Restoration Strategies Regional Water Quality Plan
Science Plan for the Everglades Stormwater Treatment Areas

Detailed Study Plans
Fiscal Years 2013–2018

Prepared by the

Applied Sciences Bureau
Water Quality Treatment Technologies Section
South Florida Water Management District

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SECTION 1:
USE OF SOIL AMENDMENTS/MANAGEMENT TO CONTROL P FLUX

1.1 OVERALL STUDY PLAN SUMMARY

The purpose of this study is to investigate whether internal loading of phosphorus (P) in the STAs (i.e., the flux of soluble P from the soil to the overlying water column) can be reduced by application of soil amendments or management techniques and thereby reduce total P (TP) outflow concentrations. This study will be conducted in three phases with a STOP/GO decision made at the conclusion of each of the first two phases. Phase I will involve (1) data mining and synthesis of past District-supported research relevant to this study, (2) continuation of an existing literature review on technologies for controlling soil P flux in wetlands or lakes and (3) to the extent practicable, assessing the feasibility of implementing any of these technologies at full-scale in the STAs. Phase II will involve screening candidate technologies identified in Phase I through small-scale laboratory or field tests to assess their ability to sequester P and, if warranted, select a subset of technologies for further investigation in Phase III. Phase III will involve conducting large-scale field trials using some form of enclosure at the outflow regions in one or more of the existing STAs and/or within the STA-1W expansion area. The experimental approach for the field trials will be to compare the ability of treated enclosures to reduce outflow TP concentrations with the performance of a control (untreated) enclosure. The field trials will be of sufficient size to minimize uncertainty surrounding the scale-up of the technology/technologies in the STAs. The successful outcome of this study will be to identify a technology or set of technologies that when implemented will sufficiently reduce TP concentrations at the STA outflows in order to achieve the permitted TP Water Quality Based Effluent Limits (WQBELs). These technologies may be applied during construction or during operation of an STA to meet the discharge limits.

1.2 BASIS FOR THE PROJECT

Key Science Plan Question Study Addresses

- Key Question 2: How can internal loading of phosphorus to the water column be reduced or controlled, especially in the lower reaches of the treatment trains?

Science Plan Sub-Question Study Addresses

- What are the treatment efficacy, long-term stability, and potential impacts of soil amendments or management?

1.3 BACKGROUND/LITERATURE REVIEW

Biogeochemical cycling of nutrients in wetlands is mediated by a number of factors, one of which is the flux of dissolved nutrients from the soil to the overlying water column. There is a need to evaluate if the flux of dissolved P from the soil in the STAs can be reduced in order to reduce the concentration of TP in water discharged at the outflow of the STAs. Reducing soil P flux during STA startup may shorten the time required for the wetland to achieve its TP startup criterion. The District and other researchers have investigated a number of management approaches to reducing soil P flux, including removing all soil down to the caprock layer, covering the soil with a layer of low-P material (such as limerock), deep tilling the surface soil down into the underlying soil layers and adding soil amendments, either by broadcasting or incorporation.

To date, a number of studies have assessed the efficacy of soil manipulation. The removal of the accrued sediment layer in STA-1W Cell 1B reduced sediment TP concentrations from 1,300 to <400 mg kg^{-1} (SFWMD, 2007). The rate of P release from the inverted soils in the littoral zone of Lake Okeechobee was orders of magnitude lower than P release from undisturbed soils, although there were no significant differences in P release from tilled and scraped (topsoil removed) soils (Water and Soil Solutions, LLC, 2009). Muck removal followed by re-vegetation in the inflow cells of the Orlando Easterly Wetland northern flow-train greatly improved the hydraulic performance and P removal effectiveness of the rejuvenated wetland (Wang et al., 2006). Removing the top 30 cm of sediments in Lake Okeechobee decreased the equilibrium phosphorus concentration (EPC_0) from 0.03 to 0.01 mg L^{-1}, indicating that subsurface sediments had greater affinity for P. Dredging significantly reduced P flux under oxygenated water-column conditions, with P flux in the range of 0.1 to 0.35 mg P m^{-2} d^{-1} (Reddy et al., 2002).

In another recent study, physical removal of the accreted organic soil in combination with alum treatments significantly reduced P flux from a municipal wastewater treatment wetland (Malecki-Brown et al., 2009). Because of the difficulties and costs associated with the removal and disposal of soils from a treatment wetland, it was suggested that alum addition alone may be the most cost-effective and efficient means of sequestering P in aging wetlands experiencing reduced P removal rates, but organic soil removal would be a more permanent solution to reducing P flux (Lindstrom and White, 2011).

A number of studies have been conducted on the effects of soil amendments to reduce P flux. The most common soil amendments are aluminum (Al), iron (Fe), or calcium Ca) salts that bind P and have been effective in reducing water column TP concentrations in several experiments. Three soil amendments [Al-, Fe- and Ca-based products (polyaluminum chloride, ferric chloride and hydrated lime, respectively)] were tested to determine their ability to reduce P flux from a flooded organic soil in a four-month mesocosm study (CH2MHill, 2003). None of the amendments completely controlled P flux, although polyaluminum chloride and ferric chloride were more effective than hydrated lime.

