

**LANDSCAPE MONITORING AND BIOLOGICAL INDICATORS FOR
SEAGRASS CONSERVATION IN
TEXAS COASTAL WATERS**

**QUALITY ASSURANCE PROJECT PLAN
Revision 1**

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LIST OF ACRONYMS AND ABBREVIATIONS

| | |
|--------|--|
| CBBEP | Coastal Bend Bays & Estuaries Program |
| CV | Coefficient of Variation |
| DMP | Data Management Plan |
| DQO | Data Quality Objectives |
| EMAP | Environmental and Monitoring Assessment Program |
| EPA | Environmental Protection Agency |
| ESS | Environmental Sciences Section |
| FKNMS | Florida Keys National Marine Sanctuary |
| GIS | Geographic Information Software |
| GPS | Global Positioning System |
| IBI | Index of Biological Integrity |
| IISWR | International Institute for Sustainable Water Resources |
| MDM& A | Monitoring Data Management and Analysis |
| NIST | National Institute of Standards and Technology |
| OSHA | Occupational Safety and Health Administration |
| PAM | Pulse-Amplitude-Modulation |
| PAR | Photosynthetically Active Radiation |
| QA | Quality Assurance |
| QC | Quality Control |
| QAPP | Quality Assurance Project Plan |
| R-EMAP | Regional Environmental and Monitoring Assessment Program |
| RPD | Relative Percent Difference |
| SCUBA | Self-contained Underwater Breathing Apparatus |
| SGS | Sediment grain size |
| SOP | Standard Operating Procedure |
| SWQM | Surface Water Quality Monitoring |
| TCEQ | Texas Commission on Environmental Quality |
| TMDL | Total Maximum Daily Load |
| TPWD | Texas Parks and Wildlife Department |
| TRACS | TCEQ Regulatory Activities Compliance System |
| TSS | Total Suspended Solids |
| TSU-SM | Texas State University – San Marcos |
| TSWQS | Texas Surface Water Quality Standards |
| USEPA | United States Environmental Protection Agency |
| UT | University of Texas at Austin |
| UTCRWR | University of Texas Center for Research in Water Resources |
| UTMSI | University of Texas Marine Science Institute |

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Each copy of the Quality Assurance Project Plan (QAPP) will be given a serial number and mailed by the Project Officer to each recipient via “returned receipt” mail. The domestic return receipt (US Postal Service form PS 3811) when returned to the Quality Assurance (QA) Officer will be maintained as proof of receipt and to assure traceability of QAPPs. Individuals who are located at the same facility where the QAPP is prepared will be required to sign for the most recent QAPP copy.

SECTION I

A4: Project/Task Organization

The Landscape Monitoring and Biological Indicators for Seagrass Conservation project represents a collaborative effort among UTMSI, TSU-SM, CBBEP, TCEQ, and USEPA. The personnel at UTMSI is responsible for sample collection, data management, and data submission for the biological indicators portion of the project, while TSU-SM is responsible for these same tasks for the landscape monitoring portion. Data from both portions of the project will be submitted to the TCEQ Project Manager in the appropriate ASCII format. The Project Manager will then submit the data to the TCEQ SWQM Team Leader for inclusion of the data into the TRACS database. Personnel at CBBEP, TCEQ, and USEPA will oversee progress towards the project goals and participate in quality assurance procedures. A detailed description of the responsibilities of the project personnel follows:

EPA

Betty Ashley
EPA Project Officer

The EPA Project Officer serves as the contact between the CBBEP/TCEQ and EPA in order to achieve the project goals. The EPA Project Officer is also responsible for monitoring the progress of the project and tracks the deliverables.

Phil Crocker
EPA Aquatic Biologist

The EPA Aquatic Biologist reviews and approves the QAPP and all revisions. Additional responsibilities include assisting the EPA Project Officer and the CBBEP QA Officer in implementing the CBBEP Quality Management Plan and coordinating any necessary corrective actions.

CBBEP

Leo Trevino
Acting Project Manager

The CBBEP Project Manager is responsible for contract management and serves as the contact between the CBBEP, UTMSI, and TCEQ. The Project Manager also tracks the deliverables and project progress to ensure that the deliverables are accurate and submitted to the TCEQ according to schedule. The Project Manager reviews and approves the initial QAPP and all revisions or modifications. Additionally, the Project Manager is responsible for the submission of all written reports to the TCEQ and all data, in the proper format, to be included in the SWQM section of the TRACS database. The Project Manager will submit all data to the TCEQ SWQM Team Leader for entry into the TRACS database.

TCEQ

Jeff Foster Project Coordinator

The project coordinator is responsible for managing the TCEQ contract and serving as the contact between CBBEP and TCEQ. In addition to the Project Manager, the Project Coordinator reviews and approves all versions of the QAPP and any proposed amendments. The Project Coordinator also maintains TCEQ QA records and monitors the progress of the project and production of deliverables.

Sharon Coleman Quality Assurance Specialist

The Quality Assurance Specialist is responsible for TCEQ QA oversight of CBBEP projects. The QA Specialist also reviews the QAPP and all revisions or changes. In the event that the quality of data is compromised, the QA Specialist notifies the TCEQ/CBBEP Project Coordinator of the situation. Additionally, the QA Specialist may conduct audits of the monitoring and quality systems.

David Sullivan Manager, MDM&A

The TCEQ MDM&A Manager reviews the QAPP for valid monitoring stations, checks validity of parameter, program, and source codes, and ensures that data will be reported following instructions in the Surface Water Quality Monitoring Data Management Guide, most recent version. The MDM&A Manager surveys the TRACS database to monitor the submittal of scheduled sampling data and/or receives the data set from the TCEQ project manager for data summary/validation prior to submittal to the Information Resources division at TCEQ. The MDM&A Manager also provides data completeness reports to Project Managers as necessary and analyzes the TRACS database to identify level 1 data validation inconsistencies. Any inconsistencies are then reported to the Project Manager. The MDM&A serves as Monitoring Operations data management customer service representative for the TCEQ Project Manager and provides training to the TCEQ Project Manager to ensure that data are submitted according to documented procedures. There are no lines of communication between MDM&A and contractors.

Patrick Roques Team Leader, Surface Water Quality Monitoring Program

The SWQM Team Leader performs technical reviews of the QAPP and oversees the review of data for assessment purposes.

UTMSI

Kenneth Dunton Project Director

The UTMSI Project Director provides oversight for the Biological Indicator portion of the project and is responsible for implementing the CBBEP requirements in the contract and the QAPP. The Project Director coordinates activities that ensure comprehensive monitoring within the study. The Project Director identifies, receives, and maintains project quality assurance records. The UTMSI Project Director works in conjunction with the TSU-SM Project Director to provide accurate and timely deliverables to the Project Manager, and both Project Directors are responsible for compiling and submitting the Final and Draft Reports to the CBBEP. All references to the Project Director in this document refer to the UTMSI Project Director, Dr. Kenneth Dunton, unless otherwise specified.

Susan Schonberg Quality Assurance Officer

The UTMSI QA Officer is responsible for implementing the quality system as defined in both the contract and the QAPP. The QA Officer validates the data prior to submission to the CBBEP, and thereby, ensures the quality of the data submitted to the CBBEP. The entities responsible for data generation are independent of the Quality Assurance Officer.

Troy Mutchler Project Officer

The Project Officer is responsible for overseeing field monitoring operations, sample analyses, and data processing duties. The Project Officer also writes, maintains, and distributes the QAPP. The Project Officer is also responsible for maintaining records of QAPP distribution, including appendices and amendments. The Project Officer transfers all data and data management checklists to the UTMSI Project Director in the appropriate format for submission to the CBBEP. The Project Officer oversees all field and laboratory data collection, sample analyses, and data management.

Kimberly Jackson Field Operations Supervisor

The Field Operations Supervisor oversees the field personnel during sampling. It is the responsibility of the Field Operations Supervisor to ensure that the field personnel are properly trained and equipped to conduct field tasks. The Field Operations Supervisor guarantees that appropriate personnel, supplies, and equipment are available when necessary.

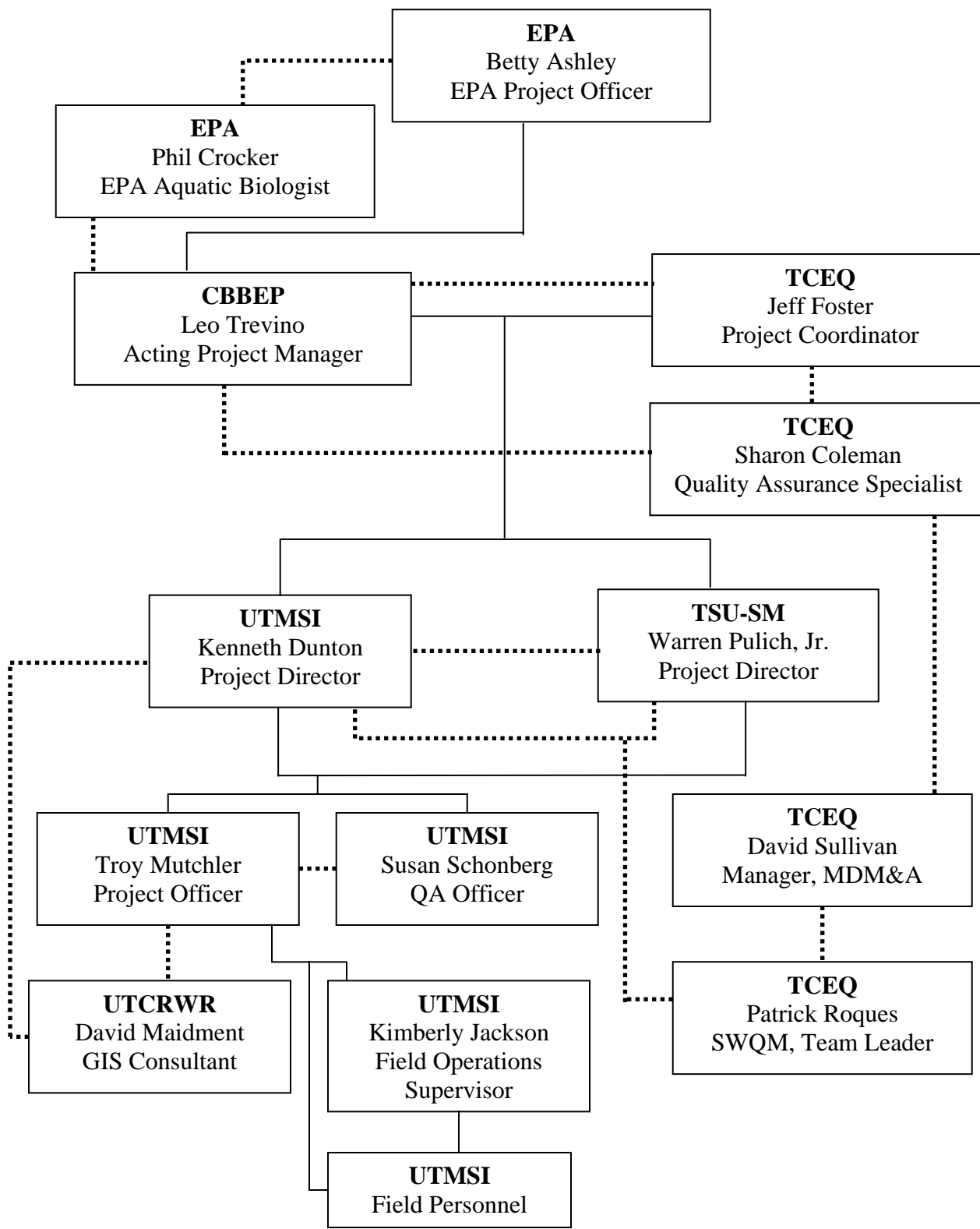
Research/Field/Technical Personnel

These personnel are responsible for performing field sampling, sample analyses, and data processing duties according to the project QAPP.

TSU-SM

Warren Pulich, Jr.
Project Director

The TSU-SM Project Director provides oversight for the acquisition of remote sensing imagery and monitoring of Landscape Indicators. The TSU-SM Project Director is responsible for implementing the CBBEP requirements in the contract and the QAPP and coordinating activities that ensure comprehensive monitoring within this portion of the study. The Project Director identifies, receives, and maintains project QA records. The TSU-SM Project Director works in conjunction with the UTMSI Project Director to provide accurate and timely deliverables to the Project Manager, and both Project Directors are responsible for compiling and submitting the Final and Draft Reports to the CBBEP.



—— Lines of Supervision
 Lines of Communication

Figure 1. Project/Task organization.

A5: Problem Definition/Background

Recently, a Seagrass Habitat Monitoring program was proposed for Texas coastal waters in the Seagrass Conservation Plan. This document, which describes many of the potential problems facing Texas seagrass habitats, recommended monitoring of key indices to detect ecosystem changes that occur before actual loss of seagrasses. Texas Parks & Wildlife (TPWD) and the Texas Commission on Environmental Quality (TCEQ) have initiated a planning process with resource managers and coastal researchers in Texas to develop a statewide, seagrass monitoring plan. This plan is to be based on sound scientific knowledge of seagrass resources and the relationship between temporally and spatially variable environmental parameters and seagrass distributions. A similar monitoring program developed for the Chesapeake Bay system significantly improved our knowledge of the biological and physical environment, resulting in an increase in public awareness and more effective resource management (Dennison et al. 1993). This project addresses the development of indicators and criteria that would be used in a future long-term monitoring plan for the State of Texas. This includes studies that test the effectiveness of various indicators and determination of optimum spatial and temporal sampling frequency that can be used to monitor seagrasses for management and conservation purposes.

Under a recent addition to the TCEQ Texas Surface Water Quality Standards, seagrass propagation is a protected aquatic life use, and water quality criteria should now be defined for use in regulatory applications (TCEQ 2000). In order to establish quantitative criteria for healthy Texas seagrass beds, environmental parameters and ecological indicators must be surveyed as part of a State-sponsored sampling and monitoring program. However, a monitoring protocol has not been identified for Texas seagrass beds. This includes selection of appropriate parameters or indicators and a probability-based sampling strategy. This project, which is a continuation of sampling completed under an EPA R-EMAP project, will focus on the development of a regional sampling design, the selection of specific techniques for seagrass ecological assessment, and the generation of a geospatial database in the latest version of Arc/Info (8.1) that permits rigorous data analysis.

This QAPP is reviewed by the TCEQ to help ensure that data generated for the purposes described above are scientifically valid and legally defensible. This process will insure that all data submitted to the TCEQ Regulatory Activities and Compliance System (TRACS) database have been collected and analyzed in a way that guarantees its reliability and therefore, can be used in TMDL development, stream standards modifications, permit decisions, and water quality assessments.

A6: Project/Task Description

This project will sample approximately 45 sites within the Mission-Aransas and East Flats areas of the South Texas coast. Within each of the two study areas, the stratified random method of hexagonal tessellation, developed by the USEPA EMAP program, will be used to select our sampling locations. This will ensure even, yet random selection of sampling sites. TCEQ Station Location requests will be submitted so that each site has a TRACS station ID. Core EMAP seagrass indicators will be measured along with additional parameters that have been identified as a result of recent research activities (Table 1). This effort will require the development of a detailed bathymetric base map in digital form using Geographic Information System (GIS) software.

Sampling sites will be sampled in winter (December/January) 2004/2005 and late summer (August/September) 2005. Each site will be marked with a PVC pole driven one meter into the sediments.

The major activities and products of the proposed research include site selection, intensive sampling over both temporal and spatial time scales, GIS and probability based design development, presentation of data on a dedicated Web site, and the production of a final report and peer-reviewed manuscripts (Table 2). Specific visual products of this monitoring effort include:

- synoptic views of water quality and seagrass measurements using GIS visualization and interpolation techniques
- temporal variations in seagrass distribution and abundance as a function of changes in physicochemical parameters on seasonal time scales

For further project details, refer to the Work Plan in Appendix A.

