0
Oligochaeta					0	0	2
<b>Anthropoda</b>							
Crustacea							
	Cirripedia				3	3	7
	Cladocera				0	1	0
	Copepoda				0	0	0
		Cyclopoida			0	0	0
		Harpacticoida	<i>Scottolana</i>	<i>Canadensis</i>	2	6	7
	Ostracoda				0	0	0
<b>Insecta</b>		Chironomidae			4	15	28
<b>Nematoda</b>					13	28	30
<b>Nemertea</b>					75	53	36
<b>Mollusca</b>							
Gastropoda					0	0	1
Bivalvia			<i>Rangia</i>	<i>Cuneata</i>	0	1	1
<b>Platyhelminthes</b>							
Turbellaria					0	0	1

**Table 16: Benthic species collected in Cow Bayou for the May 13, 1997 field trip**

Phylum	Class/SubClass	Order/Family	Genus	Species	Station 1	Station 2	Station 3
			<i>Acari</i>		0	0	0
<b>Annelida</b>							
Polychaeta							
			<i>Hopsonia</i>		1	0	0
			<i>Laeonereis</i>	<i>Culveri</i>	0	0	0
			<i>Mediomastus</i>		0	0	0
			<i>Nereis</i>		0	0	0
		Spionidae	<i>Streblopsio</i>	<i>Benedicti</i>	0	0	0
				other	0	0	0
Oligochaeta					4	2	0
<b>Anthropoda</b>							
Crustacea							
	Cirripedia				3	6	8
	Cladocera				0	0	0
	Copepoda				0	0	0
		Cyclopoida			0	0	0
		Harpacticoida	<i>Scottolana</i>	<i>Canadensis</i>	0	1	3
	Ostracoda				1	1	5
<b>Insecta</b>		Chironomidae			38	42	37
<b>Nematoda</b>					19	12	18
<b>Nemertea</b>					54	38	33
<b>Mollusca</b>							
Gastropoda					0	1	0
Bivalvia			<i>Rangia</i>	<i>Cuneata</i>	0	3	0
<b>Platyhelminthes</b>							
Turbellaria					0	0	3

**Table 17: Benthic species collected in Cow Bayou for the June 6, 1997 field trip**

Phylum	Class/SubClass	Order/Family	Genus	Species	Station 1	Station 2	Station 3
			<i>Acari</i>		0	0	0
<b>Annelida</b>							
Polychaeta							
			<i>Hopsonia</i>		0	0	0
			<i>Laeonereis</i>	<i>Culveri</i>	0	0	0
			<i>Mediomastus</i>		0	0	0
			<i>Nereis</i>		0	0	0
		Spionidae	<i>Streblopsio</i>	<i>Benedicti</i>	0	0	0
				other	0	0	0
Oligochaeta					0	5	6
<b>Anthropoda</b>							
Crustacea							
	Cirripedia				2	14	17
	Cladocera				0	0	0
	Copepoda				0	0	0
		Cyclopoida			0	0	0
		Harpacticoida	<i>Scottolana</i>	<i>Canadensis</i>	0	2	0
	Ostracoda				0	1	2
<b>Insecta</b>		Chironomidae			15	46	51
<b>Nematoda</b>					26	28	17
<b>Nemertea</b>					58	40	10
<b>Mollusca</b>							
Gastropoda					0	1	0
Bivalvia			<i>Rangia</i>	<i>Cuneata</i>	0	0	0
<b>Platyhelminthes</b>							
Turbellaria					0	0	0