In a field enclosure study involving the application of wastewater treatment residuals consisting of hydrated lime, gypsum and alum and flooded to a depth of 25 cm, alum residuals strongly reduced P flux to the overlying water column (Hoge et al., 2003). P flux reduction in enclosures with lime and gypsum application was much less. Broadcasting calcium silicate ($CaSiO_3$) slag on top of the soil to create a surface barrier reduced the flux of soil P up to 84 percent compared to an un-amended soil control. However, incorporation of the material into the soil was only minimally effective at reducing P release (Chimney et al., 2007).

Additional studies generally confirm the results described above (Reddy et al., 1998; Ann et al., 2000). However, ongoing concerns remain about the quantity of amendments necessary to control soil P flux, the length of time that the amendments will remain effective and potential toxicity associated with different chemicals.

1.4 CONCEPTUAL MODEL

The conceptual model for the major pathways involved in P cycling in the STAs is shown in **Figure 1-1**. This figure highlights the flux of soil P to the overlying water column, the pathway that is the focus of this study.

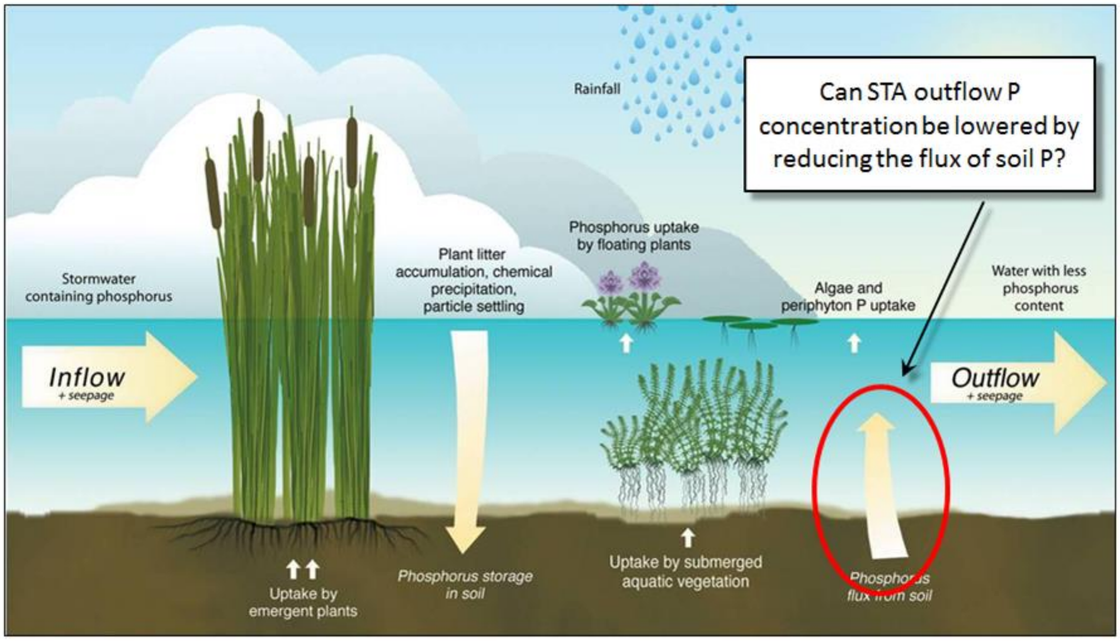

Figure 1-1. A conceptual model showing the major pathways involved in phosphorus (P) cycling in the STAs and the specific pathway that is the focus of this study, i.e., the flux of soluble P from the soil to the overlying water column.

1.5 STUDY PLAN OBJECTIVES

There are two operating hypotheses for this study:

H_0: Reducing the flux of dissolved soil P to the overlying water column in an operating STA will lower the TP concentration in water discharged at the outflow.

H_0: Reducing the flux of dissolved soil P to the overlying water column during startup of a new STA will shorten the time required for the wetland to achieve compliance with the WQBEL.

This study may include a number of separate sub-studies, each of which will focus on a particular aspect of the Key Question and Sub-question. These sub-studies may include, but are not limited to:

- Investigate the use of soil amendments to reduce soil P flux in the existing STAs. The objective would be to reduce the outflow TP concentration.
- Investigate using deep tilling to reduce soil P flux during the start-up of a new STA; the objective would be to reduce the time required for the STA to achieve compliance with the WQBEL.
- Investigate using soil amendments to reduce soil P flux during the start-up of a new STA. The objective would be to reduce the time required for the STA to achieve compliance with the WQBEL.
- Investigate the use of adding a layer of limerock (locally obtained limestone) to cap the soil layer and reduce P flux to the water column in the existing STAs and in the STA-1W expansion area and thereby reduce the outflow TP concentration.

1.6 DETAILED STUDY PLAN AND EXPERIMENTAL DESIGN

1.6.1 Study Plan Description

The purpose of this study is to investigate whether internal loading of P in the STAs can be reduced by application of soil amendments or management techniques and thereby reduce TP concentrations in water discharged from the STA outflows.