Table 1. Core EMAP coastal indicators to be measured at each permanent site. Parameter codes will be created for parameters not currently in the TRACS database.

| Parameter Name | Measurement Units | Matrix | Parameter Code | Method |
|---|--------------------------------|------------|----------------|----------------|
| Water Quality | | | | |
| Dissolved oxygen | mg L ⁻¹ | water | 00300 | YSI sonde |
| Conductivity | µmhos cm ⁻¹ | water | 00094 | YSI sonde |
| Salinity | ppt | water | 00480 | YSI sonde |
| Temperature | °C | water | 00010 | YSI sonde |
| NH ₄ ⁺ | µM | water | | UTMSI SOP 0201 |
| NO ₂ ⁻ & NO ₃ ⁻ | µM | water | | UTMSI SOP 0201 |
| PO ₄ ³⁻ | µM | water | | UTMSI SOP 0201 |
| Chlorophyll <i>a</i> | µg L ⁻¹ | water | | ESS 150.1 |
| Total suspended solids (TSS) | mg L ⁻¹ | water | | EPA 160.2 |
| Light attenuation (<i>k</i>) | m ⁻¹ | water | | UTMSI SOP |
| Surface irradiance (%SI) | % | | | LI-190SA |
| Sediment Quality | | | | |
| SGS Clay | % dry wt | sediment | 82009 | UTMSI SOP |
| SGS Silt | % dry wt | sediment | 82008 | UTMSI SOP |
| SGS Sand | % dry wt | sediment | 89991 | UTMSI SOP |
| SGS Gravel | % dry wt | sediment | 80256 | UTMSI SOP |
| Total organic carbon | mg kg ⁻¹ | sediment | 81951 | UTMSI SOP |
| Pore water NH ₄ ⁺ | mg L ⁻¹ | sediment | P1004 | UTMSI SOP |
| Seagrass Light Response Indicators | | | | |
| Biomass (above- & below-ground) | g dry m ⁻² | vegetative | | UTMSI SOP |
| Root:shoot ratio | -- | vegetative | | UTMSI SOP |
| Leaf area index | m ² m ⁻² | vegetative | | UTMSI SOP |
| Blade width | mm | vegetative | | UTMSI SOP |
| Shoot density | shoots m ⁻² | vegetative | | UTMSI SOP |
| Chlorophyll fluorescence | ratio | vegetative | | UTMSI SOP |
| Species composition | -- | vegetative | | UTMSI SOP |
| Maximum depth limit | m | vegetative | | UTMSI SOP |
| Plant Nutrient Response Indicators | | | | |
| C:N:P blade ratios | -- | vegetative | | UTMSI SOP |
| Epiphytic algal biomass | g dry cm ⁻² (leaf) | vegetative | | UTMSI SOP |
| Epiphytic algal species composition | -- | vegetative | | UTMSI SOP |
| Drift macroalgal abundance | g dry m ⁻² | vegetative | | UTMSI SOP |
| Drift macroalgal composition | -- | vegetative | | UTMSI SOP |

Table 2. Project timeline and deliverables

| Activities | 2005 | | | | | | | | | | | | 2006 | | | | | |
|----------------------------|------|---|---|---|---|---|---|---|---|---|---|---|------|---|---|---|---|---|
| | J | F | M | A | M | J | J | A | S | O | N | D | J | F | M | A | M | J |
| Project Planning & QA/QC | | | | | | | | | | | | | | | | | | |
| Field Sampling | | | | | | | | | | | | | | | | | | |
| Sample Processing | | | | | | | | | | | | | | | | | | |
| Data Management & Analysis | | | | | | | | | | | | | | | | | | |
| Spatial Analysis | | | | | | | | | | | | | | | | | | |
| Quarterly Reports | | | | | | | | | | | | | | | | | | |
| Draft Report Preparation | | | | | | | | | | | | | | | | | | |
| Final Report | | | | | | | | | | | | | | | | | | |

Deliverables:

1. **Water quality data:** Temporal and spatial analysis of water quality parameters.
2. **Biological indicators of stress:** Morphological and physiological indicators of stress.
3. **Spatial Analysis:** Geospatial database of seagrass distribution and other measurements.
4. **Final Report Document**

A7: Quality Objectives and Criteria

In this project, data will be collected semiannually within two estuarine systems along the south Texas coast. Spatial and temporal data will be used to address the following objectives concerning seagrass beds along the Texas coast:

- Integrate monitoring of landscape indicators (bed morphology patterns, vegetation species, and disturbance features) from high-resolution photography with monitoring of seagrass plant and habitat parameters (biomass, species composition, water and sediment nutrients, etc.)
- Continue collection of critical seagrass and environmental parameters in Redfish Bay through August/September 2005.
- Conduct geostatistical analyses on spatial and temporal scales to identify key parameters and appropriate scales of measurement for long-term monitoring.

Relevant data quality parameters such as sensitivity, precision, and expected range are listed in Table 3. Three replicates (n=3) will be collected and analyzed for each parameter at each of the sampling locations on each sampling date. A mean value [\pm 95% C.L. or \pm SE] will be calculated from the three replicate samples. Data from this project will not be used for TCEQ 305(b) assessments.

The conclusions of the project will be based on scientifically sound interpretations of the database. To achieve this end, and as required by EPA and TCEQ for all monitoring and measurement programs, objectives must be established for data quality based on the proposed uses of the data. The primary purpose of the QA program is to maximize the probability that the resulting data will meet or exceed the data quality objectives (DQOs) specified for the project. DQOs established for this project, however, are based on control of the measurement system because error bounds cannot, at present, be established for end use of indicator response data. As a consequence, management decisions balancing the cost of higher quality data against program objectives are not presently possible. As data are accumulated on indicators and the error rates associated with them are established, end use DQOs can be established for determining acceptable data quality to meet pre-established program objectives.

Completeness is defined as “a measure of the amount of data collected from a measurement process compared to the amount that was expected to be obtained under the conditions of measurement.”(Stanley and Verner 1985) An aspect of completeness that can be expressed for all data types is the amount of valid data (i.e., not associated with some criteria of potential unacceptability) collected. A criteria ranging from 75 to 90 percent valid data from a given measurement process is suggested as being reasonable for this project. As data are compiled for the various indicators, more realistic criteria for completeness can be developed.

Table 3. Data sensitivity, precision, and expected range for parameters to be measured as proposed in this R-EMAP study.

| Parameter | Units | Sensitivity | Precision | Expected Range |
|---|--------------------------------|-------------|-----------|----------------|
| Water Quality | | | | |
| Dissolved oxygen | mg L ⁻¹ | 0 | ±0.01 | 0-10 |
| Conductivity | mS cm ⁻¹ | 0 | ±0.001 | 0-100 |
| Salinity | ppt | 0.1 | ±0.01 | 10-55 |
| Temperature | °C | -5.0 | ±0.01 | 10-33 |
| NH ₄ ⁺ | μM | 0.1 | ±0.05 | 0-20 |
| NO ₃ ⁻ , NO ₂ ⁻ | μM | 0.05 | ±0.5 | 0-20 |
| PO ₄ ⁻³ | μM | 0.05 | ±0.3 | 0-5 |
| Chlorophyll <i>a</i> | μg L ⁻¹ | * | ±5% | 1-50 |
| Total suspended solids (TSS) | mgL ⁻¹ | 0.01 | ±0.5 | 0-50 |
| Light attenuation (<i>k</i>) | m ⁻¹ | -- | ±0.1 | 0.2-4.0 |
| Surface irradiance (%SI) | % | 0.1 | ±7 | 5-60 |
| Sediment Quality | | | | |
| Sediment grain size | phi | 14 | -- | -4-14 |
| Total organic carbon | mg kg ⁻¹ | -- | -- | 0-5 |
| Pore water NH ₄ ⁺ | mg L ⁻¹ | 0.1 | ±0.05 | 25-600 |
| Seagrass Light Responses | | | | |
| Biomass (above & below) | g dry m ⁻² | 0.001 | ±0.001 | 20-200 |
| Root:shoot ratio | -- | -- | ±0.01 | 0.5-8 |
| Leaf area index | m ² m ⁻² | 0.0001 | ±0.0001 | 0-10 |
| Blade width | mm | 1.0 | 0.5 | 4-8 |
| Shoot density | shoots m ⁻² | 40 | ±2% | 5000-9000 |
| Chlorophyll fluorescence | ratio | 0.1 | 0.1 | 0.2-0.8 |
| Maximum depth limit | m | 4.0 | 0.1 | 0.5-2.0 |
| Plant Nutrient Responses | | | | |
| C:N:P blade ratios | -- | -- | ±5% | 600:25:1 |
| Epiphytic algal biomass | g dry cm ⁻² (leaf) | 0.0001 | ±0.0001 | 0.1-100 |
| Drift macroalgal abundance | g dry m ⁻² | 0.0001 | ±0.0001 | 0.1-100 |

*Depends on volume of water filtered.

Bias, defined as “systematic or persistent distortion of a measurement process that causes errors in one direction” (Kirchner 1983; Hunt and Wilson 1986; Taylor 1987) may originate from calibration errors, sample contamination, or unaccounted for interference. Regular instrument calibration, proper cleaning of sampling and laboratory containers and use of gloves when handling and processing samples will minimize bias. Bias for nutrient analyses is tested regularly by analyzing both blind standards and standards of known values. Standard curves are prepared prior to processing each dataset and when using new chemicals. (Only standard curves with greater than 98% accuracy are used.) Precision, defined as the degree of mutual agreement among individual measurements (Kirchner 1983; Hunt and

Wilson 1986; Taylor 1987), represents an estimate of random error. Collectively, bias and precision provide an estimate of total error or uncertainty associated with an individual measured value. Bias and precision goals may not be definable for all parameters due to the nature of the measurement type because “true” or expected values do not exist for some measurement parameters (Table 3).

Sensitivity is defined as “the capability of a method or instrument to discriminate between measurement responses representing different levels of the variable of interest” (Kirchner 1983; Hunt and Wilson 1986; Taylor 1987). Sensitivity levels for each measurement are presented in Table 3.

Representativeness is defined as “the degree to which the data accurately and precisely represent a characteristic of a population parameter, variation of a property, a process characteristic, or an operational condition.”(Stanley and Verner 1985) Representativeness applies to the location of sampling or monitoring sites, the collection of samples or field measurements, the analysis of those samples, and the types of samples being used to evaluate various aspects of data quality. The sampling locations and program design in the R-EMAP Region 6 Project provide for representative seagrass population estimates along the south Texas coast. The proposed sampling design combines the strengths of systematic and random sampling with an understanding of estuarine systems to collect data that will provide estimates of seagrass status in Texas.

Comparability is defined as “the confidence with which one data set can be compared to another”(Stanley and Verner 1985). Comparability of reporting units and calculations, database management processes, and interpretative procedures must be assured if the overall goals of this project are to be realized. The comparability of the data produced is predetermined by commitment of the staff to use only approved procedures as described in this QAPP. Comparability is also improved by reporting data in standardized units, by using accepted rules for rounding figures, and by reporting data in a standard format as specified in the Data Management Plan (Appendix D). This project and the R-EMAP Region 6 Project that preceded it will generate a high level of documentation for the above topics to ensure that future efforts will be comparable. For example, both field and laboratory methods are described in full detail in manuals that will be made available to all field personnel and analytical laboratories. Field crews will undergo intensive training prior to the start of fieldwork. Finally, the sampling design for the project has been made flexible enough to allow for analytical adjustments, when necessary, to ensure data comparability.

A8: Special Training/Certification

UTMSI has been designated as the principal laboratory for this project, and will therefore provide oversight and implementation support for all activities. The UT Center for Research in Water Resources (UTCRWR) will provide technical support for spatial analysis of the data using ArcView and ArcInfo Geographical Information Systems (GIS). Additionally, personnel involved in the selection of sampling sites and GPS data collection will have appropriate training in the collection of spatial data.

Proper training of field personnel represents a critical aspect of quality control. Field technicians are trained to conduct a wide variety of activities using standardized protocols to ensure comparability in data collection. The field crew consists of a boat captain, the Project Officer, and a minimum of one technician. All are employees of UTMSI and under the supervision of the Project Directors. Members of this group will remain as consistent as possible for the duration of the project. Crewmembers generally are required to hold B.S. degrees and, preferably, a minimum of one year's experience. The captain will be a qualified and experienced boat handler. At UTMSI, all small boat captains are required to enroll in a U.S. Coast Guard Auxiliary course and pass both written and practical exams to operate an outboard motor vessel.

Laboratory personnel will undergo laboratory-specific training, which requires them to thoroughly review all methodologies and safety procedures prior to working in the laboratory. During the training, personnel will be shown each laboratory SOP by another experienced personnel. All laboratory procedures will undergo "practice runs" prior to analysis of actual project samples. Lab personnel are required to take a hazard communication course (OH 101) and a hazardous waste management course (OH 202) from the Environmental Health and Safety Department at UT.

A9: Documents and Records

All documents and records associated with this project will be permanently stored in the UTMSI laboratory of Dr. Ken Dunton. Copies of the QAPP and Corrective Action Documents will also be stored at CBBEP (Table 4).

Data collected in the field will be entered onto a pre-printed data form within a waterproof bound field notebook. Notes will be taken regarding weather conditions, personnel involved, sampling duties and summary of samples collected. We will also note any deviations from the sampling procedures or design and any problems (e.g. difficulty in gathering specimens) or unusual occurrences. The Project Officer will complete a formal fieldwork write-up in ink immediately following each field expedition. These write-ups will be kept permanently in the laboratory of Dr. Kenneth Dunton at UTMSI (Table 4).

Pre-printed spreadsheets will be used to record results of analyses as samples are processed in the lab. Data recorded on these Laboratory Data Reports will be entered into an Excel spreadsheet and the appropriate calculations performed. Each data sheet will be dated and signed by the person who collected the data in the field, processed the sample in the lab or entered the data on the computer. Data entered on computer spreadsheets will be verified to ensure accuracy of data transcription and calculations by visually proofing for entry errors and by periodic manual checks of computer calculations. Data spreadsheets and analytical results will be maintained on a computer hard drive as well as copied onto compact discs that will be retained permanently. Final data sheets will be printed out and put into a project binder that will be stored permanently in the laboratory.

Data that will contribute to the SWQM portion of the TRACS database will be entered in the appropriate ASCII file format and electronically transmitted to the Project Manager for submission to the database. A paper copy of a Chain of Custody Record (Appendix C) will be mailed to the Project Manager to document receipt of the data set.

The Project Officer will utilize a checklist to document Chain-of-Custody of each sample. The checklist will track each sample from its collection to its processing and entry into the computer. The checklist will include notes reflecting any anomalies in the samples (e.g. damaged samples) and reasons for flagging samples (e.g. contamination). A logbook with all calibrations and standard information will be kept by the Project Officer to document generation of Quality Control samples. The Chain-of-Custody checklists and calibration and standards logbook will be permanently stored in the UTMSI laboratory.

Table 4. Location and form of documentation records associated with this project.

| Document/Record | Location | Retention Time | Form |
|--|-----------------|-----------------------|-------------|
| QAPP, amendments, and appendices | CBBEP/UTMSI | Permanently | Paper |
| QAPP distribution documentation | UTMSI | Permanently | Paper |
| Field notebooks and Field Data Sheets | UTMSI | Permanently | Paper |
| Field equipment calibration and maintenance logs | UTMSI | Permanently | Paper |
| Chain of Custody Records | UTMSI | Permanently | Paper |
| Field SOPs | UTMSI | Permanently | Paper |
| Laboratory QA Manuals | UTMSI | Permanently | Paper |
| Laboratory SOPs | UTMSI | Permanently | Paper/CD |
| Laboratory Data Reports | UTMSI | Permanently | Paper |
| Data spreadsheets and analytical results | UTMSI | Permanently | CD |
| Corrective Action Documentation | CBBEP/UTMSI | Permanently | Paper |

B1: Sampling Process Design (Experimental Design)

Our sampling protocol utilizes a stratified-random approach to locate permanent monitoring sites within the Mission-Aransas and East Flats study areas. The presence of substantial seagrass meadows in both areas ensures that seagrasses will be well represented in the sampling design and that no particular portion of the sampling area is favored more than another. The areas delineated in both estuaries are based on the generalized distribution of existing and historical seagrass beds over the last 50 years. Stratified-random sampling in both systems will provide the opportunity to assess seagrass responses to variations in water quality parameters and physical disturbance.

To ensure even, yet random selection of sampling sites (i.e. to prevent location bias), we will use the stratified-random method of hexagonal tessellation, developed by the USEPA EMAP program. Study areas are divided into hexagonal subunits, each with an area of 1 km². One location will then be randomly selected as a sample site from within each hexagonal sub-unit. These randomly chosen permanent sites will be located in the field using a Global Positioning System (GPS) which is accurate to ± 5 m in south Texas and placed on a digital bathymetric map in GIS. The sites will be permanently marked by driving a PVC rod into the sediments. If the randomly chosen site is inaccessible (e.g. dredge location, a small island not on map, near or at a fishing house), another randomly chosen location will be used (10 potential “back-up” sites will be available for each of the hexagonal subunits if necessary). Deviations from the sampling plan, including utilization of “back-up” sites, will be noted in field notes.