**Table 18: Benthic species collected in Cow Bayou for the July 10, 19976 field trip**

Phylum	Class/SubClass	Order/Family	Genus	Species	Station 1	Station 2	Station 3
			<i>Acari</i>		0	0	0
<b>Annelida</b>							
Polychaeta							
			<i>Hopsonia</i>		0	0	0
			<i>Laeonereis</i>	<i>Culveri</i>	0	0	0
			<i>Mediomastus</i>		0	0	0
			<i>Nereis</i>		0	0	0
		Spionidae	<i>Streblopsio</i>	<i>Benedicti</i>	0	0	0
				other	0	0	0
Oligochaeta					2	0	1
<b>Anthropoda</b>							
Crustacea							
	Cirripedia				1	1	10
	Cladocera				0	0	0
	Copepoda				0	0	0
		Cyclopoida			0	0	0
		Harpacticoida	<i>Scottolana</i>	<i>Canadensis</i>	0	0	0
	Ostracoda				0	0	0
<b>Insecta</b>		Chironomidae			36	23	82
<b>Nematoda</b>					1	4	4
<b>Nemertea</b>					116	70	23
<b>Mollusca</b>							
Gastropoda					1	0	0
Bivalvia			<i>Rangia</i>	<i>Cuneata</i>	0	3	0
<b>Platyhelminthes</b>							
Turbellaria					0	0	0



**Table 19: Benthic species collected in Cow Bayou for the July 31, 1997 field trip**

Phylum	Class/SubClass	Order/Family	Genus	Species	Station 1	Station 2	Station 3
			<i>Acari</i>		0	0	0
<b>Annelida</b>							
Polychaeta							
			<i>Hopsonia</i>		0	0	0
			<i>Laeonereis</i>	<i>Culveri</i>	0	0	0
			<i>Mediomastus</i>		0	0	0
			<i>Nereis</i>		1	0	1
		Spionidae	<i>Streblopsio</i>	<i>Benedicti</i>	0	0	0
				other	0	0	1
Oligochaeta					47	15	1
<b>Anthropoda</b>							
Crustacea							
	Cirripedia				0	0	3
	Cladocera				0	0	0
	Copepoda				0	0	0
		Cyclopoida			0	0	0
		Harpacticoida	<i>Scottolana</i>	<i>Canadensis</i>	0	0	7
	Ostracoda				1	0	0
<b>Insecta</b>		Chironomidae			25	10	41
<b>Nematoda</b>					50	20	27
<b>Nemertea</b>					2	56	20
<b>Mollusca</b>							
Gastropoda					0	0	1
Bivalvia			<i>Rangia</i>	<i>Cuneata</i>	0	0	0
<b>Platyhelminthes</b>							
Turbellaria					0	0	0

**Table 20: Benthic species collected in Cow Bayou for the September 5, 1997**

**field trip**

Phylum	Class/SubClass	Order/Family	Genus	Species	Station 1	Station 2	Station 3
			<i>Acari</i>		0	0	0
<b>Annelida</b>							
Polychaeta							
			<i>Hopsonia</i>		10	2	1
			<i>Laeonereis</i>	<i>Culveri</i>	0	0	0
			<i>Mediomastus</i>		0	0	0
			<i>Nereis</i>		1	2	1
		Spionidae	<i>Streblopsio</i>	<i>Benedicti</i>	19	26	61
				other	0	0	0
Oligochaeta					3	0	0
<b>Anthropoda</b>							
Crustacea							
	Cirripedia				0	0	0
	Cladocera				0	0	0
	Copepoda				0	0	0
		Cyclopoida			0	0	0
		Harpacticoida	<i>Scottolana</i>	<i>Canadensis</i>	4	17	1
	Ostracoda				0	0	0
<b>Insecta</b>		Chironomidae			1	1	3
<b>Nematoda</b>					86	44	23
<b>Nemertea</b>					0	6	3
<b>Mollusca</b>							
Gastropoda					0	0	0
Bivalvia			<i>Rangia</i>	<i>Cuneata</i>	0	1	0
<b>Platyhelminthes</b>							
Turbellaria					0	0	0

**Table 21: Benthic species collected in Cow Bayou for the October 16, 1997 field trip**

Phylum	Class/SubClass	Order/Family	Genus	Species	Station 1	Station 2	Station 3
			<i>Acari</i>		0	0	0
<b>Annelida</b>							
Polychaeta							
			<i>Hopsonia</i>		0	0	1
			<i>Laeonereis</i>	<i>Culveri</i>	0	0	0
			<i>Mediomastus</i>		0	0	0
			<i>Nereis</i>		1	1	0
		Spionidae	<i>Streblopsio</i>	<i>Benedicti</i>	3	2	12
				other	0	0	0
Oligochaeta					0	0	0
<b>Anthropoda</b>							
Crustacea							
	Cirripedia				13	0	0
	Cladocera				0	0	0
	Copepoda				0	0	0
		Cyclopoida			0	0	0
		Harpacticoida	<i>Scottolana</i>	<i>Canadensis</i>	0	4	1
	Ostracoda				0	0	0
<b>Insecta</b>		Chironomidae			10	0	1
<b>Nematoda</b>					51	62	60
<b>Nemertea</b>					37	14	15
<b>Mollusca</b>							
Gastropoda					0	0	0
Bivalvia			<i>Rangia</i>	<i>Cuneata</i>	0	0	0
<b>Platyhelminthes</b>							
Turbellaria					0	0	0