This study will be conducted in three phases with a STOP/GO decision for continuing with the study made at the conclusion of each of the first two phases. Phase I will involve (1) data mining and synthesis of past District-supported research relevant to this study, (2) expansion of an existing literature review on technologies for controlling soil P flux in wetlands or lakes and (3) to the extent practicable, assessing the feasibility of implementing any of these technologies at full-scale in the STAs. Phase II will involve screening candidate technologies identified in Phase I through small-scale laboratory or field tests for their ability to sequester P and (potentially) select a subset of technologies for further investigation. Phase III will involve conducting large-scale field trials using some form of enclosure at the outflow regions in one or more of the existing STAs and/or within the STA-1W expansion area. The experimental approach for the field trials will be to compare the ability of treated enclosures to reduce outflow TP concentrations with the performance of a control. The field trials will be of sufficient size to minimize uncertainty surrounding the scale-up of the technology/technologies to the STAs.

1.6.2 Experimental Design and Study Plan Components

This study will be conducted in three phases. **Phase I** will include the following activities:

- Summarize data and findings of past District and DB Environmental, Inc. studies on controlling soil P flux in wetlands.
- Expand the preliminary literature review on technologies for controlling soil P flux in wetlands that was performed during development of the Science Plan. Information gained from the literature review (e.g., technology description, unit costs, treatment efficacy, adverse side effects, etc.) will be compiled into an Excel spreadsheet.
- To the extent practicable, assess the engineering, logistical and economic feasibility of applying any of these technologies at full-scale in the STAs. Where possible, stakeholder and public comments received on this study plan will be addressed during Phase I and will include, but not be limited to, questions concerning the cost of implementation, logistics of applying amendments to the STAs, the long-term treatment efficacy of amendments, potential downstream toxicity and other marsh readiness issues.

The findings from Phase I and any recommendations for continuing the study will be compiled in a summary report. A STOP/GO decision will be made whether to initiate Phase II of the study.

If the Phase I STOP/GO decision is to continue the study, **Phase II** will begin with the preparation of a more detailed research plan. Phase II investigations may use small experimental units, such as soil cores or mesocosms, to characterize the ability of different soil amendments, soil management methods and amendment application methods to reduce the flux of dissolved P from soil to the overlying water column in short-term (days to weeks) trials. Soil used in these trials will be collected from operating STAs or the footprint of the STA-1W expansion area. Findings from Phase II and any recommendations for continuing the study and those technologies to carry forward will be compiled in a summary report. Another STOP/GO decision will be made whether to proceed to Phase III based on the outcome of the Phase II trials.

If the Phase II STOP/GO decision is to continue the study, **Phase III** will begin with the preparation of a more detailed research plan and a request for engineering support to design and construct the field-scale research facilities. Phase III investigations will involve long-term field trials using the most promising soil amendment(s), soil management method(s) or some combination of both identified in Phase II. Field-trials focused on reducing outflow TP concentration in existing STAs will be conducted in large enclosures constructed in the outflow regions of operating STAs, while field trials investigating reducing soil P flux during STA startup will, by necessity, have to be conducted within the footprint of the STA-1W expansion area. The experimental approach will be to compare reduction in TP concentration at the outflow of treated enclosures with the performance of a control enclosure. If feasible, the inflow/outflow water quality data will be supplemented with *in situ* measurements of soil P flux to the overlying water column. All water quality samples collected during this study will be analyzed by the District's Chemistry Laboratory. To the extent practicable, the enclosures will be operated to mimic the hydraulic conditions experienced in the STAs (i.e., water depth, flow regimes and hydraulic retention time). The success of this study will be defined relative to the WQBEL established for the STAs, i.e., can the treated enclosures produce an effluent that meets the TP concentration limits mandated by the permits and Consent Order. The expected duration of the Phase III field trials will be 4-5 years. The findings from Phase III and any recommendation for full-scale implementation of these technologies in the STAs will be compiled in a summary report.

1.6.3 Data Management

All water quality analyses and the data generated from these procedures will be subject to the District Chemistry Laboratory's QA procedures. The water quality data will be stored in the District's DBHYDRO database.

1.6.4 Reporting

Summary reports, as described above, will be prepared at the conclusion of each phase of the study. Letter reports on the progress of the study will be prepared quarterly. An annual summary of study progress will be included in the annual South Florida Environmental Report (SFER). The summary reports for each study phase also will be incorporated into the annual SFER, if possible.

1.6.5 Study Schedule

Phase I: Initiate work – FY2014

Complete work – FY2014

Phase II: Initiate work – FY2014, if needed

Complete work – FY2015

Phase III: Initiate work – FY2015, if needed

Complete work – FY2020

1.7 LITERATURE CITED

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SECTION 2:

DEVELOPMENT OF OPERATIONAL GUIDANCE FOR FEB AND STA REGIONAL OPERATION PLANS

2.1 OVERALL STUDY PLAN SUMMARY

The purpose of this is study is to develop toolsets and methodologies that support the development of operational guidance and regional operation plans for FEBs/STAs that best enable Everglades STAs to consistently achieve the Water Quality Based Effluent Limits (WQBELs) for total phosphorus (TP). This is a three and a half year effort that will include hydrologic, hydraulic and water quality modeling in support of the development of operational guidance for the A1, L-8, and C-139 Annex FEBs, associated STAs and the surrounding regional water management infrastructure. The current proposed study is expected to support a larger series of efforts spread across the construction, implementation and monitoring work associated with the Restoration Strategies program. As project planning and design efforts proceed, information from this study will continually be incorporated into modeling support provided to various restoration strategies project components.