At the sampling sites, a rapid visual assessment technique developed early in the 20th century by plant sociologist Braun-Blanquet will be used to assess the abundance of seagrass and macroalgae. At each site a 50-m transect will be established by extending a meter tape along the bottom in an up-current direction from the marker rod. Ten quadrats (0.25 m² each) will be placed along each transect at pre-determined random distances from one of the marker rods. A new set of random sampling positions are chosen before each visit to a site. Each quadrat is examined using SCUBA. All seagrass species occurring in the quadrat are listed, and a score based on the cover of the species in that quadrat is assigned (Table 5). Cover will be defined as the fraction of the total quadrat area that is obscured by a particular species when viewed from directly above. From the observations of cover in each quadrat at a site, three statistics will be computed for each species: density, abundance and frequency.

Table 5. Braun-Blanquet abundance scores (S). Each seagrass species will be scored in each quadrat according to this scale.

| S | Interpretation |
|-----|--|
| 0 | Species absent from quadrat |
| 0.1 | Species represented by a solitary short shoot, < 5 % cover |
| 0.5 | Species represented by a few (< 5%) short shoots, < 5% cover |
| 1 | Species represented by a many (> 5%) short shoots, < 5% cover |
| 2 | Species represented by many (> 5%) short shoots 5%-25% cover |
| 3 | Species represented by many (> 5) short shoots, 25%-50% cover |
| 4 | Species represented by many (> 5) short shoots, 50%-75% cover |
| 5 | Species represented by many (> 5) short shoots, 75%-100% cover |

Slightly different methods are used to ensure an unbiased placement of sampling quadrats in the three monitoring programs. In this study, we propose to adapt the procedures used in the FKNMS program in which the 50 m transect is placed in a north-south direction at each site.

At each site we propose to collect replicate samples (n=3; e.g. three-way field splits) of all media that require analytical analysis. An exception is grain size analyses that will be performed during the initial and final sampling only. These replicate analyses will permit the use of statistical analyses over the duration of this study. Because the project's overall goal is to determine which sampling parameters are necessary for assessing seagrass health along the south Texas coast, each parameter to be measured is assumed to be "critical." Classification as "non-critical" can only be determined following termination of the project sampling and statistical analysis of data.

Data collected during this project will be incorporated into a geospatial database in various GIS layers for assessment and statistical analyses. These data layers will include seagrass distribution and measurements from a variety of indicators. Since parameters are clearly linked to an underlying ecological process, geostatistical analyses can be used effectively to evaluate the power and reliability of a given indicator.

B2: Sampling Methods

At each site, all water samples will be collected in acid-washed, pre-labeled polyethylene bottles. Three replicates will be taken for each of the following measurements: inorganic nitrogen (NH_4^+ and $\text{NO}_3^- + \text{NO}_2^-$), phosphate (PO_4^{3-}), total suspended solids (TSS) and chlorophyll *a*. Water samples for NH_4^+ and $\text{NO}_3^- + \text{NO}_2^-$ will be collected in 125 ml bottles; PO_4^{3-} and chlorophyll *a* samples will be concurrently collected in 500 ml bottles, and TSS will be collected in 1 L bottles. Samples for sediment grain size, total organic carbon and pore water NH_4^+ will be collected and placed in separate, sterile Whirlpak bags. All samples will then be placed on ice for transport to the laboratory. Dissolved oxygen, conductivity, salinity, and temperature will be measured in the field using the YSI 600XLM-Sonde.

Upon return to the laboratory, chlorophyll *a* measurements will be performed immediately. Samples to be analyzed for inorganic nitrogen, total inorganic carbon, pore water NH_4^+ , TSS, and phosphate will be frozen for later processing (Parsons et al. 1984). The holding time will not exceed 30 days, with analyses typically occurring within 10 days of collection.

Samples for sediment grain size determination will be stored at 4°C until they can be processed.

Replicate biomass cores will be used for estimates of above- and below-ground biomass, root:shoot ratio, leaf area index, blade width, and shoot density. A 15 cm diameter corer will be used to sample *Thalassia*, and a 9 cm diameter corer will be used to sample *Halodule*, *Syringodium*, *Ruppia*, and *Halophila*. Samples of each species present will be collected at each site. Species presence (i.e. seagrass species composition) will be determined by visual *in situ* analysis of plants observed within a 25 m radius of each site. Samples will be placed in pre-labeled Ziploc bags and immediately placed on ice. Biomass samples will be refrigerated until they can be processed. Processing of all biomass samples will be completed within 30 days of their collection.

The variation in water depth will be estimated from measurements of depth made on each visit to a sampling site. The maximum depth for seagrasses at a site will be determined from depth measurements made at the edge of continuous meadows closest to the site. Aerial reconnaissance will be used to facilitate the location of meadow edges in each sampling area.

Estimates of algal epiphyte biomass and composition will be made from separate leaf samples of entire shoots taken directly adjacent to the biomass cores. Drift macroalgal abundance and composition will be determined from the collection of all algal material within 0.25 m² quadrats placed randomly on the seabed. Material from each triplicate is placed in sealed plastic bags and then transported to the laboratory in cooled containers. Upon arrival at the laboratory, the samples will be frozen until processing commences. Processing will begin as soon as possible and will be completed within 30 days of sample collection.

Additional seagrass blade of all species will be collected and put into Ziploc bags for determination of C:N:P ratios. The samples will be placed on ice for transport to the laboratory, where they will be frozen immediately upon arrival. Upon completion of processing the biomass samples, the seagrass blades will be thawed, dried, and prepared for elemental analysis. Analysis will be completed within 90 days of collection.

The percent surface irradiance (% SI) and the diffuse light attenuation coefficient (k) will be calculated from simultaneous measurements of surface and underwater irradiance. Measurements of photosynthetically active radiation (PAR = ca. 400 to 700 nm wavelength) will be collected on the surface using an LI-190SA quantum-sensor that provides input to a LI-1000 datalogger (LI-COR Inc., Lincoln, Nebraska, USA). Underwater measurements will be measured simultaneously at two separate depths using a LI-192SA quantum sensor. Estimates of % SI and k will be based on four replicate determinations of instantaneous PAR collected by surface and underwater sensors and recorded by the LI-1000.

In situ measurements of chlorophyll fluorescence will be made using the Diving-PAM, a submersible version of the portable Mini-PAM (Pulse-Amplitude-Modulation, Walz, Germany). A custom-built cuvette, positioned parallel to the blade (~2-6 cm above the sheath), holds the fiber optic light probe at a uniform 45° from the leaf surface during each trial. Steady state measurements of $\Delta F/F_m'$ will be made on a 4-6 replicate blade tissues from each site.

Equipment failures will be handled directly by the Project Director. Sampling problems noticed in the field will be addressed with corrective actions (e.g. re-sampling and a strong emphasis on sampling protocol). The Project Officer will be responsible for implementing, documenting and determining the effectiveness of corrective actions both in the field and in the laboratory. If problems are not detected while in the field, the Project Director will assess if re-sampling is necessary. If so, re-sampling will occur at the earliest possible date, and if not, the sampling problem will be noted.

When possible, samples will be preserved and archived in the laboratory at UTMSI. Dried biomass samples will be sealed in foil and stored in boxes. Dried tissues for C:N:P analysis will be stored in glass vials. Unused portions of all tissue samples will be stored indefinitely to permit future verification of measurements as necessary.

B3: Sample Handling and Custody

Bottles and bags for holding samples will be labeled in the lab prior to field collection. Standard labeling procedures will be followed; each bottle is labeled with waterproof labeling tape prior to field sampling. Each label will contain: sample type, station number, distance along transect, replicate number and date. To ensure that all samples are obtained, a standardized checklist of samples will be taken into the field (Appendix B). All samples will be placed on ice in darkness immediately upon collection in the field. Transport time to the laboratory will not exceed 6-8 hours. All samples will be stored in one designated location and remain in the custody of the Project Officer. Samples will be processed within the limits of the appropriate holding time to ensure the validity of all measurements. The Project Officer will utilize a checklist to document Chain-of-Custody of the samples in any instance in which possession changes between project personnel (Appendix C). The checklist will track each sample from its collection to when it is processed in the laboratory to when analytical results are entered into the computer. The checklist will include notes documenting any anomalies in the samples (e.g. damaged samples) and reasons for flagging samples (e.g. contamination). Since the Project Officer will oversee all sample collection, storage, analysis, and data entry at UTMSI, custody changes will be infrequent.

B4: Analytical Methods

The analytical methods used in this project are listed in Table 1. Most of the methods are UTMSI SOPs that have been used within the laboratory of the UTMSI Project Director in the past. Copies of the UTMSI SOPs are available for review by the CBBEP, TCEQ, and EPA

Ammonium, nitrate + nitrite, and phosphate water samples will be immediately frozen upon return to the laboratory (Parsons et al. 1984). Concentrations of NH_4^+ will be determined within 7 days using a standard colorimetric technique following the method of Parsons et al. (1984). Within 1-2 days of collection, concentrations of $\text{NO}_2^- + \text{NO}_3^-$ will be determined colorimetrically after cadmium reduction (Parsons et al. 1984). Water column PO_4^{3-} concentrations will be measured by adding a reagent containing molybdic acid, ascorbic acid, and trivalent antimony. Phosphate reacts with the reagent and the resulting complex is reduced to a blue solution measured at 885 nm on a spectrophotometer (Parsons et al. 1984).

Water column chlorophyll *a* levels will be determined by filtering water samples of known volume onto cellulose nitrate filters, and then extracting with 90% acetone (Parsons et al. 1984). Cellulose nitrate filters are used because they dissolve easily in acetone. Samples will be processed immediately upon return to the lab. TSS water samples will be frozen upon return to the lab. Total suspended solids will be determined by filtering a 1 L seawater sample onto a pre-weighed glass fiber filter. The sample filter is dried at 60°C to a constant weight.

Sediment grain size will be determined following the methods of Folk (1964). Percent contribution by weight is measured for four components: rubble, sand, silt, and clay. A 20-ml sediment sample is mixed with 50 ml of hydrogen peroxide and 75 ml of de-ionized water to digest organic material in the sample. The sample is wet sieved through a 62 μm mesh stainless steel screen using a vacuum pump and a Millipore Hydrosol SST filter holder to separate rubble and sand from silt and clay. After drying, the rubble and sand are separated on a 125 μm screen. The silt and clay fractions are measured using pipette analysis. Briefly, the settling velocity will be used to classify the particles and to determine the percent composition of each fraction, based on weight.

To determine total organic carbon, sediment samples will be dried to constant weight (at 60° C), cooled, weighed and combusted at 500° C for 3 hours to calculate the percent organic material present. Pore water NH_4^+ is measured by centrifuging (5,000 x g for 5-10 min) the thawed sediments and analyzing the supernatant for NH_4^+ following Parsons et al. (1984).

Biomass samples will be sieved within 2-3 days upon returning from the field. To determine above and below ground biomass, first sediment is gently washed from plants and epiphytes are removed. After sieving, plants will be refrigerated until they can be sorted into above- and below-ground components for each species. Sorting will occur within a week of sieving. Aboveground components are separated into leaves (including sheath material) and floral parts, while belowground tissues include roots and rhizome materials. Dead plant

material is discarded and the live tissues dried to a constant weight (60 °C) and weighed to the nearest milligram. The biomass values for above- and below-ground biomass will be used to calculate a root: shoot ratio.

Shoot density will be determined by counting the number of shoots in replicate cores of known area and scaling to appropriate units (shoots m⁻²). The average number of leaves per shoot will also be determined from core samples sorted in the lab. Leaf area index is calculated as a product of blade width measurements, blade length, shoot density, and the average number of leaves on each shoot. Blade width of the leaves collected from the aboveground portion of core samples are measured to the closest 0.5 mm.

Leaf tissues sorted from biomass cores will be used for C:N:P determination. Newly formed leaves (the youngest leaf in a shoot bundle) will be gently scraped and rinsed in tap water to remove algal and faunal epiphytes. These rinsed samples will be dried to a constant weight at 60 °C and homogenized by grinding to a fine powder using a mortar and pestle. Total C and N contents in the leaf tissues will be determined from duplicate subsamples of each sample by oxidation in a Carlo Erba model EA 1109 CHN elemental analyzer). P content will be measured with a modification of the method of Solorzano and Sharp (1980) as described by Fourqurean et al. (1992). Molar C:P, C:N, and N:P ratios are then calculated for evaluation of temporal and spatial trends.

Estimates of algal epiphyte biomass will be made from separate leaf samples of entire shoots taken directly adjacent to the biomass cores. In the laboratory epiphytes are separated from the leaf surface by scraping a constant area with a scalpel. Scraped material is then collected and retained on Whatman GF/C filters for biomass determination. This procedure removes >90% of the epiphytic algae at low algal densities. Algal epiphyte biomass will be expressed as a function of both the surface area and biomass of seagrass tissue scraped. The species composition of epiphytes (including the relative percentage of algal and faunal material) will be determined from microscopic analysis of scraped subsamples.

Drift macroalgal abundance will be determined from the collection of all algal material within 0.25 m² quadrats placed randomly on the seabed. Material from each replicate is placed in sealed plastic bags and then transported to the laboratory in cooled containers. Samples from each quadrat are sorted and identified to species. Tissues from individual species are dried at 60° C to a constant weight, weighed, and archived.

Failures in the analytical procedures or equipment will be handled directly by the Project Director. Problems noticed immediately will result in re-processing if sufficient sample remains. If re-processing is not possible, the lost sample will be noted and corrective action taken (e.g. reviewing lab protocol, remixing chemicals, re-cleaning glassware, running additional standards or blanks). The Project Officer will be responsible for implementing, documenting and determining the effectiveness of corrective actions. If any approved procedures are violated and knowingly yield invalid or even suspect data, the flawed data will not be submitted for entry into the TRACS database.

B5. Quality Control

Data quality will be ensured during the project by the use of standard curves and blanks (NH_4^+ , $\text{NO}_3^- + \text{NO}_2^-$, PO_4^{-3} , chlorophyll *a*). Nutrient/chlorophyll deplete-water samples will be taken into the field on each sampling trip to serve as trip blanks. These trip blanks will go through the same analytical processes as the field collected samples to serve as a check for contamination or other procedural errors. If analysis of a trip blank yields anomalous output, then the integrity of the sampling process has been compromised and diagnostic procedures will be undertaken to eliminate further errors.

Standard curves will be established for NH_4^+ , $\text{NO}_3^- + \text{NO}_2^-$, PO_4^{-3} analyses utilizing a blank (nutrient deficient seawater) and at least three analytical standards of increasing concentration, covering the range of expected sample concentrations. Linearity of the standard curve ($R^2 \geq 0.995$) must be established prior to the analysis of samples (UTMSI SOP 0201). Blanks will be used during every analysis to auto-zero the spectrophotometer, and complete standard curves will be performed when new chemicals are used and prior to processing each data set. Select standards will be used during each analysis to check the assumption that the original standard curve continues to be valid. Analysis of standards should occur at the beginning of a sample set and after the last analytical sample. If blanks or initial standards deviate from known concentrations, sample processing will be suspended and procedures will be undertaken to determine the source of error (i.e. troubleshoot to determine if the error is due to contamination, bad chemicals, poor technique, a technical error or instrument failure). If the standards run following the last analytical sample deviate from the original values obtained, the last sample analyzed before the check sample (standard) that failed the control limit criteria should then be reanalyzed. If the relative percent difference (RPD) between the results of this reanalysis and the original analysis exceeds 20 percent, the instrument is assumed to have been out of calibration during the original analysis and the earlier data will be flagged or replaced. If possible, reanalysis of samples should progress in reverse order until it is determined that there is <20 RPD between initial and reanalysis results. If it is not possible or feasible to perform reanalysis of samples, all earlier data (i.e., since the last successful standard control check) will be flagged.

Irradiance values will be compared with other sensors calibrated against a National Institute of Standards and Technology (NIST) standard. YSI-sonde measurements of salinity, temperature, dissolved oxygen, and pH will be validated with other instruments measuring the same parameters. Comparison to other instruments will occur prior to each sampling period.

C:N ratios will be compared against a known standard (chitin) for every 10 samples processed. Molar C:P and N:P ratios are calculated for evaluation of temporal and spatial trends. NIST orchard leaves will be used as a primary standard to assess the accuracy of our procedures for measuring P. A mean value [$\pm 95\%$ C.L. or $\pm \text{SE} (n)$] will be provided from three replicate sample measurements collected quarterly over the entire period of study ($n=3$ for each parameter on each sample date).