**Table 22: Benthic species collected in Cow Bayou for the December 5, 1997**

**field trip**

Phylum	Class/SubClass	Order/Family	Genus	Species	Station 1	Station 2	Station 3
			<i>Acari</i>		0	0	0
<b>Annelida</b>							
Polychaeta							
			<i>Hopsonia</i>		2	2	2
			<i>Laeonereis</i>	<i>Culveri</i>	0	0	0
			<i>Mediomastus</i>		0	0	0
			<i>Nereis</i>		0	0	0
		Spionidae	<i>Streblopsio</i>	<i>Benedicti</i>	5	14	12
				other	0	0	1
Oligochaeta					0	1	0
<b>Anthropoda</b>							
Crustacea							
	Cirripedia				15	9	10
	Cladocera				0	0	0
	Copepoda				0	0	0
		Cyclopoida			0	0	0
		Harpacticoida	<i>Scottolana</i>	<i>Canadensis</i>	0	1	1
	Ostracoda				0	0	0
<b>Insecta</b>		Chironomidae			1	3	3
<b>Nematoda</b>					17	18	22
<b>Nemertea</b>					50	137	37
<b>Mollusca</b>							
Gastropoda					0	0	0
Bivalvia			<i>Rangia</i>	<i>Cuneata</i>	0	0	0
<b>Platyhelminthes</b>							
Turbellaria					0	0	2

**Appendix B: Results of Microtox Basic Solid Phase Tests**

**Table 23: EC50 values for all sediment samples collected from Cow Bayou**

Field Trip Date	Replicates	Station 1	Station 2	Station 3
Dec. 18, 1996	1	0.37	0.44	0.70
	2	0.35	0.54	0.67
	3	0.48	0.51	0.69
	Average	0.40	0.50	0.69
Feb. 14, 1997	1	2.87	2.56	3.19
	2	2.06	2.10	2.10
	3	3.05	2.16	2.24
	Average	2.66	2.27	2.51
March 19, 1997	1	1.90	0.59	0.93
	2	1.80	0.47	1.32
	3	1.44	0.45	1.33
	Average	1.71	0.50	1.19
April 18, 1997	1	1.75	0.96	0.53
	2	1.67	1.01	0.80
	3	1.63	1.59	1.13
	Average	1.68	1.18	0.82
May 13, 1997	1	0.89	1.17	0.87
	2	1.19	1.57	0.67
	3	1.33	1.39	0.91
	Average	1.13	1.37	0.82
June 6, 1997	1	0.89	1.56	0.82
	2	1.26	1.78	0.81
	3	0.86	1.39	1.13
	Average	1.00	1.58	0.92
July 10, 1997	1	0.44	0.36	0.21
	2	0.43	0.36	0.17
	3	0.54	0.35	0.18
	Average	0.47	0.36	0.19
July 31, 1997	1	1.70	1.05	0.80
	2	1.27	1.43	0.94
	3	1.48	1.33	1.32
	Average	1.48	1.27	1.02
Sept. 5, 1997	1	0.53	0.67	0.59
	2	0.71	1.02	0.88
	3	1.04	1.32	0.46
	Average	0.76	1.00	0.64
Oct. 16, 1997	1	0.62	1.30	1.40
	2	0.81	0.70	1.41
	3	1.09	1.08	1.68
	Average	0.84	1.03	1.50
Dec. 5, 1997	1	0.71	1.10	2.00
	2	1.43	1.32	1.40
	3	0.95	1.60	0.91
	Average	1.03	1.34	1.44