Overall, this study seeks over the next several years to develop a set of decision support tools, including analytical tools and models, that will consolidate known sources of information on WQBEL requirements, regional objectives/constraints, phosphorous dynamics, and vegetation sustainability in an effort to provide more robust and understandable operational guidance. Through extensive information gathering, field testing, and data analysis, a set of tools will be identified that can subsequently be tested and improved during real-time operations of STAs and FEBs. This effort will support many aspects of Restoration Strategies, including design and permitting work, and will ultimately help to maximize the likelihood of achieving WQBEL compliance.

2.2 BASIS FOR THE PROJECT

Key Science Plan Question Study Addresses

- Key Question 1: How can the FEBs be designed and operated to moderate phosphorus concentrations and optimize phosphorus loading rates and hydraulic loading rates entering the STAs, possibly in combination with water treatment technologies, or inflow canal management?
- Key Question 5: What operational or design refinements could be implemented at existing STAs and future features (i.e., STA expansions, FEBs) to improve and sustain STA treatment performance?

Science Plan Sub-Question Study Addresses

- How should storage in the FEBs be managed throughout the year so water can be delivered to the STAs in a manner that allows them to achieve desired low outflow P concentrations?

In addition to the above-listed questions, the following questions also have been raised for consideration in this study:

- How should the individual FEB/STA treatment trains best be operated within the constraints of the larger water management infrastructure to maximize the opportunity to meet WQBELs?

- How should inflows (and associated concentrations) and outflows to and from the STAs be managed to achieve desired low outflow phosphorus concentration?
- How should storage in the FEBs be managed throughout the year so that water can be delivered to the STAs in a manner that allows them to achieve desired low outflow phosphorus concentration?
- How should vegetation management and hydraulic considerations be factored into operational protocols?
- How are non-water quality regional objectives (flood protection, water supply, environmental deliveries, etc.) considered in FEB/STA management protocols?
- To what extent are pulsed versus continuous operations critical to STA phosphorus uptake performance and does this affect the need for remote system controls?
- Which FEB filling and release strategies minimize STA dryout?
- Are there ways through operations to minimize the effects of episodic events or to address STA rehydration strategies following a dryout?

2.3 STUDY PLAN OBJECTIVES

FEBs and STAs can be considered as engineered systems that are can provide quantifiable level of performance in terms of flood control, phosphorus (P) treatment, other regulatory compliances and environmental sustainability. Information gathering, field testing and model hypothesis testing will be performed in order to improve the understanding of FEB/STA system dynamics in terms of response times, attenuation rates, and numerous other relationships between control variables (gate openings) and state variables (water level, TP concentration). Development and implementation of analytic and computer models at varying temporal and spatial scales will help to incorporate improved understanding of the system dynamics into tools that can assist in analyzing infrastructure design and in optimizing operational control algorithms.

The Restoration Strategies program planning relied primarily on the Dynamic Model for Stormwater Treatment Areas (DMSTA) for water quality modeling, predicated on underlying hydrology provided by the South Florida Water Management Model (SFWMM). While this approach was appropriate for and will continue to be used for planning purposes, real time operations support will focus on consolidating multiple sources of information into a broader set of modeling and decision support tools including analytical tools, the iModel (an optimization/ inverse modeling tool) and the Regional Simulation Model (RSM) which can be used to represent both regional (i.e., similar to the SFWMM) and subregional/STA specific hydrology.

This effort will at times support work that is being conducted by others and may also leverage work performed by others into operating protocols and decision support tools that clarify real-time operating protocols by integrating multiple system considerations with the goal of achieving consistent, sustainable low STA outflow TP concentrations that achieve the WQBEL. It is also anticipated that staff across the SFWMD will work with the modeling resources to help develop and review FEB and STA operational guidance. It is anticipated that these protocols will also be revisited over time as FEBs are constructed and operated and additional experience is gained.