B6: Instrument/Equipment Testing, Inspection and Maintenance

The UTMSI Laboratory is equipped to provide analytical support for physical, chemical, and biological analyses, has facilities to store and prepare samples, and has the appropriate instrumentation and trained staff to generate data of the required quality within the time period dictated by the project. Operations are conducted using good laboratory and field practices, including:

- A program of scheduled maintenance of analytical balances, microscopes, laboratory equipment, refractometer, light meter, GPS units, YSI sonde, and other instrumentation.
- Recording all analytical data in bound logbooks in ink.
- Monitoring and documenting the temperatures of cold storage areas and freezer units.
- Verifying the efficiency of fume hoods.
- Standardized labeling of all containers used in the laboratory with date prepared, contents, and initials of the individual who prepared the contents.
- Dating and safely storing all chemicals upon receipt. Chemicals are disposed of properly when the expiration date is reached.
- Using a laboratory information management system to track the location and status of any sample received for analysis.

All information regarding these laboratory and field practices is stored in the laboratory of Dr. Kenneth Dunton at UTMSI.

All laboratories at UTMSI are routinely inspected by Environmental Health and Safety personnel from UT-Austin to check adherence to strict University policies regarding chemical safety and handling. Laboratory personnel performing EMAP analyses will be well trained in good laboratory practices, including standard safety procedures. It is the responsibility of the Project Officer to ensure that safety training is mandatory for all laboratory personnel. The UTMSI laboratory maintains a current safety manual in compliance with the Occupational Safety and Health Administration (OSHA) and the equivalent state or local regulations. The safety manual is available to all laboratory personnel. Proper procedures for safe storage, handling and disposal of chemicals are followed at all times. Chemicals are treated as potential health hazards and good laboratory practices are implemented accordingly.

B7: Instrument/Equipment Calibration and Frequency

Analytical balances are calibrated before each use using a standard weight. At the onset of each field season, the membrane on the YSI sonde will be replaced, and the sonde will be calibrated according to the manufacturer's specifications. Calibration will be repeated in the lab prior to each sampling trip and the performance of the sonde will be monitored in the field. If, during a sampling trip, the charge on the sonde falls out of the range of 50 ± 25 units, or the percent dissolved oxygen readings in saturated solution are not within 2% of 100%, the sonde will be recalibrated on site.

LI-COR light sensors are re-calibrated annually to within $\pm 5\%$ of NIST standards by LI-COR, Inc. These checks are performed routinely and the results recorded in a log kept for each instrument. Routine calibration of pipettes occurs off-site by the pipette suppliers on a quarterly basis. The spectrophotometer will be calibrated on-site using standards every six months. A trained technician will calibrate the mass spectrometer. A calibration of weight percent is done everyday, and a mass calibration is performed every two weeks. Instrument calibration will be recorded in a logbook. Summary data documenting initial calibration and any events requiring recalibration and the corresponding recalibration data will be included with the analytical results.

B8: Inspection/Acceptance of Supplies and Consumables

The Project Officer will verify all supplies (e.g. bottles, bags) and consumables (e.g. reagents) with shipping/receiving documents to ensure accurate quantities and types. Damaged, opened or incorrect packages or contents will be reported to the shipping department and returned to the supplier for replacement. All problems with shipments will be immediately reported to the project director and the purchasing department so that all expenses are accounted. The Project Officer will document any problems with supplies. Following acceptance of supplies and consumables, the items will be stored in the lab.

B9: Non-direct Measurements

No non-direct measurement sources will be required for this project. Only data collected directly under this QAPP will be submitted to the TRACS database. All data collected under this QAPP will comply with all requirements of the project and the Surface Water Quality Monitoring QAPP.

B10: Data Management

All data generated during this project will be handled as outlined in the Data Management Plan (Appendix D). Data collected in the field will be recorded on pre-printed data sheets (Appendix B). The data collector will sign and date each data sheet. Upon return from the field, the Project Officer will enter these data into Microsoft Excel spreadsheets on a PC computer and sign and date the data sheet following each entry. The data will then be proofed by the QA Officer to detect and correct for data entry errors. This person will also sign and date the data sheets. Mean values and standard errors will be calculated. The data

will then be saved on the computer hard drive, backed up on a CD-ROM, and printed out as a hard copy, which will be put into a project specific binder. Backup files will be stored in a separate location from the main computer files in the laboratory at UTMSI. Data Review Checklists (Appendix E) will be completed for laboratory and field data to document completion of quality control procedures, data verification, and data validation. All data will be incorporated into a Geographical Information System (GIS) where it will be analyzed and interpolated using Geostatistical Analyst, an Arc/Info 8.1 extension. Following review by the QA Officer, Project Officer, and the Project Director, all data spreadsheets will be sent to the Project Manager for transmittal to TCEQ. All data that will enter the TRACS database will be submitted in the format outlined in Chapter 7 of the Data Management Reference Guide (TCEQ 2003).

C1: Assessments and Response Actions

The Project Director will conduct routine surveillance throughout the course of the study. The Project Officer will perform informal technical system audits to ensure compliance with the QAPP. Field data sheets, sampling methodology, and equipment calibration will be reviewed during each sampling period for conformance to standards outlined in the QAPP. Similar audits will take place during laboratory analyses to ensure the use of proper techniques, analysis of blanks and standards, and documentation of standard curves and sample handling. Any deviations from approved protocols will be immediately corrected and noted. The MDM&A Manager will analyze the data and identify inconsistencies and report them to the Project Manager. The Quality Assurance Officer will perform regular data quality audits and assessments. In the event of invalid data, the QA Officer will notify the Project Officer so that the appropriate response may be taken. Invalid or suspect data will not be included in the TRACS database.

C2: Reports to Management

Quarterly reports summarizing the status of field and laboratory work, data analyses performed, and budget status will be submitted to the CBBEP Project Manager. Quarterly reports will contain information regarding data quality problems, possible solutions, and needs for additional resources. A final report encompassing all data analyses, including GIS, and conclusions will be submitted to the CBBEP Project Manager. Project results will also be presented on a web site and at a scientific conference. All reports will be completed by the Project Officer and approved by both Project Directors.

D1: Data Review, Verification, and Validation

Review of data will occur at the field, laboratory, and database levels as outlined in Table 6. Data will be reviewed by the Project Officer, QA Officer, and Project Director following each sampling period. The Project Officer will check each sample in the field to make sure it is representative of the sampling location, in the correctly labeled container, and of the correct quantity. Deviations from the QAPP will be noted in sufficient detail in the laboratory field notebook and the affected samples will be flagged. During sample transport from the field to the laboratory, sample and storage containers will be evaluated by the

Project Officer for damage and contamination, and all problems will be noted. A checklist of each sample to be collected and processed will be completed before sampling occurs. Data will be validated by using QC checks (e.g. trip blanks and standards) and proper calibration of instruments and equipment. If problems occur, the data will be further validated by documenting corrective actions taken, samples affected, and effects of the actions on the validity of the data. During data recording and processing, mean values and standard errors will be calculated. Prior to statistical analyses, all appropriate assumptions will be tested (e.g. homogeneity of variance, normal distribution). Statistically significant differences in mean data values will require a 95% Confidence Level ($p < 0.05$).

Table 6. Data review, verification, and validation

| Data to be Verified | Field Task | Laboratory Task | Database (or Lead Organization Data Manager) Task |
|--|-------------------|------------------------|--|
| Sample documentation complete; samples labeled, sites identified | √ | √ | |
| Field QC samples collected for all analytes as prescribed in the TCEQ SWQM Procedures Manual | √ | | |
| Standards and reagents traceable | √ | √ | |
| Chain of custody complete/acceptable | √ | √ | |
| Sample preservation and handling acceptable | √ | √ | |
| Holding times not exceeded | √ | √ | |
| Collection, preparation and analysis techniques consistent with SOPs and QAPP | √ | √ | √ |
| Field documentation complete | √ | | |
| Instrument calibration data complete | √ | √ | |
| QC samples analyzed at required frequencies | √ | √ | √ |
| QC results meet performance and program specifications | √ | √ | √ |
| Results, calculations, transcriptions checked | √ | √ | |
| Nonconforming activities documented | √ | √ | √ |
| Outliers confirmed and documented; reasonableness check performed | | | √ |
| Data formatted correctly | | | √ |
| Depth reported correctly | | | √ |
| TCEQ ID number assigned | | | √ |
| Valid Parameter codes | | | √ |
| Source codes 1 and 2 and program code used correctly | | | √ |
| Absence of transcription error confirmed | √ | √ | √ |
| Absence of electronic submittal errors confirmed | √ | √ | √ |
| Sampling and analytical data gaps checked | √ | √ | √ |
| Field QC results attached to data review checklist | | | √ |
| Verified data log submitted | | | √ |

D2: Verification and Validation Methods

All data will be reviewed to ensure that they meet the project specifications. Data will be verified to make certain that they are representative of the samples that were analyzed and the locations at which the measurements were taken. Review and verification will be performed by the staff and management responsible for the generation and handling of the data. A cursory screening will occur as data are recorded or upon generation of the first meaningful data. In addition, quality control checks will be performed in the midst of processing to assess the general level of data quality. Comparison of sample values with those of blanks and standards will provide an indication of technical problems that could yield flawed data. This type of routine QC check permits early detection of errors so that appropriate actions can be taken to prevent transmission further along the data stream. Data will be reviewed at all phases of generation and management, including raw data, electronic data, hard copy output, and data on required forms.

The verification of data will consist of both self-assessments and peer review, depending on the nature of the task (Appendix E). The task manager will also perform a technical review to ensure that the data meet project specifications and that errors in transcription, calculation, and data input are identified. All data will be maintained and managed by the Project Officer. Because of the direct involvement in data collection, generation and management, the Project Officer will retain custody of the data until it is presented to the Project Director, QA Officer and CBBEP Project Manager on a quarterly basis for review. After review for verification and validation, the Project Officer will be formally informed of any discrepancies or problems with the data via written notification. Problems that can be rectified will be corrected. Corrections will be documented electronically or by initialing and dating the appropriate paperwork. Final decisions regarding potential data outliers and their acceptance or rejection will be made by the Project Director and will be based on (1) whether the data point is within the expected data range and (2) if the resultant data point is suspected to have been skewed due to technical or collection errors (i.e. poor data validation). Data will be verified (i.e. ensuring that conclusions can be correctly drawn) by comparison of values to expected ranges determined from previous studies in similar environments.

Following the data review, all verified and validated data will be given to the Project Manager for submittal to TCEQ for inclusion in the TRACS database. Results of data analysis will be synthesized and conveyed to data users through peer reviewed journal publications and presentations at scientific meetings.

D3: Reconciliation with User Requirements

Data will be reconciled with the DQOs by performing data analyses according to standardized procedures established by the EMAP program. Point data on species density will be used to produce continuous maps of the density of seagrass species, as well as maps of species richness. Krigging algorithms will be used to interpolate between the random point data and spatial analysis programs will be used to compute areas of seagrass coverage from these interpolated surfaces.

Two main types of seagrass health indicator variables can be identified at each station: Condition Indicators and Stressor Indicators. Condition Indicators are “characteristics of the environment that provide quantitative estimates of the state of ecological resources that are important to society,” and stressor indicators are “characteristics of the environment that are suspected to elicit a change in the state of ecological resources.” Statistical analyses are then implemented to relate the condition and stressor indicators.

Interpretation of condition and stressor indicator data may require characterization of sites using descriptive criteria for “impaired” sites or separation of sites into various classes of impairment based on quadrisection of indicator values. Least impaired sites would be defined as remotely located from potential sources of perturbation with little or no observed physical damage.

Transformation of indicator values may be required for site separation into “impairment classes,” because of assumptions of linear response and homoscedasticity. The coefficient of variation (CV) will be calculated for indicator values measured among least impaired sites for the entire study period and within seasons. Indicators with a low CV have the potential to yield a more sensitive multimetric index or multivariate response model than indicators with a high CV. Simple linear correlation will be used as a test for redundancy among the response indicators as a step in identifying those that provide the most unique information at the least cost.

There are several approaches that will be used for observing the relative strength of the response of each indicator to the combined effects of stressors. The development of response models for individual indicators may prove to be the most informative approach, allowing visualization of the response across a gradient of conditions. Stepwise discriminant analysis may be used in conjunction with classification of sites as “impaired” or “least impaired” to identify the indicators that contribute the most weight to a discriminant function. Canonical Correlation Analysis may also be used to observe the multivariate relationship and canonical loadings for each indicator in the relationship between all stressor and response indicators.

It is an implicit assumption in all these analyses that the random samples in adjacent hexagonal boxes are statistically independent, that is, the spatial placement of the boxes relative to one another has no bearing on the measurements made within them. Geostatistical analysis using GIS can be used to test the validity of this assumption by constructing a variogram measuring the range of spatial correlation among the sampled

measurements. Geostatistical analysis is also useful for interpolating gridded maps of estimated values of the measured indicators (and standard errors for those estimates), which may reveal spatial patterns in the data that observation of the point values would not reveal. This is particularly useful when there is a great deal of variability in measurements at any given sample point relative to the variability between sample points.

One goal of a future study may be the development of a summary statistic that combines a set of variables that reliably identifies the health of seagrass beds. One method of doing this is an Index of Biological Integrity (IBI), which is a weighted set of indicator values that provide a score of biological health. Gradations of the score from high to low would then be used to characterize the health of seagrass population. Such indicators need to be:

- Statistically sound in the sense that they discriminate between sampled conditions in a statistically significant manner.
- Scientifically sound in the sense that inclusion of variables in the IBI is empirically defensible.
- Reasonable in the sense that they may be used by resource staff to determine an IBI value following standardized protocol within a reasonable amount of time with limited resources. Statistical summarization techniques are helpful in forming such indices but common sense selection of reasonable variables should always take precedent.

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APPENDIX A. APPROVED WORK PLAN

**LANDSCAPE MONITORING AND BIOLOGICAL INDICATORS FOR SEAGRASS
CONSERVATION IN TEXAS COASTAL WATERS**

by

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**Project Period
29 April 2004 – 31 December 2005**

Approval Date: _____

**Coastal Bend Bays & Estuaries Program Executive Director: Ray Allen
www.CBBEP.org**

UT-Austin PROPOSAL NUMBER UTA04-316
10 February 2004

Approval page/distribution list

Project Name: Landscape Monitoring and Biological Indicators for Seagrass Conservation in Texas
Coastal Waters

Contract Number:

CBBEP Project Manager: _____ Date: _____

CBBEP Executive Director: _____ Date: _____

TCEQ Representative: _____ Date: _____

EPA Representative: _____ Date: _____

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ARTICLE I

PROJECT DESCRIPTION

Introduction

Recently, a Seagrass Habitat Monitoring program was proposed for Texas coastal waters in the Seagrass Conservation Plan (TPWD, 1999). This document, which describes many of the potential problems facing Texas seagrass habitats, recommended monitoring of key indices to detect ecosystem changes that occur before actual loss of seagrasses. Texas Parks & Wildlife (TPWD) and the Texas Commission on Environmental Quality (TCEQ) have since started a planning process with resource managers and coastal researchers in Texas to develop a statewide seagrass-monitoring plan based on sound scientific knowledge of the resource to link the dynamic variability in environmental parameters to changes in seagrass distribution on both temporal and spatial scales. The proposed project addresses the development of indicators and criteria that began under a R-EMAP project funded to Dr. Ken Dunton (2002-2004) but also integrates a critical landscape component proposed by Dr. Warren Pulich. The proposed project will address the effectiveness of various plant, water column, and landscape indicators to determine the optimum scales for both spatial and temporal sampling frequency that can be used to monitor seagrasses for management and conservation purposes. The project also includes a separate effort for the acquisition of high-resolution baseline images in areas defined as critical seagrass habitat.

State resource managers in TPWD, TGLO, and TCEQ, along with coastal research scientists, have recommended coastwise monitoring to assess the status of seagrass beds and to detect sub lethal impacts prior to actual plant losses (Seagrass Conservation Plan for Texas 1998). The Texas Seagrass Monitoring Plan (TSGMP 2003) has recently proposed a combination of intensive field surveys (plant scale) and landscape (bed scale) monitoring with color aerial photography to measure indicators of seagrass health and stress. Changes in species composition or grassed distributions, water clarity conditions, proliferation of algal blooms or epiphytes, and physico-mechanical impacts to grass beds, are typical indicators of stressed seagrass beds and degradation from dredging, urbanization, boating/ship traffic, or high nutrient loading. Seagrass dynamics at both the plant level (microscale) and landscape (bed scale) level are critical in interpreting the degree of stress from these diverse anthropogenic or natural disturbance factors.