**Table 24: EC50 values for the soil mixtures tested in the experimental procedure developed to recognize the effects of particle sizes on the basic solid phase test**

Soil mixture by weight (% < 75 $\mu\text{m}$ )	Sediment composition by volume (% < 75 $\mu\text{m}$ )	Initial Dilution	1st replicate	2nd replicate
0	23.94	0	2.778	2.667
10	39.35	0	3.114	2.713
50	55.99	0	2.597	3.199
70	69.11	0	1.677	1.836
100	82.65	0	1.305	1.483
0	23.94	1/1.5	2.811	
10	39.35	1/1.5	2.919	2.837
50	55.99	1/1.5	2.646	3.468
70	69.11	1/1.5	1.729	1.865
100	82.65	1/1.5	1.676	2.185
0	23.94	1/3	3.118	3.974
10	39.35	1/3	3.37	4.056
50	55.99	1/3	3.851	4.139
70	69.11	1/3	1.997	2.605
100	82.65	1/3	2.517	4.02



**Appendix C: Results from Particle Size Analyzer (Mastersizer)**

**Table 25: Percent by volume of particles smaller than 75  $\mu\text{m}$  in soil mixtures created after a manual and a mechanical sieving process**

Soil mixture by weight (% < 75 $\mu\text{m}$ )	Mechanical sieving results by volume (% < 75 $\mu\text{m}$ )	Manual sieving results by volume (% < 75 $\mu\text{m}$ )
0	27.9	39.95
10	39.35	
20	42	49.2
30	46.67	54.63
40	49.99	62.09
50	56	65.4
60	59.4	72.77
70	69.11	
80	74.12	81.42
90	76.87	86.02
100	82.65	90.07



**Table 24: Percent by volume of particles smaller than 75  $\mu\text{m}$  in all sediment samples collected in Cow Bayou**

Field Trip Date	Station	Sediment composition by volume (% < 75 $\mu\text{m}$ )
Aug. 30, 1996	1	
	2	
	3	
Sept. 10, 1996	1	
	2	
	3	
Oct. 4, 1996	1	
	2	
	3	
Nov. 8, 1996	1	
	2	
	3	
Dec. 18, 1996	1	85.15
	2	81.4
	3	94.57
Jan. 24, 1997	1	
	2	
	3	
Febr. 14, 1997	1	71.02
	2	67.39
	3	66.15
March 7, 1997	1	95.03
	2	90.11
	3	96.62
March 19, 1997	1	72.31
	2	88.82
	3	91.15
April 18, 1997	1	
	2	
	3	
May 13, 1997	1	93.45
	2	81.07
	3	94.87
June 6, 1997	1	95.41
	2	76.01
	3	88.87
July 10, 1997	1	93.08
	2	89.82
	3	97.89
July 31, 1997	1	87.73
	2	89.75
	3	94.12
Sept. 5, 1997	1	92.09
	2	92.11
	3	90.5
Oct. 16, 1997	1	96.14
	2	86.8
	3	96.28
Dec. 5, 1997	1	95.01
	2	80.12
	3	85.45