2.4 DETAILED STUDY PLAN

2.4.1 Study Plan Description

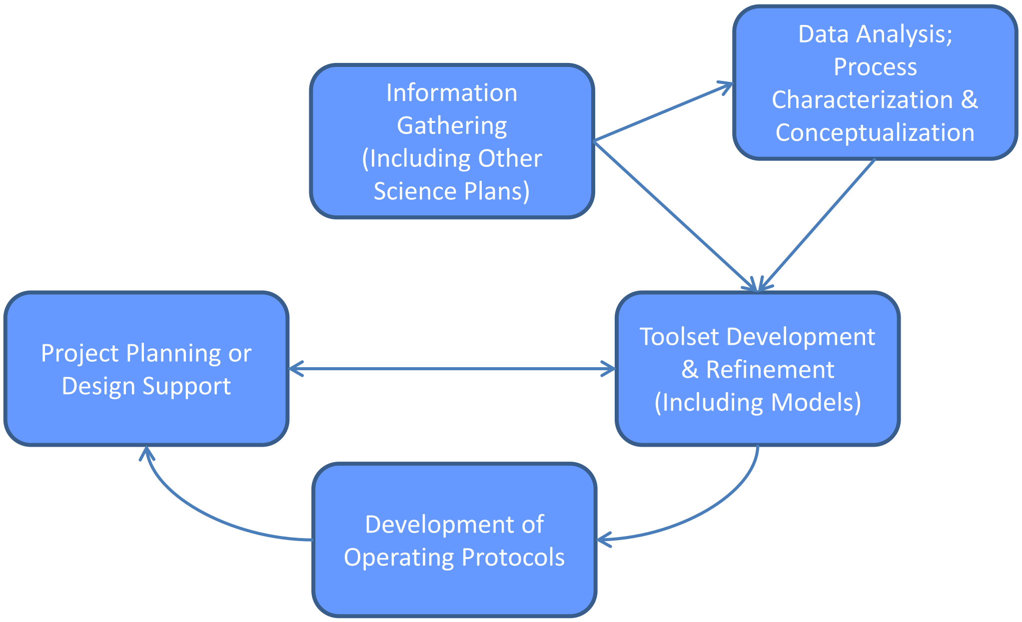
This effort is aimed at integrating multiple considerations of regional hydrology and STA performance dynamics into operational decision support tools. The work includes, but is not limited to, development and application of hydrologic, hydraulic and water quality modeling to test guidance for operation plans.

2.4.2 Study Plan Components

The following work tasks will be implemented throughout the course of the effort toward achieving the project requirements:

- Information Gathering:
    - Field tests: Hydraulics and water quality
    - Incorporation of information from other forums, including science plan projects
- Development of hydraulics, hydrologic and operational model parameters
- Development of local (STAs/FEBs) operating strategies and rules
    - Formulation of overarching operation strategies
    - Local gate opening / pump criteria
    - Application of system optimization tools
- Development of the regional system operating strategies and rules
    - Formulation of overarching operation strategies
    - Regional routing decision trees or algorithms
    - Application of system optimization tools
- Project documentation

A generalized lifecycle interaction is shown in the following figure that encompasses the proposed workflow for the project and organizes the project tasks.



2.4.3 Data Management

The data obtained through the various information gathering efforts will be applied to develop new or improved analytical tools (including models), that seek to represent the complex water quantity and water quality aspects of FEB and STA operation. It is anticipated that the toolset refinement aspect of this project will primarily utilize the RSM due to its relative flexibility in being able to represent a highly varied set of project and operational features, but will also consider improvements to implementations of the SFWMM, DMSTA, and hydraulic/hydrodynamic tool suite (e.g., TUFLOW).

While historical data analysis will be performed and can provide some answers, a series of structured field experiments is proposed to help determine both bulk vegetation resistance and vegetation resistance spatial distributions within STAs/FEBs. The bulk resistance is useful in understanding the overall systems response (e.g., how long would it take to empty or fill an STA/FEB). The spatial and temporal variation of resistance is important during the management of hydraulics within STAs/FEBs, the management of water depth (stage duration) and hydraulic efficiency (short-circuiting). The proposed method of analysis for these field experiments is the use of Solinst portable data loggers to measure the spatial variability of water levels/depths at different locations within the desired system (e.g., Cell 3A in STA-3/4 or Cell 3 in STA-2). Field tests will be carried out by generating small perturbations in hydraulics superimposed on steady or near-steady state conditions. These perturbations are generated by controlling the operations of the facility to honor pre-determined flow patterns capable of producing the desired stage responses. Wave propagation speeds and decay behaviors are monitored within the system for detailed calculations. Changes in water levels recorded by the loggers will be used to calculate wave speed and attenuation within the vegetated area. Data collected will also be used to calculate emptying time of STAs and the shape of the wetting front during filling. These hydraulic phenomena have analytical solutions that relate the hydraulic behavior to the local vegetation resistance parameter. A parallel set of tests is anticipated within the STA-1W test cells. Performing near steady-state analysis within these locations will determine what aspects of bulk vegetation parameters can be inferred from the much smaller test cell locations.

Addition of calcium chloride salt ($CaCl_2$), if allowed, would assist in providing information to determine phosphorous transport mechanics. Existing vegetation (both emergent aquatic vegetation (EAV) and submerged aquatic vegetation (SAV)) may be exposed to several times the background level of conductivity (up to ~ 3000 microsiemens/centimeter (µS/cm) for a period of less than one hour for high and low frequency waves during a test. It is expected that the added salt will become diluted as it is being transported thorough the STA and exists the cell at background concentrations. The spatial extent of the higher salt concentrations during dispersion should also be relatively limited, on the order of tens of meters compared to the wide flow paths of most STAs. It is important to note that since water is constantly flowing during the test that a combination of movement and dilution should prevent vegetation damage.

During execution of the proposed field tests, Remote Phosphorus Analyzers (RPA) will be deployed at inflow and outflow locations. RPAs have been shown to be comparable to other sampling methods and are capable of detecting very low TP concentrations, but provide a much more frequent sampling interval, allowing for the collection of a phosphorus data set to accompany the hydraulic information acquired during the tests. It is anticipated that a more long-term deployment of RPAs at a seasonal scale will also be pursued in order to develop a temporally discrete data set of a representative STA. It is anticipated that this data set will ultimately be utilized to analyze whether operational decision making is sensitive to TP collection frequency.