Intensive field sampling is traditionally used to detect and quantitatively measure effects of specific factors related to seagrass stress or growth (Neckles ed. 1994). But, such microscale field sampling of plant indicators is very labor intensive and expensive. At the seagrass bed scale, landscape indicators and patterns should also reflect major causes of disturbances, including poor water quality, human or natural physical disturbances, and hydrologic conditions. High-resolution photography over wide areas would be very cost effective in identifying characteristic human or natural disturbances, and possibly water quality impacts (Dobson et al. 1995, Robbins 1997). If microscale measurements from field sampling were statistically correlated with specific landscape features and geomorphological patterns of beds evident in photography, it should be possible to extrapolate the extent of seagrass impacts over wide coastal areas.

Identifying the factors responsible for seagrass plant dynamics (i.e. causes of seagrass stress or growth) can be difficult, even from carefully measured plant scale indicators (e.g. plant biomass or root/shoot ratios). When landscape indicators are monitored (e.g. bed morphology or patchiness,

macroalgae, species composition, etc.), the same problem exists; and inferring the causes of landscape changes from these effects must be approached cautiously. Environmental conditions and ecological factors may exert positive or negative effects, either singularly or in combination. Consequently, identifying the responsible factors and their effects on the seagrass bed involves deciphering complex interactions through both site specific and landscape level measurements. Extrapolation from specific field site measurements over large seagrass landscape areas requires integration of both remote sensing and field sampling data through geostatistical analysis.

ARTICLE II

PROJECT OVERVIEW

This proposed project will identify the plant, water column, and landscape indicators that provide the most critical information on water quality criteria that is relevant to successful maintenance and growth of seagrasses. The project will also allow investigators to generate data to assess the relative value of various indicators with respect to cost, inherent variability on spatial and temporal scales, and effort. The project will focus on Redfish Bay in the Mission-Aransas estuarine system and utilize East Flats in the adjacent Nueces Estuary (Corpus Christi Bay) as a reference site. Both sites are located within the Coastal Bend Bays and Estuaries Program (CBBEP) study area. Redfish Bay and East Flats support extensive seagrass meadows that contain all five species of seagrasses common to the Texas coast (*Halodule wrightii*, *Ruppia maritima*, *Thalassia testudinum*, *Syringodium filiforme*, and *Halophila engelmannii*).

This study will validate a landscape analysis approach to seagrass monitoring and establish protocols for evaluating stress on seagrass systems from landscape-scale dynamics determined from aerial remote sensing data. Since landscape scale dynamics are best extrapolated from change analysis of wetland landscapes over reasonable time periods (Dobson et al. 1995; Heggem et al. 1999), we propose to compare seagrass landscapes at target sites over two time periods, and to correlate classified landscape features with discrete plant scale data or process measurements for the same time periods. Effects of plant ecosystem processes will be correlated with change dynamics of landscape features between beginning and end of annual growth cycles, or before and after an environmental disturbance. Statistical relationships between plant level data and landscape features and patterns would also provide the basis for deriving seagrass bed indicators of biological integrity.



Figure 1. A map of watersheds that drain into the Mission-Aransas, Nueces, and Laguna Madre estuaries of the Texas coast. The map was created from the USGS National Hydrology Dataset.

ARTICLE III

PROJECT OBJECTIVES

This proposed project includes several critical features:

This proposal will integrate monitoring of landscape indicators (bed morphology patterns, vegetation species, and disturbance features) for seagrass beds with monitoring of seagrass plant/habitat parameters (biomass, species composition, water and sediment nutrients, etc.) in the CBBEP study area. The lead PI for each component, Ken Dunton (KD) or Warren Pulich (WP) is listed following each objective.

- Integrate monitoring of landscape indicators (bed morphology patterns, vegetation species, and disturbance features) from high-resolution photography with monitoring of seagrass plant/habitat parameters (biomass, species composition, water and sediment nutrients, etc.) at three sites in the CBBEP study area. {WP and KD}
- Establish 7 - 8 priority target sites in the CBBEP study area and collect remotely-sensed data for long-term seagrass monitoring as recommended by the TSGMP (2003). {WP}
- Acquire true color aerial photography at 1:24000 and 1:9600 scales for the sites and archived as digital imagery in an electronic database. {WP}
- Develop a GIS tool to define landscape indices of biological integrity based on an analysis that combines landscape indicator data and field-based sampling data. {WP}
- Continue collection of critical seagrass and water column indicators in Redfish Bay in December 2004/January 2005 and August/September 2005. {KD}
- Expand monitoring to the diverse and productive seagrass meadows at East Flats. {KD}
- Conduct a gestatistical analyses of the spatial and temporal dataset to identify key parameters and scales of measurement for long-term monitoring. {KD}
- Address the question of scale in the interpretation of aerial imagery. {WP}

This proposed project will integrate and coordinate the efforts of various state and federal agencies, including the United States Geological Survey/National Wetlands Research Center, Texas Parks and Wildlife, the Texas Natural Resources Conservation Commission, the Texas General Land Office (TGLO) and local academic institutions. The PI's on this project will coordinate their activities with the Seagrass Monitoring Work Group, which was developed to facilitate and organize seagrass monitoring programs in Texas, and act as liaisons in linking this research effort with agency objectives.

ARTICLE IV

TASKS

The PERFORMING PARTY shall perform the following tasks:

- (a) Task 1. Quality Assurance Project Plan - Submit a written Quality Assurance Project Plan (QAPP) to the CBBEP Project Manager. Upon final review by the CBBEP Authorized Representative, the QAPP shall become part of this contract by reference. All project sampling, analysis, and reporting protocols will continue to meet or exceed the protocols identified in the *EPA National Coastal Assessment Program* (formally Coastal 2000) *QAPP and Field Manual*, or approved alternate methods.
- (b) Task 2A. Indicators and Monitoring –
- (c) Task 2B. Landscape Monitoring
- (d) Task 3. Analysis –
- (e) Task 4. Submit Results – Primary assistance will be provided to the CBBEP Project Manager in making the transfer of the comprehensive project results submitted to CBBEP.
- (f) Task 5. Report – The draft and final reports will be incorporated into a single comprehensive report for the entire project.

Task 2A. Indicators and Monitoring

Indicators

This project will sample a minimum of 30 sites within the Mission-Aransas estuary (Fig. 2). Core indicators will also be measured at East Flats sites in coordination with the remote sensing measurements at this landscape reference site. Within each study area, core EMAP seagrass indicators will be measured (Neckles, 1994) along with additional parameters that we have identified based on recent research activities (Table 1). This effort will require the development of a detailed bathymetric base map in digital form using Geographic Information System (GIS) software (Melancon et al., 1999).

Sampling Design and Site Selection

Our sampling protocol follows the procedures and standards established by Fourqurean et al. (2001) for the EPA sponsored seagrass status and trends monitoring project in the Florida Keys National Marine Sanctuary (<http://www.fiu.edu/~seagrass/>). A stratified-random approach was used to locate 30 permanent monitoring sites within the Mission-Aransas study area under the 2002-2004 EMAP program (Fig.2). The presence of substantial seagrass meadows in both areas ensures that seagrasses were well represented in the sampling design and that no particular portion of the sampling area is favored more than another (Volstad et al., 1995). The areas delineated in Redfish Bay are based on the generalized distribution of existing and historical seagrass beds over the last 50 years. Input from the Texas Seagrass Monitoring Work Group (consisting of officials from USGS-NWRC, USF&WS,

Table 1. Core coastal indicators to be measured at each permanent site in this study.

| Water Quality | Sediment Quality | Seagrass Light Response Indicators | Plant Nutrient Response Indicators |
|---|---|---|---|
| dissolved oxygen | grain size | biomass (above- & below-ground) | C:N:P blade ratios |
| conductivity, salinity, and temperature | total organic carbon | root:shoot ratio | epiphytic algal species composition and biomass |
| nutrients (NH ₄ ⁺ , NO ₃ ⁻ , NO ₂ ⁻ , PO ₄ ⁻³) | pore water NH ₄ ⁺ | leaf area index; blade width | drift macroalgal abundance/ composition |
| chlorophyll <i>a</i> | | shoot density | |
| total suspended solids (TSS) | | chlorophyll fluorescence | |
| light attenuation (<i>k</i>) | | species composition | |
| Surface irradiance (%SI) | | maximum depth limit | |

TPWD, TCEQ, TGLO) and scientists with EPA-ORD and EMAP were used to select the relevant biological and environmental parameters (indicators) and to conduct the necessary field sampling. QA/QC procedures developed for the R-EMAP study, which are necessary to ensure the necessary precision, accuracy and completeness of field and analytical procedures, will be provided to the CBBEP office.

Although seagrass beds in the Mission-Aransas (Redfish Bay) and Nueces (East Flats) estuarine systems are often noted as pristine habitats, Redfish Bay is subject to suspected non-point and/or point sources of inflows from adjacent watersheds. In the Redfish Bay area, Pulich et al. (1997) noted possible water quality problems and physical impacts from dredging, prop scarring, and storms. Consequently, random sampling in both systems provides the opportunity to assess seagrass response indicators to variations in water quality parameters and physical disturbance. The maximum depth limitation for seagrasses on the Texas coast is about 1.7 m based on seagrass minimum light requirements of 18-20% surface irradiance (Onuf, 1994; Dunton, 1996). Random site selection thus requires the use of digitized NOAA bathymetric charts (see Fig. 2).

It is important that the sites selected in both estuarine systems can be used in a probability-based design for geostatistical analyses, and are representative of the physiographic variability within each estuary based on existing data. All points within the landscape must have an equal probability of being sampled, and sampling effort be quasi-evenly distributed across the landscape. Pure random distribution of sampling points often leads to clumped and non-uniformly distributed data points. To insure even, but yet random selection of sampling sites, we used the stratified random method of hexagonal tessellation, developed by the USEPA's EMAP program, to locate our sampling locations under the R-EMAP program. The Redfish Bay study area, which is approximately 36 km², was divided into 30 hexagonal subunits. One random location was then chosen as a sample site from within each hexagonal sub-unit. The 30 randomly-chosen permanent sites are located in the field using a Global Positioning System (GPS) which is accurate to ± 5 m in south Texas and placed on a digital

bathymetric map in GIS. In concept, this task is similar to that carried out by the Texas Parks and Wildlife Department for principal Texas bays and estuaries (Fig. 3).

The permanent sites selected in Redfish Bay under the R-EMAP program, along with sites selected in East Flats, will be sampled in Winter 2004/05 and Summer 2005. This will ensure the collection of a continuous 3.0-yr year dataset for Redfish Bay. The sites will be marked with PVC poles driven one meter into the bottom. The measurements made at the permanent sites will be used to rapidly estimate the spatial extent and cover of benthic macrophytes in the study area (see following section).



Figure 2. Proposed study area (enclosed by black line) in Mission Aransas estuary (Redfish Bay).

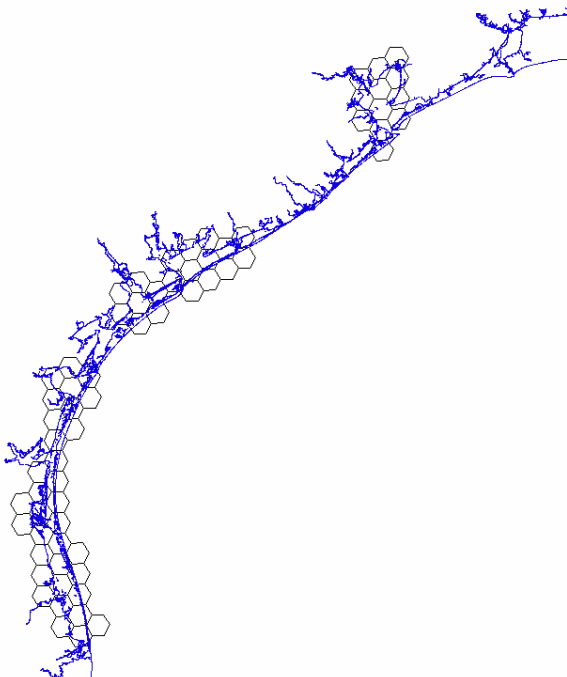


Figure 3. R-EMAP hexagonal sampling units defined by TPWD for Texas bays and estuaries.

Field Procedures, Frequency of Sampling, and Replication

Frequency, Abundance, and Density of Seagrasses and Macroalgae (procedures adapted from Fourqurean et al. (2001))

At both permanent sites, a rapid visual assessment technique developed early in the 20th century by plant sociologist Braun-Blanquet (Braun-Blanquet 1972) will be used to assess the abundance of seagrass and macroalgae. This method is used in the EPA sponsored seagrass status and trends monitoring project in the Florida Keys National Marine Sanctuary (FKNMS). It is very quick, requiring only minutes at each sampling site, yet is robust and highly repeatable, thereby minimizing among-observer differences, and has recently been applied to seagrass research. At each permanent seagrass monitoring site, a 50-m-long transect will be established at the beginning of the study period by driving steel rods into the substratum at both ends of the transect. At each survey site a 50-m transect will be established by extending a meter tape along the bottom in an up-current direction. Ten quadrats (0.25 m²) will be placed along each transect at pre-determined random distances from one of the marker rods. A new set of random sampling positions are chosen before each visit to a site. Each quadrat is examined using SCUBA. All seagrass species occurring in the quadrat are listed, and a score based on the cover of the species in that quadrat is assigned (see Table 2). Cover will be defined as the fraction of the total quadrat area that is obscured by a particular species when viewed from directly above.

From the observations of cover in each quadrat at a site, three statistics will be computed for each species: density, abundance and frequency following the detailed procedures of Fourqurean et al. (2001).

Table 2. Braun-Blanquet abundance scores (S). Each seagrass species will be scored in each quadrat according to this scale (from Fourqurean et al., 2001).

| S | Interpretation |
|-----|--|
| 0 | Species absent from quadrat |
| 0.1 | Species represented by a solitary short shoot, < 5 % cover |
| 0.5 | Species represented by a few (< 5%) short shoots, < 5% cover |
| 1 | Species represented by a many (> 5%) short shoots, < 5% cover |
| 2 | Species represented by many (> 5%) short shoots 5%-25% cover |
| 3 | Species represented by many (> 5) short shoots, 25%-50% cover |
| 4 | Species represented by many (> 5) short shoots, 50%-75% cover |
| 5 | Species represented by many (> 5) short shoots, 75%-100% cover |

Slightly different methods are used to ensure an unbiased placement of sampling quadrats in the three monitoring programs. In this study, we propose to adapt the procedures used in the FKNMS program: 10 quadrats are placed at each site by locating the quadrats at pre-determined random distances along a 50 m transect placed in a N-S direction at each site.

Frequency of Sampling and Replication

We propose to sample permanent sites in December 2004/January 2005 and again in August/September 2005 within the two estuarine systems. Bi-annual sampling will be used (1) to enable accurate calculation of indicator sensitivity on a temporal scale (i.e. what is the minimum indicator response time to changes in water quality?), and (2) to identify the specific seasonal period that various measures are most sensitive indicators of ecosystem health. This information provides the critical criteria needed in the design of an annual monitoring program in which sampling is confined to certain periods and a specific frequency. The data thus provides a sound statistical basis for management decisions regarding the resource.

At the permanent sites we propose to collect replicates (n=3; e.g. three-way field splits) of all samples that require analytical analysis. An exception is grain size analyses that will be performed during the initial and final sampling only. These replicate analyses will provide an ample sample size for statistical analyses over the duration of this study.

Temporal and Geospatial Analysis

Data collected during the field effort will be incorporated into the geospatial database in various GIS layers for assessment and statistical analysis. These data layers will include seagrass distribution and measurements from a variety of indicators. Since indicators are clearly linked to an underlying ecological process, geostatistical analyses can be used effectively to evaluate the power and reliability of a given indicator.

The expected benefits of these activities include:

- Evaluation of the relative importance of various seagrass indicators for a state-wide seagrass monitoring program.
- A demonstration of a seagrass monitoring program in two different Texas estuaries characterized by widespread cover and diversity of seagrasses.