## Appendix D: Sample Water Quality Data

**Table 25: Field and laboratory measurements of water quality in Cow Bayou**

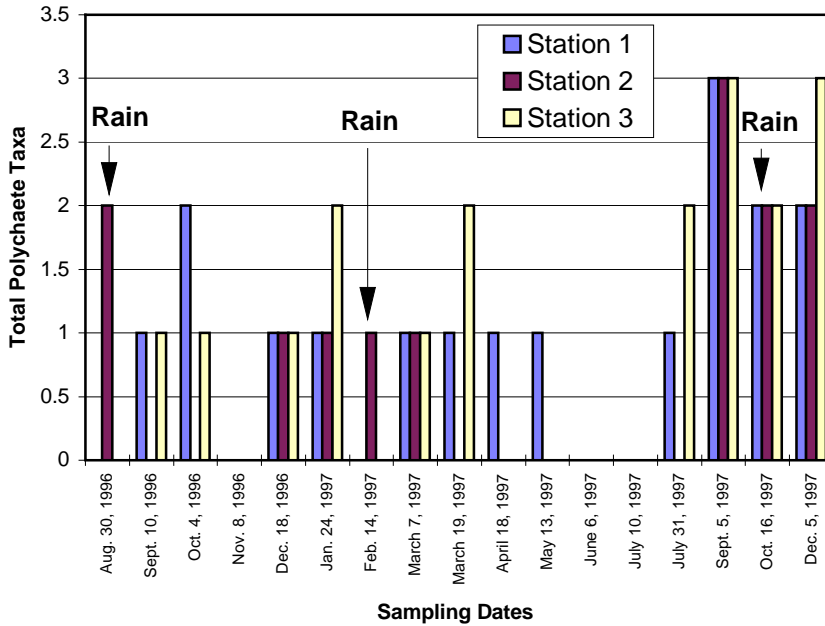
Field Trip Date	Station	Light Penetration (ft)	pH	DO (mg/L)	Conductivity (ms/cm)	TDS (g/L)	SS (mg/L)	Temperature °C
Aug. 30, 1996	1	1.1	6.6	4.36	0.32	0.15	33	26.2
	2	1.1	6.6	3.8	0.31	0.15	32	26.1
	3	0.75	5	4.26	0.29	0.14	74	25.8
Sept. 10, 1996	1	1.8	7.29	3.12	0.32	0.16	7	30.4
	2	1.7	7.25	3.03	0.3	0.14	24	28.8
	3	1.6	7.35	3.34	0.33	0.16	25	27.9
Oct. 4, 1996	1	1.75	8.05	5	5.62	2.8	18	24.4
	2	1.75	8.26	5.63	5.91	2.95	17	25.5
	3	1.75	8.35	6.22	5.88	2.98	10	24.8
Nov. 8, 1996	1	1.6	5.3	6.34			23	19
	2	1.6	7.43	6.5			21	19.3
	3	1.6	7.65	7.98			30	19.1
Dec. 18, 1996	1		7.56	9.36	4.79	2.83	42	8
	2		8.86	11.1	2.67	1.61	37	9.1
	3							
Jan. 24, 1997	1	1.2	6.2		0.52	0.24	45	21.2
	2	1.2	6.33		0.63	0.33	40	21.5
	3	1.1	6.6		4.73	2.3	40	20.6
Febr. 14, 1997	1	1.1	6.49	11.04			52	12
	2	0.9	6.36	11.03			46	11.5
	3	1	6.25	11.1			56	11.5
March 7, 1997	1	1.5	7.5	13.22	0.76	0.38	12	20
	2	1.25	7.85	13.31	0.75	0.37	40	19.4
	3	1.2	8.07	13.16	0.77	0.38	48	18.9
March 19, 1997	1	1.25	5.74	7.62	0.21	0.1	51	18.8
	2	0.83	5.86	7.4	0.19	0.09	49	18.6
	3	1.2	6.03	7.3	0.18	0.08	45	17.9
April 18, 1997	1	1.25	7.66	10.5			32	21.4
	2	1.67	7.55	9.93			41	21.1
	3	1.83	8	10.15			63	21
May 13, 1997	1	1.58	7.05	6.56	0.31	0.15	32	27.4
	2	1.58	8.15	8.25	0.41	0.2	36	27.4
	3	1.58	8.34	9.8	0.49	0.24	34	27.8
June 6, 1997	1	1.83	8.28	8.22	0.55	0.27	22	28.1
	2	1.67	8.6	9.17	0.8	0.4	29	26
	3	1.42	8.71	9.75	0.8	0.4	39	29.6
July 10, 1997	1	1.67	6.9	4.9	0.18	0.09	35	29.6
	2	1.25	7.83	5.5	0.37	0.18	36	30.5
	3	1.25	8.28	6.91	0.92	0.46	40	31.45
July 31, 1997	1	1.92	7.1	2.71			16	30.8
	2	1.67	7.08	3.46			13	31.1
	3	1.75	7.4	4.33			20	31.7
Sept. 5, 1997	1	1.67	8.19	7.36	17.25	9.11	8	31
	2	1.25	8.43	8.55	18.49	9.25	6	31
	3	1.417	8.65	9.57	18.66	9.33	10	31.6
Oct. 16, 1997	1	1.58	7.27	4.7	0.36	0.18	14	19.8
	2	1.58	7.7	4.26	0.36	0.18	14	20.1
	3	1.5	7.7	4.64	0.33	0.16	18	20.8
Dec. 5, 1997	1	0.75	6.55	8.35	0.56	0.28	31	15
	2	0.75	6.79	8.2	0.62	0.31	36	16.4
	3	0.75	6.88	8.2	0.82	0.4	37	17.6