Outcomes from these tests will be combined with other sources of information and used to identify proposed toolset refinement, thus initiating the project lifecycle. Due to the unpredictable nature of field conditions (availability of water, conditions within proposed STA sites, etc.), the field test efforts will seek to perform tests as logistics allows for a given area of inquiry (e.g., SAV or EAV vegetation).

As tools continue to be refined and are available for project support, a decision support framework will be developed to identify operational guidance for new infrastructure projects and water managers. A key aspect of this step of the work will include the use of inverse modeling and system optimization tools (such as the iModel) to identify mechanisms by which multivariate, competing objectives can be balanced. The decision support system will be developed to consider both short and long-term goals of FEB and STA management and will also attempt to explicitly identify potential risks or benefits of decisions in the context of permit compliance. The decision support system will be designed in a way to facilitate transparent dissemination of information, providing context to users on why and how certain protocols were selected to help facilitate acceptance or feedback.

It is anticipated that implementation of this lifecycle will, at a minimum, result in the following advancements:

1. Improved understanding and formulation of vegetation resistance
2. Improved understanding of flow dynamics (e.g., governing equations of flow)
3. Development of tools for STAs/FEBs structures automation
4. Development of analytical tools for STAs/FEBs operations
5. Improved parameterization and formulation of computer models
6. Development of a decision support framework for operational guidance
7. Improved application of analytical and modeling tools in project planning and design support
8. Development of communication products to explain project outcomes

Summary reports will be prepared at the conclusion of each phase of the study. Letter reports on the progress of the study will be prepared quarterly. An annual summary of study progress will be included in the annual South Florida Environmental Report (SFER). The summary reports for each study phase also will be incorporated into the annual SFER, if possible.

2.4.4 Reporting

Multiple avenues will be utilized to report on project progress, to identify potentially beneficial cross-over collaborations with other SFWMD projects and to continually refine and assess achievement of project requirements:

- Project progress will be reported at the regularly scheduled Restoration Strategies Update biweekly meeting and project tasking and resource decisions will be vetted though the Work Intake System, which is used to manage and prioritize limited modeling resources in support of agency initiatives. Technical findings will be presented to the Technical Solutions Forum for review.
- Project status and progress will be reported via Study Plan Progress Reports and at Restoration Strategies Science Plan Core Team meetings at least quarterly, and technical findings will be presented for review and feedback as products are developed.
- Project status and progress will be communicated at Restoration Strategies Technical Representatives meetings and Long-Term Plan Quarterly Communications meetings, when practical and when requested.

- Project status and progress will be summarized in the annual South Florida Environmental Report (SFER).
- Another forum for inter-SFWMD coordination of this effort will be the Restoration Strategies Modeling Coordination Meetings which include representatives from the Everglades Policy and Coordination, Engineering and Operations groups at the SFWMD.

2.4.5 Activities and Milestones

This project will span a three and a half year timeframe comprised of an initial startup period of information gathering beginning in March 2013 and then a continuous, repeating process of testing, tool development and project planning/design support through FY2016. Within the information gathering phase, each field testing cycle is expected to be on the order of three months from initial study design through final execution. These tests will be followed by a multiple-month data analysis and algorithm development period.

The project will begin with an initial period of information gathering followed by an initiation of the project lifecycle identified in the previous section. Once the lifecycle is initiated, quarterly work tasking will be performed under the existing Work Intake System (WIS) procedures. A primary initial objective of the information gathering step is to further understanding of how vegetation roughness changes within the STAs/FEBs (as a function of water depth, vegetation density, vegetation type, extent of sediment accumulation and other factors) and how the flow conditions change under various extreme operational conditions (i.e., flood control versus phosphorus treatment or removal).

Key highlights of the DSP tasks and work breakdown structure for this study are as follows:

<u>WBS Task 1: Information Gathering</u>

1) Review of literature / project data
    a) Review available literature on STA operations management – the science plan team has compiled a list of prior studies
    b) Bring project team up to speed on Restoration Strategies project features and objectives
2) Historical Data Analysis
    a) Collect, review and analyze historical flow data related to the STAs for applicability to current Restoration Strategies objectives.
    b) Collect, review and analyze historical water quality data related to the STAs for applicability to current Restoration Strategies objectives.
3) Meetings with SFWMD Subject Matter Experts (SME)
    a) Seek out SFWMD project managers, modelers, water managers, permit compliance staff, etc. with experience in operational decision support and meet to gather perspective
        i) Kissimmee Basin Modeling and Operations Study (KBMOS)
        ii) Big Cypress real-time Mike modeling
        iii) Lake Okeechobee operations/objectives
        iv) Others
    b) Summarize findings in a brief memorandum.