Intensive measurements of selected indicators will occur twice over the duration of this study beginning Winter 2004/2005. Beginning in February 2005, this data will be analyzed and interpolated using Geostatistical Analyst, an Arc/Info 8.1 extension. Four deterministic interpolation techniques are available. In addition, geostatistical techniques, which involve kriging or cokriging methods (for multivariate cases) can be used to create prediction surfaces. Several methods are available in the Geostatistical Analyst extension.

Understanding the combination of spatial and temporal trends in data requires a combination of techniques from geospatial analysis using GIS and time series analysis. Some of the issues are the following:

- A given indicator at a particular location may vary seasonally (e.g. dissolved oxygen) or have a value that shows little seasonal variation (e.g. total suspended solids). Figure 4 illustrates this behavior for dissolved oxygen in Lavaca Bay.
- There may be consistent differences from one year to the next in the level of an indicator. Figure 5 illustrates this behavior for chlorophyll *a* in South Texas estuaries in 1994 and 1996.
- The indicators are linked by physical, chemical and biological relationships.

The techniques we propose to use in this study to clarify these relationships are:

- Geostatistical analysis of point information to generate spatial maps of expected concentrations of variables and their standard error of estimate. The Geostatistical Analyst extension of ArcInfo 8.1 will be used for this purpose. We have successfully employed this technique in a study of the benthic community in the Western Arctic ocean (Jonsdottir, et al., 2000).
- Fourier analysis of indicators showing seasonal variations, as illustrated in Figure 4. This is fairly easy to accomplish using regular regression methods, which can be combined with annual variables to examine combinations of year-to-year trends and seasonal variations within a year.
- Regression or other mathematical or physical models to describe the interrelationships of the indicator variables. This analysis will likely be programmed in Visual Basic to operate on data in ArcInfo 8.1 or a combination of ArcInfo 8.1 and Excel.

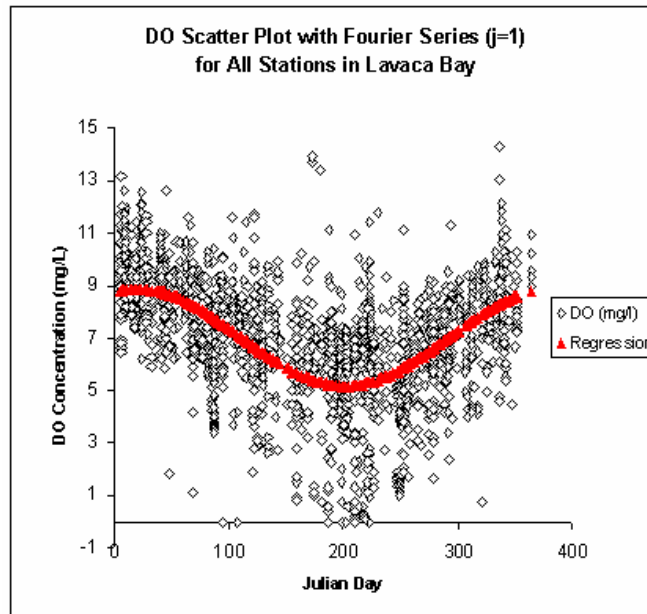


Figure 4. Dissolved oxygen measurements at 13 locations in Lavaca Bay (after Baguley, 2000)

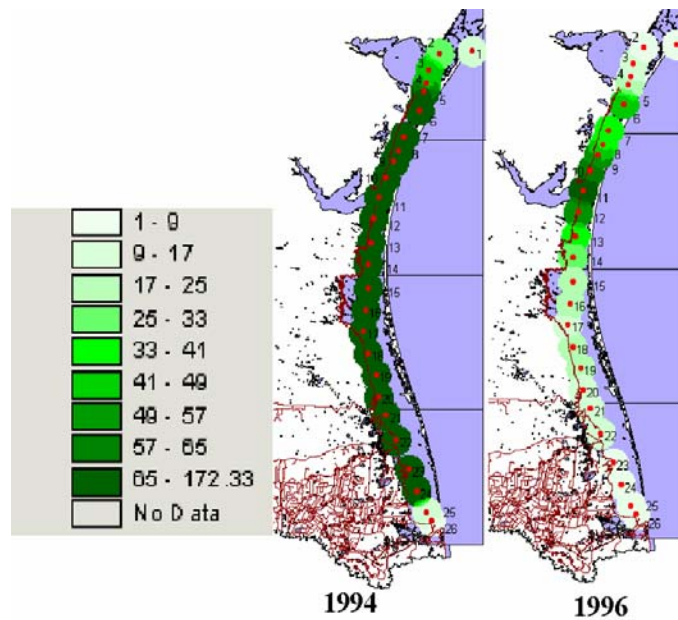


Figure 5. Spatial patterns of chlorophyll *a* concentrations in South Texas estuaries in 1994 and 1996 (after Aumack, 2000).

Morphological and Physiological Indicators of Stress

Neckles (1994) listed several morphological and physiological features as possible response indicators to assess seagrass health. Of these, biomass is most often measured in seagrass monitoring studies (Burd and Dunton, 2000; Dunton, 1990; Dunton, 1994; Kaldy and Dunton, 2000; Lee and Dunton, 1997), although other indicators have shown considerable promise. Indices of below-ground to above-ground biomass ratios, shoot density, and blade width have been documented as measurable early warning indicators of chronic underwater light stress in both *Thalassia testudinum* and *Halodule wrightii* (Dunton, 1994; 1996; Lee and Dunton, 1997, Kaldy and Dunton, 2000). Since these morphometric measurements are relatively easy to make, they are excellent candidates as indicators of plant stress.

Physiological indicators have been historically associated with complex and time intensive measurements that are unrealistic for long term monitoring. However, recent advances in instrumentation have allowed researchers to collect physiological information that has enormous potential for assessing ecosystem health. Elemental composition of plant biomass has been used to assess the nutrient status of a plant's environment. This has been demonstrated for macrophytes as well as phytoplankton (Gerloff and Krombholz, 1966). Fourqurean et al. (1992) provided one of the first examples of the application of C:N:P ratios to assess nutrient status in seagrasses. In Texas, Lee and Dunton (1999) found C:N ratios in *Thalassia testudinum* ranged from about 15 in a nutrient replete system (Corpus Christi Bay) to 24 in the oligotrophic Laguna Madre. In situ fertilization of plants in the Laguna Madre caused C:N ratios to drop from 24 to 16, providing clear evidence that the Laguna plants were N-limited (Lee and Dunton, 1999). Unfortunately, none of the indicators listed above provide an instantaneous assessment of plant health. They can provide information on sub-lethal conditions to which the plants are exposed, which by their nature are integrative over temporal and spatial scales. Coastal managers and research scientists alike have continued to search for innovative methods to efficiently assess anthropogenic impacts on seagrass primary production on much shorter time scales, before degradation of plant tissues becomes non-reversible. Blade growth measurements are difficult to do in the field, are destructive, and require considerable time. On the other hand, physiological measurements have traditionally involved measurements of photosynthesis (i.e. ^{14}C uptake and O_2 evolution) that involve hazardous materials, special incubation chambers, and considerable time.

Recently, pulse amplitude modulated fluorescence (PAM) was developed as an innovative approach to measure photosynthetic performance of both vascular plants and algae. The technique shows enormous promise as an indicator of environmental stress in plants (Schreiber et al., 1994). PAM fluorescence requires little set-up and no sample preparation. The measurement is rapid, non-intrusive, and is free of chamber effects. The fluorescence measurement is an instantaneous reflection of seagrass physiological condition and primary production. The approach is one of the most innovative techniques to emerge for rapid assessment of seagrass health in response to any stressor that would impact photosynthesis. In addition, chlorophyll fluorescence yields can be used to estimate seagrass primary production (Beer and Bjork, 2000). Preliminary data collected on high and low-light exposed *Thalassia testudinum* plants in Texas has demonstrated that PAM fluorescence is a potentially excellent indicator of plant physiological condition (Major and Dunton, 2001).

In situ measurements of chlorophyll fluorescence will be made using a submersible version of the portable Mini-PAM (Pulse-Amplitude-Modulation, Walz, Germany). A custom-built cuvette, positioned parallel to the blade, holds the fiber optic probe at a uniform angle for repeated

measurements. The ratio of variable to maximum fluorescence (F_v/F_m) is a reflection of light stress and is proportional to the maximum photon yield of PS II. High F_v/F_m ratios (0.7-0.8) are exhibited by plants that have optimum photosynthetic potential and pigment concentrations under an ambient light field. Low F_v/F_m ratios (0.2) can be indicative of plants that have sustained some photodamage or partial impairment of photosynthesis. Care will be taken to standardize the measurement of fluorescence, which is inherently complex and poorly understood by many plant biologists. We are well aware of the potential problems with in situ fluorescence measurements; one of us (KHD) has a large project that is focused on the application of PAM chlorophyll fluorescence in the assessing the effects of UV on algal photosynthesis. A field protocol for accurate and consistent measurement chlorophyll fluorescence is currently being developed for seagrasses on a separate project funded by Texas Sea Grant. Procedures will take into account such factors as light acclimation, dark adaptation of leaf tissues, time of day, ambient light at time of measurement, the number of replicates required, etc. The collection of PAM chlorophyll fluorescence will be dependent on the establishment of an accepted and field-tested protocol developed and funded independently of this project.

Data Analysis

Data analysis for this project will be carried out according to standardized procedures established by the EMAP program (<http://www.epa.gov/emap/>). Point data on species density will be used to produce continuous maps of the density of seagrass species, as well as maps of species richness. Krigging algorithms will be used to interpolate between the random point data and spatial analysis programs (eg. SURFER, Golden Software, Golden, CO, USA) will be used to compute areas of seagrass coverage from these interpolated surfaces.

Central to these procedures is the creation of hexagonal sampling units covering the two study sites as discussed in Section 3.2. One component of EMAP is a National Coastal Assessment (<http://www.epa.gov/emap/nca/>), and within this assessment there exist various regional EMAP studies (<http://www.epa.gov/emap/html/>).

Each program has a standardized statistical sampling procedure in which a sampling site is randomly selected spatially within each hexagonal unit (not all hexagonal units may be sampled in a given year of a particular study). Two types of variables are measured: *Condition Indicators* and *Stressor Indicators*. Condition indicators are “characteristics of the environment that provide quantitative estimates of the state of ecological resources that are important to society”, and stressor indicators are “characteristics of the environment that are suspected to elicit a change in the state of ecological resources”. Statistical analyses are then implemented to relate the condition and stressor indicators.

Interpretation of condition and stressor indicator data may require characterization of sites using descriptive criteria for “impaired” sites or separation of sites into various classes of impairment based on quadrisection of indicator values. Least impaired sites would be defined as remotely located from sources of perturbation with little or no observed physical damage.

Transformation of indicator values may be required for site separation into “impairment classes”, because of assumptions of linear response and homoscedasticity (Hair et al. 1995, Zar 1996). The coefficient of variation (CV) will be calculated for response indicator values measured among least impaired sites for the entire study period and within seasons. Indicators with a low CV have the potential to yield a more sensitive multimetric index or multivariate response model than indicators with a high CV. Simple linear correlation (Zar 1996) will be used as a test for redundancy among the

response indicators as a step in identifying those that provide the most unique information at the least cost.

There are several approaches that may be useful for observing the relative strength of the response of each indicator to the combined effects of stressors. The development of response models for individual indicators may prove to be the most informative approach (Leps and Smilauer, 1999), allowing visualization of the response across a gradient of conditions. Stepwise discriminant analysis may be used in conjunction with classification of sites as “impaired” or “least impaired” to identify the indicators that contribute the most weight to a discriminant function (Barbour et al. 1992). Canonical Correlation Analysis may also be used to observe the multivariate relationship and canonical loadings for each indicator in the relationship between all stressor and response indicators (Hair et al. 1995).

It is an implicit assumption in all these analyses that the random samples in adjacent hexagonal boxes are statistically independent, that is, the spatial placement of the boxes relative to one another has no bearing on the measurements made within them. Geostatistical analysis using Geographic Information Systems can be used to test the validity of this assumption by constructing a variogram measuring the range of spatial correlation among the sampled measurements (Isaaks and Srivastava, 1989; Cressie, 1991). Geostatistical analysis is also useful for interpolating gridded maps of estimated values of the measured indicators (and standard errors for those estimates), which may reveal spatial patterns in the data that observation of the point values would not reveal. This is particularly useful when there is a great deal of variability in measurements at any given sample point relative to the variability between sample points.

One goal of this study is to identify an indicator variable or combined set of variables that reliably identifies the health of seagrass beds. One method of doing this is an Index of Biological Integrity (IBI), which is a weighted set of indicator values that provide a score. Gradations of the score from high to low are then used to characterize regions of health of the seagrass population. Such indicators need to be:

- Statistically sound in the sense that they discriminate between sampled conditions in a statistically significant manner.
- Scientifically sound in the sense that the variables included in the IBI are rational.
- Reasonable in the sense that they may be used by resource staff to determine an IBI value following standardized protocol within a reasonable amount of time with limited resources. Statistical summarization techniques are helpful in forming such indices but common sense selection of reasonable variables should always take precedent.

Task 2B. Landscape Monitoring

Our landscape studies will analyze bed morphology patterns, vegetative species composition, and human disturbance features of target seagrass beds in the CBBEP study area using high-resolution aerial color photography. Three priority study sites (i.e. North and South Redfish Bay, East Flats in Corpus Christi Bay) are selected for intensive field sampling since work has been conducted there for previous 3 – 5 years by the PI’s (Fig. 6), and their dynamics are fairly well understood. Interpretation and analysis of the aerial film photography will be aided by simultaneous acquisition of multispectral airborne imagery at discrete wavelengths using digital videography. These areas will be photographed

at 2 altitudes, resulting in 2 camera film scales: 1:9600 and 1:24000. Figure 6 shows the three target sites and relative areas covered (i.e. photo footprints) by the 2 scales of photography.

In general the approach based on 1:24000 photography follows the protocol recommended by Dobson et al. (1995) for the NOAA-CCAP Program and reiterated by the NOAA-Benthic Habitat Mapping Program (NOAA-CSC 2001) for mapping seagrass. However recent work described in the TSGMP (Pulich, Fletcher, and Hardegree, unpubl. 2003) indicates that 1:9600 scale photos, because of their better resolution (< 0.5 m ground feature delineation), produce more accurately identified landscape feature data. In order to develop the landscape health indicators desired from seagrass landscape monitoring, high-resolution remote sensing data is necessary, but the exact film scale needed is still equivocal. By acquiring photography simultaneously for study sites at both scales, we will be able to compare the subsequent classification and interpretation accuracy for landscape indicators derived from each film scale. This will enable a decision on the relative accuracy of 1:24000 vs. 1:9600 photos and the source data needed for future landscape monitoring work.

During intensive field surveys conducted at the time of photo acquisition, measurements will be made of water quality and other environmental indicators, and ground truth data will also be obtained on the plant species at target sites. These field data will be incorporated into a GIS database and form the basis for a spatial analysis model (using ARC-View) to correlate Seagrass plant indicators with landscape indicators.

Four additional sites (1. Adjacent to Packery Channel; 2. Adjacent to the Kennedy Causeway in upper Laguna Madre; 3. In the Shamrock Island area of Mustang Island; and 4. North Harbor Island area) will also be photographed for baseline data acquisition at both the 1:24000 and 1:9600 scale. However, intensive field studies will not be conducted at these sites, rather only minimal groundtruthing.

Acquisition of Remote Sensing Data and Ground-truthing Samples

True color aerial photography (large format 9" x 9" Aerocolor 2445 film) will be flown by a commercial aerial photography contractor at both 1:24000 and 1:9600 photo scales. In addition, sub areas of the three intensive study sites will be flown with airborne multispectral videography by USDA-Weslaco collaborators to acquire narrow band imagery at similar high resolution. One site (i.e. East Flats area) is considered to be a control area (relatively undisturbed seagrass beds), as opposed to other more disturbed seagrass areas (e.g. Redfish Bay or Packery Channel). At the three intensive study sites, we will acquire 1 set of photos during Year 1 for temporal change analysis, an end-of-year series taken in Dec 2004. During Year 2 a photo series would be taken in December 2005 for archival purposes (and additional analyses, time permitting). At the 4-5 other baseline sites, only the Dec. 2004 photography will be acquired. Photomissions will be scheduled during good weather and calm, clear water conditions and low to average tides.