## **Appendix E: Total Polychaete Taxa and Sheldon's Evenness Index**

Total polychaete taxa is a biotic indices. It reflect the health of the community through a measurement of the type of taxa present. In general it increases with increasing water quality. Total polychaete taxa is calculated by grouping all the species that belong in the polychaete class and summing them up.

Figure 27 displays the value of total polychaete taxa calculated for each of the three stations during this study. There are three graphical bars present for every sampling date. The height of each bar in any sampling day indicates the value recorded for the total polychaete taxa at each sampling station for that date. The height of the bar on the left represents the value of total polychaete taxa calculated for Station 1 whereas the height of the middle and right bar represent the value of the index for Station 2 and 3 respectively. Absence of one or more of the bars for a sampling date indicates that there were no species present that date that belonged in the polychaete class in the sample from one or more of the stations. The arrows shown in Figure 27 point to dates when sampling took place during or right after a major rainfall event.

Unlike the total taxa index, total polychaete index shows no apparent patterns when comparing upstream with downstream of the construction activity. The value of the total polychaete taxa varies between 0 and 3 for all three stations.



**Figure 27: Total polychaete taxa at every station**

Sheldon’s evenness index was also used in this study although it was not utilized in the Patrick Bayou assessment. This index is highly dependent on the Shannon - Wiener index and is calculated as follows:

$$\text{Sheldon's evenness index} = \frac{e^{H'}}{s}, \quad (4)$$

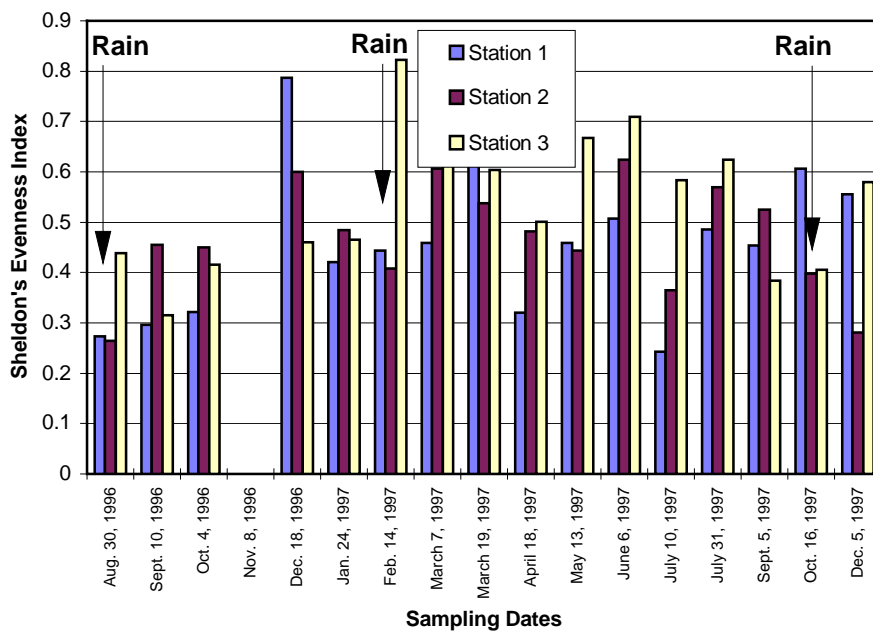
where  $H'$  is the Shannon - Wiener index,

$e = 2.71828$ , and

$s$  is the total number of different taxa.

Figure 28 shows the value of Sheldon’s evenness index calculated at each of the three stations during this study. There are three graphical bars present for every sampling

date. All field trips are shown in the x axis of the graph. The height of the outer left bar in any sampling day indicates the value recorded for the Sheldon's evenness index at Station 1 for that date. The height of the middle bar represents the value of Sheldon's evenness index for Station 2 while the height of the outer right bar in each sampling date is the corresponding value of the evenness index at Station 3. The arrows that appear on the August 30, 1996, February 14, 1997 and October 16, 1997 field trips indicate that sampling took place during or right after a major rainfall event.