WBS Task 2: Field Test Execution

1) Site Identification
    a) Identify STA sites that would be suitable and representative for investigations to determine vegetation resistance (e.g., SAV, EAV, open water)
    b) Given regional and STA conditions, propose field tests as conditions warrant for a given area of inquiry
2) Design and carry out wave experiments
    a) Develop a brief summary report of experiment design
    b) Determine equipment, field work, and control room and pump station crews support needed for experiment and summarize expected test outcomes
    c) Carry out field experiments
3) STA-1W Test Cells
    a) Improve inflow / outflow works for test cells to allow for a broader range of hydraulic analysis through use of temporary pumps and/or custom weir boxes.
    b) Perform near steady state experimentation in test cells to determine what aspects of bulk vegetation parameters can be inferred from the much smaller test cell locations

WBS Task 3: Identification of Potential Model Refinements

1) Field Test Data Analysis
    a) Identifying and quantifying vegetation and topography characteristics (including extent of sediment accumulation) that will be used to understand the STA hydraulics by analyzing and classifying the data
    b) Develop a brief summary report of data collected, methods used for data analysis, results, and its implication to FEB/STA operation protocols and consolidate data into a database for use by other efforts
    c) Develop analytical formulas relating the results of multiple field tests to response times, residence times, peak inflow and outflow rates, hydraulic efficiency and treatment efficiency
2) Identify Modeling Tools
    a) Review existing project production models, the Water Quality Modeling Strategy and current modeling contract reviews to identify candidate models for tool enhancement
    b) Meet with project team leads to agree on tool refinement strategy
    c) Initiate a stand-alone RSM application to test model parameterization

WBS Task 4: Development of Operating Protocols

1) Data Mining
    a) Identify and compile hydrologic data including imposed boundary flows, rainfall, potential evapotranspiration (PET), and all other data time series that could be useful for this project.
    b) Identify Hydrologic Data characteristics and identify data interrelationships

   c) Develop Rainfall Runoff model and any other data characterization for each flow path
   d) Preliminary attempt to model routing using the available preprocessed data given preliminary understanding of flow routing
2) Efficient modeling of the flows paths; make the best use of the available tools, data, and rules to develop a flow routing utility for each flow path covering the regional routing from the LOK to Everglades
   a) Develop expertise in the flow path features such as flow structures, routing mechanics and conditions
   b) Maximize the size and features of the basic building blocks without compromising the flexibility needed for optimizing decisions. For example, both FEB and STAs shall be external sources/sinks to allow for operational decisions into and out of such areas by the optimization model
   c) Comparative analysis and technical investigation to select the most appropriate representation of the system dynamics. This includes the review of the current RSM basin, DMSTA Regional Design Tool (DRDT), formulas developed in the Data mining task
   d) Develop the selected utility and have the ability to identify needed changes to allow for flexibility when needed
3) Intra-flow routing modeling
   a) Compile all intra flow routing conditions and requirements across the three flow paths subareas
   b) Classify the intra flow features as targets, constraints and or standalone representative model
4) STA and FEB Modeling:
   a) Search and identify the best modeling options and the level of details for the hydrologic and phosphorous processes within STAs and FEBs
   b) Examine the use of one or more of (1) real-time data, (2) well-established empirical equations, (3) physically based models and (4) Artificial Neural Network based emulators
5) WQBEL and P variability
   a) Work closely with the P data analysis team to understand the availability and inherent variability of the P data
   b) Develop understanding of the perspective of P data statistics selection in connection with the rules and policies
   c) Attempt to improve understanding of phosphorus variability and explore ways of linking it to rainfall and seasonality (compare to the current monthly averages)
   d) Provide improvement to the estimation of inflow phosphorus concentration and assess the applicability and possibility of estimation in the light of the above three items
6) Implementation of the Restoration Strategy iModel (iModel RS) to provide a tool for optimization and operational hedging

   Use the above mentioned deliverables to implement the iModel RS including hydrologic model emulators, constraints and targets (see Ali, 2009 and Ali, 2013 iModel theoretical concept presentation). Specific details for this application will be developed as work progresses.

2.4.6 Study Schedule

• Task 1: Information gathering	Initiate FY2013	Complete FY2014
• Task 2: Field test execution	Initiate FY2013	Complete FY2016
• Task 3: Identify potential model refinements	Initiate FY2014	Complete FY2016
• Task 4: Development of operating protocols	Initiate FY2014	Complete FY2016

SECTION 3:
EVALUATE P SOURCES, FORMS, FLUX AND TRANSFORMATION PROCESSES IN THE STAs

3.1 OVERALL STUDY PLAN SUMMARY

The purpose of this study is to enhance the understanding of mechanisms and factors that affect phosphorus (P) treatment performance of the Everglades STAs, particularly those that are key drivers to performance at the lower reaches of the treatment flow-ways. Understanding the mechanisms and factors that affect the P treatment performance of STAs at low total phosphorus (TP) concentrations should provide information to develop strategies that will ultimately improve capabilities of the STAs to achieve permit compliance with Water Quality Based Effluent Limits (WQBELs). In order to understand the mechanisms and factors that affect the P treatment performance of STAs at low TP concentrations, it is important to evaluate P sources, forms, flux, and transformation processes along the STA flow-ways. The study will involve data mining, literature review, development of multi-tiered conceptual models, field surveys and studies, and more focused controlled studies. It is expected that this study plan will be reviewed and enhanced as new information is derived via the data mining, literature review, conceptual model enhancement, and initial surveys.

3.2 BASIS FOR THE PROJECT

Key Science Plan Question Study Addresses

- Key Question 2: How can internal loading of phosphorus to the water column be reduced or controlled, especially in the lower reaches of the treatment trains?
- Key Question 4: How can the biogeochemical or physical mechanisms be managed to further reduce soluble reactive, particulate, and dissolved organic phosphorus concentrations at the outflow of the STAs?