“Real-time” ground-truthing with DGPS (differential GPS) will be critical to identifying and locating features in the photos. Ephemeral features such as drift algae, wrack, or sparse seagrass make it imperative to perform groundtruthing within several days (before or after) procurement of the photos. This requires careful synchronizing in flying the photomission with field survey observations. The 1:9600 photo footprints (i.e. 3 smaller colored areas inside the 1:24000 footprints in Fig. 1) define the areal extent of the landscape target areas to be sampled in the field. Field sampling will be

conducted to collect plant/algae and environmental data from fixed stations where we see potential disturbance or pattern features in the photos.

Softcopy Photogrammetry and Digital Processing

Digital scanning will be used to convert analog photographs (positive film transparencies) to high-resolution electronic images with 1 ft per pixel ground feature resolution. Multispectral imagery will be acquired in digital format using aerial videography and narrow-band filters. Then digital imagery will be processed by standard image processing software (e.g. ERDAS, Image Analyst or IDRISI) to rectify and georegister it. Ground control points for registration of the 1:9600 photographs will be taken from white reflective field targets located by DGPS (at +/- 1m accuracy) and visible in the 1:9600 photographs. The 1:24000 scale photographs will be registered from GPS points taken on visible landmarks or from points off digital orthoquads. A GIS database will be created to store all geoprocessed photoimagery, as well as field monitoring data (plant and other biological data; hydrographic, environmental and water quality data). After QA/QC, these data will be transferred to the State agency custodial database at TPWD.

Field Sampling and Plant Scale Measurements

After the photography has been acquired, a field sampling design for hydrographic, environmental and plant measurements will be developed based on the location of bed features and the coverage required for statistical analysis. This field sampling will be designed, coordinated and performed in collaboration with Dr. Kenneth Dunton at UTMSI.

Image Analysis and GeoStatistical Analysis of Landscape Indicators

Landscape features will be delineated from the scanned photos. Image analysis software (e.g. ERDAS and Image Analysis with ARC-View 8.3) will be used to classify the digital imagery and to identify features (seagrass species, plants vs. bare patches, drift macroalgae, human disturbance activities, shoals and channels, etc.). Classification and identification of spectrally distinct features will be aided by separate spectral band datasets (using color filters at 447-455 nm, 483-492nm, 555 – 565 nm, 625-635 nm) derived from the multispectral videography data. Trend analyses will be performed by comparing differences between classified imagery from two time periods to quantitate change for feature classes or species composition. After conversion of images to an ARC-Info GRID, spatial statistics (i.e. landscape metrics) will be derived for bed feature polygons such as patch size and density, edge/shape ratios, species diversity, etc. A list of metrics and corresponding indicator classes follows:

METRICS FOR MAJOR CLASSES OF SEAGRASS LANDSCAPE INDICATORS

Class 1. Morphology and Patterns of Seagrass Plants

Metrics: Shape, size, density, & edge symmetry of beds/patches;
depth range.

Class 2. Non-seagrass Natural Features

Metrics: Macroalgae, bare patches, reefs, channels, sand bars & shoals.

Class 3. Human Impact Features

Metrics: Prop scars, pipelines, “industrial activities”, dredged channels & spoil.

Class 4. Spatial Distribution of Seagrass Species

Metrics: Changes in species composition.

Class 5. Water Column Constituents

Metrics: Turbidity, chlorophyll, water chemistry.

Based on GPS point sampling data collected by Dr. Dunton’s group, spatial modeling (using kriging to interpolate between data points) will be performed with Geospatial Analysis module of ARC-View 8.x; and the model output layers will be overlaid onto the classified seagrass landscapes. From overlay analysis, statistical relationships will be derived between classified landscape features and disturbance indicators from the GPS field data. By identifying the landscape indicators that correlate with biological processes or vegetative characteristics, we will attempt to develop biotic integrity indices for the seagrass beds. Some of the landscape patterns are expected to correlate with disturbance, fragmentation or expansion (growth) processes in the seagrass bed. Multivariate statistical analyses may also be applied to characterize seagrass target sites.

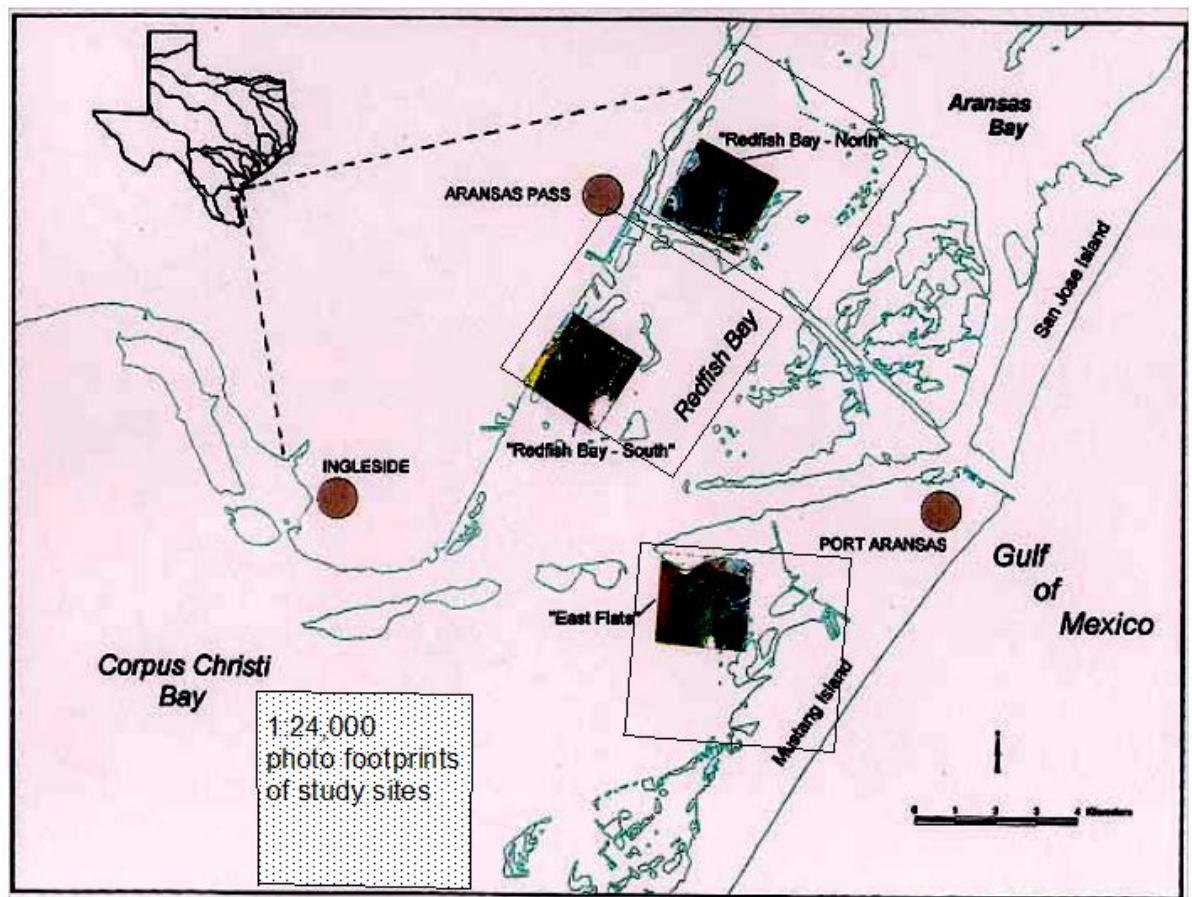


Figure 6. Site map showing three intensive sampling sites for integrated landscape and field seagrass studies in Redfish Bay and East Flats.

ARTICLE V

WORK PRODUCT DELIVERABLES FOR THIS CONTRACT

- (a) Quality Assurance Project Plan (QAPP). The PERFORMING PARTY shall submit a written Quality Assurance Project Plan (referred to as the QAPP) to the CBBEP Authorized Representative. The QAPP must be approved by the CBBEP Authorized Representative prior to the PERFORMING PARTY initiating substantial work related to the major tasks as described in this Scope of Work. Upon approval of the QAPP by the CBBEP, the QAPP shall become part of this contract by reference. The QAPP shall include an introductory overview describing the general approach and specific methods proposed by the contractor and sub-contractors to accomplish work outlined in this Scope of Work. A proposed approach and time line for each task listed in this Scope of Work is to be detailed.
- (b) Quarterly Reports. The PERFORMING PARTY shall submit written quarterly progress reports by the end of each calendar quarter, with the quarters ending on August 15, November 15, February 15, and May 15. Quarterly reports shall detail progress on all major tasks, in chronological order. The Quarterly Reports shall be submitted to the CBBEP Authorized Representative. Instructions for preparing the quarterly report will be provided by the CBBEP Authorized Representative.
- (c) Data. The PERFORMING PARTY shall submit all project data to the CBBEP Authorized Representative in electronic format, in accordance with the CBBEP Data Information Management System (DIMS). PERFORMING PARTY shall also submit the draft report in an electronic format compatible with CBBEP software.
- (d) Draft Report. The PERFORMING PARTY shall submit a written draft report of work completed for review by the CBBEP Authorized Representative. The draft report shall include at a minimum the information requested in this Scope of Work. The draft report shall be submitted as four (4) unbound copies. PERFORMING PARTY shall also submit the draft report in an electronic format compatible with CBBEP software.
- (e) Final Report. The PERFORMING PARTY shall submit a written final report of work completed by no later than the date specified in the Schedule of Deliverables. The final report shall include at a minimum the information requested in this Scope of Work, including revisions requested by the CBBEP Authorized Representative. The final report shall be submitted as four (4) unbound copies. PERFORMING PARTY shall also submit the final report in an electronic format compatible with CBBEP software.

ARTICLE VI

COST SUMMARY AND BUDGET JUSTIFICATION

The estimated project cost is 300 K over a two-year period beginning 1 January 2004 (see attached budget). Salary support is requested for Dunton (1 month total), a part time research associate with expertise in GIS (2 months), a post-doctoral research associate (19 mos), and a Ph.D. level graduate student (24 mos). Travel funds are requested for semi-annual PI meetings and for the PI's (or graduate student) to attend one national or international meeting, conference, or workshop to present results year 2. Travel support for field activities is also requested in both years. We anticipate working 55 days in the field in years 1 and 2 to collect measurements of selected indicators. A very modest sum (total \$3839) is requested for the purchase of material and supplies directly associated with field and laboratory work. This includes filters, flasks, cores, chemicals, batteries, wet suits, booties, masks, snorkels, hip waders, protective weather and safety gear, cell phone charges for a dedicated field phone, charts, herbarium paper, storage bags, maintenance and repair costs for field instruments, etc. A total of \$5500 is budgeted for boat costs and \$5,000 for analytical costs associated with sample analysis (nutrients and chlorophyll). In addition we request a PC computer for data management and GIS model development. The sub-contract to Warren Pulich at Texas State University is listed for a total of \$120,537 for both years.

ARTICLE VII

PERSONNEL AND MANAGEMENT

Dr. Ken Dunton and Warren Pulich will serve as co-principal investigators of this project. Field sampling (including ground truthing) and seagrass/water quality development will be directed and conducted by his staff at test sites. Other contractors on the project will provide geospatial and landscape data analysis support. The active participants will also prepare input to QAPP development where appropriate.

1. Dr. Dunton, a research scientist from the University of Texas-Marine Science Institute at Port Aransas, has extensive knowledge and experience related to the study and monitoring of seagrass productivity and related environmental parameters in estuarine waters.
2. Dr. Warren Pulich, acknowledged seagrass expert in landscape ecology at Texas State University will direct the remote imagery, landscape, and mapping work.

Collaborative Arrangements between the co-PI's

Dr. Warren Pulich at Texas State University (TSU) – San Marcos Inst. for Sustainable Water Resources (IISWR), specializing in seagrass ecology and wetlands remote sensing, will collaborate with Drs. Reginald Fletcher and Jim Everitt at the USDA, Agricultural Research Station, Remote Sensing Unit in Weslaco, TX. The Weslaco group will acquire all multispectral videoimagery and consult on aerial photography analyses. Complete film aerial photography missions will be flown by and purchased from a Texas commercial vendor. Dr. Pam Showalter, a remote sensing specialist in the Geography Dept. at TSU, will provide consultant support on image processing techniques and accessibility to TSU computer facilities. Dr. Kenneth Dunton seagrass biologist and marine scientist at UTMSI, Port Aransas, will direct all field sampling and microscale measurements. His group will collect and analyze plant, sediment and water samples from the field sites. Since UTMSI is located close to the proposed study area, this will also provide essential logistical support for the field research. Mr. Beau Hardegree at US Fish & Wildlife Service, Corpus Christi, will also participate in the fieldwork as part of his coastal program responsibilities. Graduate student thesis projects will be developed around this project, and lead to training in remote sensing, estuarine biology, GIS data analysis, and coastal management.

Collaborative Arrangements with EPA Region 6 and Other Partners

There will be close coordination with the Texas Seagrass Monitoring Work Group (TSWG). The purpose of the TSWG is to help facilitate and develop an effective monitoring plan for seagrass conservation in Texas coastal waters. This group includes representatives from several state agencies including TCEQ, TPWD, CBBEP, TGLO, USF&WS, and USGS-NWRC and EPA Region 6. Active exchange of information, ideas, and resources will facilitate the development of a long-term monitoring program that has widespread support from state agencies and is a model for other related projects. Development of this proposal has already benefited greatly from input from this group. In addition, the PI's have access to a broad range of in-house expertise in statistics.

This project requires the support of EPA-ORD and Region 6 personnel in the development of ecological monitoring tools for risk assessment and probability-based design. Dr. Kevin Summers (EPA, Gulf Ecology Division) is a recognized expert in this area. Communication with Dr. Summers

will be critical to the success and application of our model to other regions and advancing the state of science of monitoring. EPA Region 6 scientists (Charlie Howell, Ken Teague, Philip Crocker) played a critical role in the development of the R-EMAP proposal and are expected to provide input during the design and implementation stages of this project. Input from scientists in local, state, and federal agencies is an important aspect of this project.

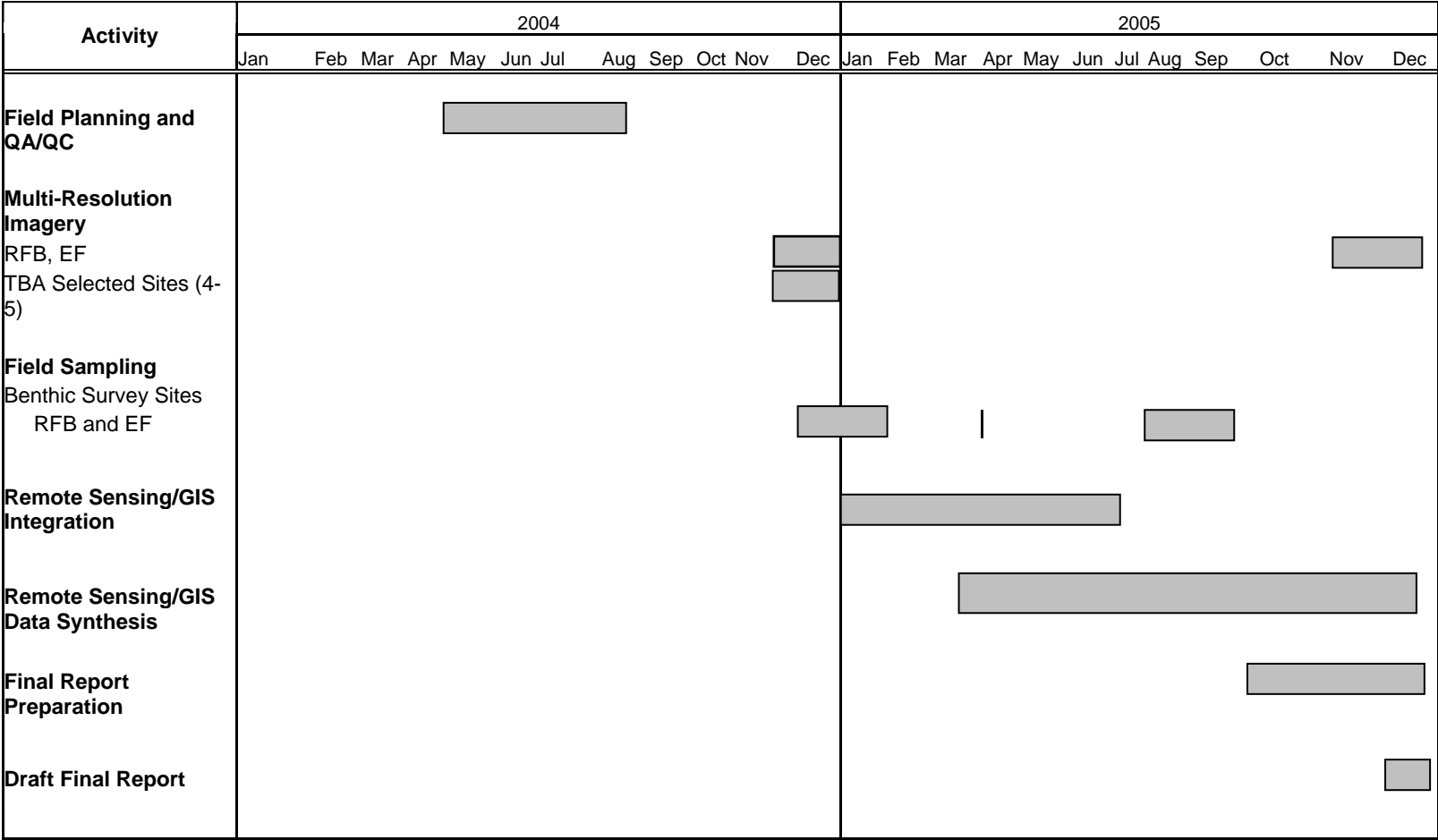


Figure 7. A timeline of activities associated with this project over the two-year period of study.