**Figure 28: Sheldon's evenness index at every station**

Like total polychaete taxa, Sheldon's evenness index shows no apparent patterns when comparing upstream with downstream of the construction activity. The value

of the Sheldon's evenness index varied between the values of 0.26 and 0.62 for all three stations.

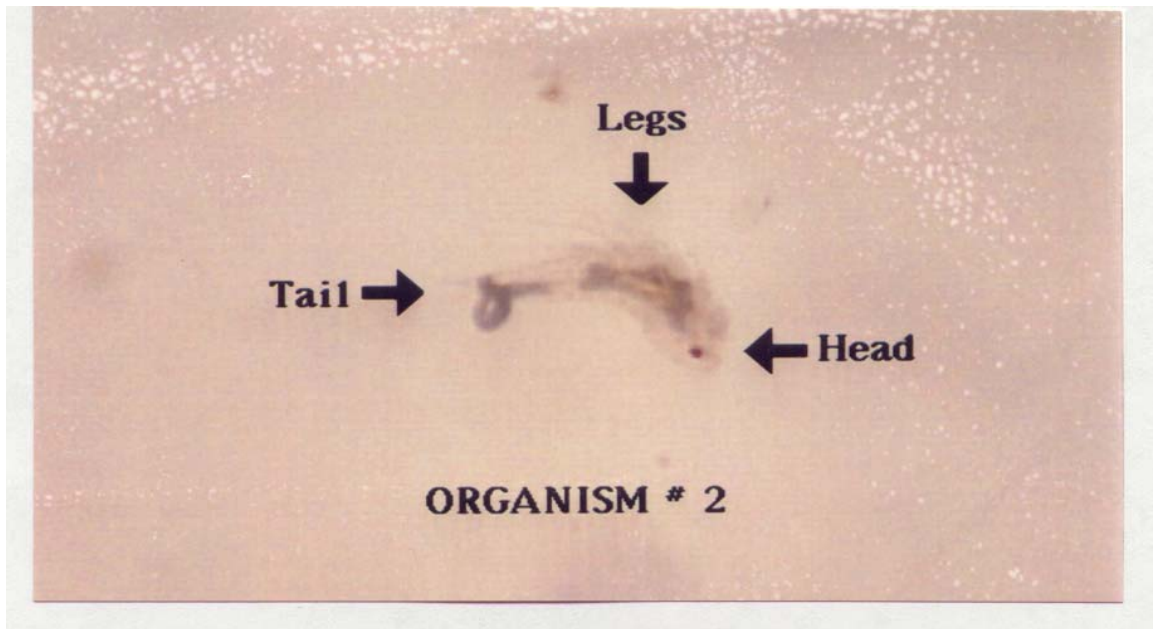
## Appendix F: Rainfall Data

**Table 26: Total monthly precipitation (in.) data from the National Climatic Data Center (NCDC) for the Houston Intercontinental and Galveston stations**

Location	State	Year	JAN	FEB	MAR	APR	MAY	JUN	JUL	AUG	SEP	OCT	NOV	DEC	ANNUAL
HOUSTON INT'CNTRL AP	TX	1996	0.88	1.29	0.12	2.05	0.56	8.37	1.11	10.58	6.96	2.6	4.55	3.74	42.81
GALVESTON	TX	1996	1.67	1.36	0.4	3.21	0.02	2.98	0.24	10.14	7.22	5.47	1.58	3.93	38.22
HOUSTON INT'CNTRL AP	TX	1997	3.8	4.9	7.96	7.17	6.19	4.47	1.67	2.26	4.86	7.11	3.38	5.42	59.19
GALVESTON	TX	1997	3.83	3.14	12.78	6.47	3.15	2.33	1.08	1.04	3.66	6.54	5.38	4.9	54.3

**Appendix G: Most Commonly Found Benthic Macroinvertebrates in Cow**

**Bayou**





**ORGANISM # 1**



**ORGANISM # 9**