ARTICLE VIII

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Education

1986-1987, Post-doctoral Research Associate, Stable Isotopic Geochemistry, The University of Texas at Austin

1985, Ph.D., Biological Oceanography, University of Alaska, Fairbanks

1977, M.S., Biology, Western Washington University, Bellingham

1975, B.S., Biology, University of Maine, Orono

Professional History:

2002-present, Professor (Marine Science Institute, University of Texas at Austin)

1996-2002, Associate Professor (MSI, UT-Austin)

1990-1996, Assistant Professor (MSI, UT-Austin)

1987-present, Research Scientist (MSI, UT-Austin)

Six Relevant Publications:

Major, K.M. and K.H. Dunton. 2002. Variations in light-harvesting characteristics of the seagrass, *Thalassia testudinum*: evidence for photoacclimation. *Journal of Experimental Marine Biology and Ecology* 275:1730189.

Dunton, K.H. and S.V. Schonberg. 2001. Assessment of propeller scarring in seagrass beds of the south Texas coast. *Journal of Coastal Research* 37: 100-110.

Lee, K.-S. and K.H. Dunton. 2000. Diurnal changes in pore water sulfide concentrations in the seagrass *Thalassia testudinum* beds: the effects of seagrasses on sulfide dynamics. *Journal of Experimental Marine Biology and Ecology* 255:201-214.

Burd, A.B. and K.H. Dunton. 2000. Field verification of a light-driven model of biomass changes in the seagrass *Halodule wrightii*. *Marine Ecology Progress Series* 209:85-98.

Lee, K.-S. and K.H. Dunton. 1999. Inorganic nitrogen acquisition in the seagrass *Thalassia testudinum*: development of whole-plant nitrogen budget. *Limnology and Oceanography* 44(5): 1204-1215.

Herzka, S.Z. and K.H. Dunton. 1998. Light and carbon balance in the seagrass *Thalassia testudinum*: evaluation of current production models. *Marine Biology* 132:711-721.

Six Other Significant Publications:

Alexander, H.D. and K.H. Dunton. 2002. Freshwater inundation effects on emergent vegetation of a hypersaline salt marsh. *Estuaries* 25(6B): 1426-1435

Dunton, K.H., B. Hardegee, and T.E. Whitedge. 2001. Response of estuarine marsh vegetation to inter-annual variations in precipitation. *Estuaries* 24:851-861.

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Professional Activities:

- Associate Editor (*Journal of Phycology*), appointed January 2002-2005.
- Advisory Board, Nature Conservancy, Coastal Bend Chapter, 2001-2004.
- Co-author, *Seagrass Conservation Plan for Texas*, Texas Parks and Wildlife, 1999
- Steering Committee, NSF Arctic System Science Program/OAII, Western Arctic Shelf-Basin Interactions (SBI) Science Plan, 1994-present
- Associate Editor (*Estuaries*), 1998-2002
- Editorial Board (*Journal of Coastal Research*), 1991-1994, 1997-present
- Review Panelist: NSF/OCE-CAREER, 2003; NOAA-COP, 2003; NOAA-CIFAR (Arctic), 2000; NOAA-NURP (West Coast and Polar), 1998
- Model Evaluation Group, Florida Bay Program Management Committee, 1998
- Review Panel, Corpus Christi Bay National Estuary Program, Technical Advisory Committee, 1994-2000
- Scientific Diving Control Board, NSF (OPP), 1993-present

Awards and Recognition:

- Outreach Award, 2003, College of Natural Sciences, UT-Austin
- Teaching Excellence Award, 1999-2000, College of Natural Sciences, UT-Austin
- Station Leader, Palmer Station, NSF(OPP), Antarctica, 1996
- Jerry McDonald Good Citizen Award, Port Aransas, 1996
- Port Aransas ISD, School Board President, Elected 2002-2004

Activities Related to K-12 Education:

Organized interactive program linking graduate students with K-12 students on climate change in the Arctic; see <<http://www.utmsi.utexas.edu/outreach/k12.htm>>

School Board Trustee, elected May 1999 (term expires May 2002)

**17th Sea Grant Marine Education Symposium (for K-12 teachers and students),
Field/Workshop Organizer (2000)**

Science Shadow Projects, Flour Bluff and Corpus Christi ISD (1993-2001)

Guest Lecturer: Port Aransas ISD, Flour Bluff ISD

Member: Long-Range Facilities and Capital Improvement Committee, Port Aransas ISD (1999-present).

Collaborators:

Leif Anderson((Univ. Goteborg)

Adrian Burd (Texas A&M School)

Lee Cooper (Univ. Tenn)

Glenn Cota (Old Dominion)

David Maidment (Univ. Texas)

Wieslaw Maslowski (Naval Postgraduate School)

Brad Moran (Univ. Rhode Island)

Patrick Neale (Smithsonian)

Mickle Flint (Shirshov Inst. Oceanology, Russia)
Jacqueline Grebmeier (Univ. Tenn)
David Kirchman (Univ. Rhode Island)

John Walsh (Univ. South Florida)
Paul Wassman (Univ. Tromso)
Terry Whitledge (Univ. Alaska)

Graduate and Postdoctoral Advisors:

Donald M. Schell, University of Alaska, Fairbanks
Maurice A. Dube, Western Washington University (deceased)
Patrick L. Parker, The University of Texas at Austin (retired))

Academic Advisement:

14 graduate students (4 Ph.D.; 10 Masters); 4 post-doctoral research associates

Ph.D. Thesis and/or Post-doctoral Advisor:

William Henley (Univ. Oklahoma)
Kelly Major (Univ. Oklahoma)
James Kaldy (EPA-Western Ecology Div.)

Kun-seop Lee (Korea)
Harlan Miller (Univ. Georgia)
David Tomasko (SW Florida Water Mgmt.
District)

RESEARCH INTERESTS

Physiological ecology of coastal marsh plants and seagrasses. (Nutritional and edaphic requirements; Trace metal physiology; Freshwater inflow effects on production).

Restoration ecology of submerged vegetation (Revegetation techniques and dynamics; Restoration responses and natural recovery of impacted wetlands).

Application of remote sensing and GIS techniques to landscape analysis of coastal wetland vegetation/seagrasses
Plant community dynamics and ecological health indicators using remote-sensing analysis.

RELEVANT PUBLICATIONS

Pulich, Jr., W. M. and William A. White. 1991. Decline of submerged vegetation in the Galveston Bay system: Chronology and relationships to physical processes. *Journal of Coastal Research* 7 (4): 1125-1138.

Pulich, Jr., W. and James Hinson. 1992. Methodology for classifying land cover and habitats from Landsat TM imagery of the Texas coastal zone. *In* Proceedings of First Thematic Conference on Remote Sensing for Marine and Coastal Environments, New Orleans, La., 15-17 June 1992, Vol. I, pp. 344-359. Published by ERIM, Ann Arbor, MI.

Pulich, Jr., W. and J. Hinson. 1995. Application of multitemporal thematic mapper data to change detection analysis of Texas coastal land cover. *In* Proceedings of Third Thematic Conference on Remote Sensing for Marine and Coastal Environments, Seattle, WA., 18-20 September 1995, Vol. II, pp.469-482. Published by Environmental Research Institute of Michigan, Ann Arbor, MI.

Pulich, Jr., W. and J. Hinson. 1996. Development of geographic information system data sets on coastal wetlands and land cover. Technical Studies Report No.1. Resource Protection Division, Texas Parks and Wildlife Dept., Austin, TX. 72 p.

Pulich, Jr., W., C. Blair, and W.A. White. 1997. Current status and trends of seagrasses in the Corpus Christi Bay National Estuary Program study area. Publication No. CCBNEP-20. Texas Natural Resource Conservation Commission, Austin, TX. 131 p.

Sheridan, P., G. McMahan, K. Hammerstrom, and W. Pulich, Jr. 1998. Factors affecting restoration of *Halodule wrightii* (shoalgrass) to Galveston Bay, Texas. *Restoration Ecology Journal* 6 (2): 144-158.

Pulich, Jr., W., W.Y. Lee, C.L. Loeffler, P. Eldridge, J. Hinson, M. Minto, and D. German. 1998. *Freshwater Inflow Recommendation for the Guadalupe Estuary of Texas*. Coastal Studies Technical Report No. 98-1. Resource Protection Division, Texas Parks and Wildlife Department, Austin, TX. 55 p. + Appendix.

Pulich, Jr., W. (ed.). 1999. *Seagrass Conservation Plan for Texas*. Resource Protection Division, Texas Parks & Wildlife Dept., Austin, TX. 78 p.

Pulich, Jr., W., J. Tolan, W.Y. Lee, and W. Alvis. (2002). *Freshwater inflow analysis and recommendation for the Nueces Estuary of Texas*. Coastal Studies Technical Report. Texas Parks and Wildlife Department, Resource Protection Division, Austin, TX. 65 p. + Appendix.

Pulich, W., Jr., B. Hardegree, A. Kopecky, S. Schwelling, C.P. Onuf, and K. Dunton. 2003. *Strategic Plan for the Texas Seagrass Monitoring Program*. Resource Protection Division, Texas Parks and

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Appendix B. Example of a UTMSI Field Data Form

UTMSI Field Data Sheet

Date: _____ Transect Meter Mark: _____
 Station ID: _____ Time collected: _____
 GPS Position: _____
 Sample Collector (s): _____
 Monitor (s) Name (s): _____

| STORET | VALUE | PARAMETER |
|--------|-----------------------------|---|
| | | Total Depth (cm) |
| | | Sensor Depth (cm) |
| | | Wind Intensity (mph) |
| 89010 | | Wind Direction 1=N 2=S 3=E 4=W 5=NE 6=SE 7=NW 8=SW |
| | | % Cloud Cover |
| 00010 | | Water Temp (C) |
| 00480 | | Salinity (ppt) |
| 00094 | | Conductivity (mS cm ⁻¹) |
| 00300 | | DO (mg L ⁻¹) |
| | | Light Attenuation (m ⁻¹) |
| | | Surface Irradiance (%) |
| | | Seagrass Percent Cover (%) |
| | | Maximum Depth Limit (m) |
| STORET | SAMPLE COLLECTION CHECKLIST | PARAMETER |
| | | SGS Clay |
| | | SGS Silt |
| | | SGS Sand |
| | | SGS Rubble |
| | | Total Organic Carbon |
| | | Pore Water NH ₄ |
| | | Ammonium (μM) |
| | | Nitrate + Nitrite (μM) |
| | | Phosphate (μM) |
| | | Total Suspended Solids (mg L ⁻¹) |
| | | Seagrass Biomass (g m ⁻²) |
| | | Leaf Area Index (m ² /m ⁻²) |
| | | Blade Width (mm) |
| | | Shoot Density (shoots m ⁻²) |
| | | Chlorophyll Fluorescence |
| | | Species Composition |
| | | C:N:P Blade Ratios |
| | | Epiphytic Algal Biomass (g cm ⁻²) |
| | | Drift Macroalgal Abundance |

Appendix C. An example of a Chain of Custody Form

| UTMSI Chain of Custody | | | | | | |
|-------------------------------|------------------|----------|--------------|------|-------------|-----------------|
| Samples collected by: | | | | | | |
| Station # | Station Location | Position | Date | Time | Sample Type | # of containers |
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| Relinquished by: | | | Received by: | | | |
| date: | | | date: | | | |
| Relinquished by: | | | Received by: | | | |
| date: | | | date: | | | |
| Relinquished by: | | | Received by: | | | |
| date: | | | date: | | | |

Appendix D. UTMSI Data Management Plan

Personnel

The Project Officer is responsible for overseeing the collection of field data and samples as well as laboratory processing of samples. The Project Officer is also responsible for entering all data into database format. The Project Officer will submit the data and Data Review Checklists to the QA Officer, Project Director, and Project Manager for quarterly reviews. After review, the data will be analyzed for writing quarterly and annual reports.

Systems Design

Hardware used to support data processing: PC computers

Software used to support data processing: Windows XP, Microsoft Office 2000, Adobe Acrobat, ArcGIS v. 8.3, SPSS 11.5, Primer 5, Sigmaplot

All data will meet the minimum requirements for submitted information to CBBEP and TCEQ. Data that will be included in the SWQM portion of the TRACS database will be submitted in the proper format as outlined in the SWQM Data Management Reference Guide (TCEQ 2003).

Data Management Plan Implementation

Field data sheets are completed in the field during sample collection. Samples are processed in the lab, and data are recorded on data sheets. All data is proof read and entered into computer spreadsheets. All entered data will be reviewed to identify entry errors or other flaws as described in section D2 of the QAPP. These data management activities occur under the supervision of the Project Officer. Following review of the data by the Project Officer, the data will be verified and validated by the QA Officer, Project Director, and Project Manager before ultimately being distributed to TCEQ and the data users. Chain of Custody forms will be completed with each transfer of data between project personnel.

Quality Assurance/Control

See QAPP.

Migration/Transfer/Conversion

Data will be transferred electronically between project personnel via email. All data will be transferred in the appropriate formats to ensure that the data is imported correctly. Proper formatting and transfer of data will be the responsibility of the Project Officer. The Project Officer will also verify the format for eventual transfer to TCEQ in ASCII pipe delimited text files. The data will be submitted to the Project Director and QA Officer for further review and verification. Following this review, the electronic data file

and a completed hard copy of the Data Review Checklist will be submitted to the Project Manager. The Project Manager reviews the database format and verifies that all data conform to the proper TCEQ TRACS format before submitting both electronic and hard copies to the TCEQ SWQM Team Leader. The SWQM Team Leader then will review and verify the data prior to loading into the TRACS database by the TCEQ Information Resources Division.

Backup/Disaster Recovery

Data will only be recorded in approved software programs, and the Project Officer will make copies of the data on a weekly basis. Back up copies will be stored on a different PC, CD-ROM, or other storage device to ensure that all copies are not stored in the same electronic location.

Archives/Data Retention

The Project Director will retain all original data sheets as well as electronic data files at UTMSI. Electronic files will be stored on CD-ROM or other suitable storage electronic storage device.

Information Dissemination

The data will be made available to the public following completion of the final report.

Appendix E. UTMSI Data Review Checklist

Data Quality Review for Field Data

1. Field Data Sheets completed? _____
2. Appropriate blanks collected or analyzed? _____
3. Chain of Custody record properly filled out? _____
4. Were there any unusual circumstances that may affect data? _____
5. Were there any sample collection problems? _____
6. QC of calibration performed? _____
7. Checks on data reasonableness performed? _____
8. Outliers confirmed and documented? _____

Data Quality Review for Laboratory Analyses

1. Samples stored/preserved properly? _____
2. Samples processed within necessary time limits? _____
3. Calibration of instruments performed? _____
4. Appropriate blanks analyzed? _____
5. Were there any unusual circumstances that may affect analysis? _____
6. Checks on correctness of analysis or data reasonableness performed? _____
7. Outliers confirmed and documented? _____

Data Quality Review of Data Input and Storage

1. Have all field data been entered? _____
2. Have all laboratory analysis data been entered? _____
3. Have all data been checked for entry errors? _____
4. Have backup data files been created? _____

COMMENTS: Explain any answers that may indicate a problem with the data.

Date: _____

Data Reviewed: _____

Project Officer Signature: _____

Date: _____