Science Plan Sub-Question Study Addresses

- What are the sources (internal/external, plants, microbial, wildlife), forms, and transformation mechanisms controlling the residual P pools within the different STAs and are they comparable to those observed in the natural system?
- What are the key physical-chemical factors influencing P cycling at very low concentrations?
- Are there things that can be done in the STAs to enhance settling, filtering, and treatment of DOP and PP in the water column?

3.3 BACKGROUND

The Everglades STAs (STA-1E, STA-1W, STA-2, STA-3/4, and STA-5/6) were built south of Lake Okeechobee to reduce TP concentrations in surface water runoff prior to discharge into the Everglades Protection Area (**Figure 3-1**). Currently, the STAs provide approximately 23,000 hectares (ha) of treatment area. From 1994–2012, the STAs treated over 13 million ac-ft of runoff water and retained over 1,700 metric tons (mt) of TP load, with an average removal of 74 percent of total inflow load. The overall annual flow-weighted mean (FWM) TP concentration attained at the outflow is 37 micrograms per liter [µg/L, or parts per billion (ppb)], with some flow-ways achieving less than 20 ppb annually or during parts of the water year. While the removal efficiency and resulting concentrations observed at the outflow demonstrate successful treatment, the new stringent effluent limits for the STAs require for further actions to continue to improve STA performance. This includes researching the mechanisms and drivers that influence P concentrations observed at the outflow.

Figure 3-1. Location of the Everglades Stormwater Treatment Areas (STAs) 1 East (1E), 1 West (1W), 2, 3/4, and 5/6 in relation to the Everglades Protection Area, the dominant STA vegetation communities [emergent vegetation (EAV) or submerged aquatic vegetation (SAV)].

3.3.1 Overview of Phosphorus Removal Process

Phosphorus reduction in wetlands occurs via many biogeochemical pathways, including adsorption, desorption, chemical precipitation reactions, uptake by biota, as well as sediment accretion (Kadlec, 2008; Reddy et al., 2005; Dunne and Reddy, 2005). Phosphorus sorption and desorption are two of the most important processes regulating the retention and release at the soil-water interface. Phosphorus co-precipitation with metallic ions such as calcium (Ca), magnesium (Mg), iron (Fe), and aluminum (Al) is also a major P uptake mechanism in many wetlands. In calcareous systems, such as the Everglades STAs, P can also sorb onto Ca-rich substrate. Because the STAs are natural systems, flora and fauna are vital components of the P cycling in these systems. The influence of vegetation and microbial P uptake and release, resulting in formation and accretion of new peat and soil, have been intensively studied for many natural and constructed wetlands (Kadlec, 2006; Richardson and Marshall, 1986; Richardson et al., 1997).

Phosphorus retention mechanisms in wetlands are regulated by many factors, e.g., pH, redox potential, concentration of metallic ions such as Ca, Mg, Fe and Al, soil characteristics, and vegetation condition (Reddy et al., 1999; Richardson and Qian, 1999). In the STAs and similar wetlands, previous analyses have concluded that hydraulic and P loading, as well as inflow concentrations are important drivers to the P uptake performance (Kadlec and Wallace, 2009; Ivanoff et al., 2013; Chen et al., 2014).

Based on an analysis of long-term internal P profile in STA-2 Cell 3, a predominantly SAV cell, Juston and DeBusk (2011) found that the lowest achievable water column P concentration was 16 ppb. Juston and DeBusk (2011) concluded that the P reduction trend was insensitive to P loading of less than 1.7 $g/m^2/yr$. Chen et al. (2014) also reported that at phosphorus loading rates PLRs) of less than 1.4 $g\ P\ m^{-2}\ yr^{-1}$, the relationship between PLR and outflow TP concentration becomes insignificant, suggesting that the outflow TP concentrations at low P environment are being influenced by other factors and processes that occur within interior of the flow-ways, e.g., internal flux and detrital P release.

3.3.2 Phosphorus Species in the STAs

Inflow water contains both organic and inorganic forms of P and can be in soluble and particulate forms. The particulate and dissolved organic forms have been further categorized into labile and refractory pools. Soluble reactive P (SRP), also known as dissolved inorganic P (DIP), is considered readily available for microbial and plant uptake while organic and particulate forms must undergo transformations before they can be bioavailable (Reddy et al., 1999). Particulate P (PP) includes phosphate sorbed onto suspended clay particles, and suspended crystalline and amorphous precipitates of $PO4^{3-}$ with Ca, Mg, Al, and Fe (Reddy and DeLaune, 2008).

Organic P refers to any P that is associated with organic compounds, including nucleic acids, phosphoamides, phosphoproteins, sugar phosphates, inositol phosphates, phospholipids, phosphonates, organophosphate pesticides, humic-associated P, and organic condensed P (Baldwin, 2013; Turner, 2005). In aquatic systems, organic P may be present in dissolved form (DOP) or particulate form (POP) and can be transformed in surface waters and flooded soils by physiochemical (e.g., adsorption onto CaCO3, precipitation as metal-humic complexes, or UV degradation) and biological (e.g., enzymatic hydrolysis processes) (Wetzel, 1999). Because DOP is conveniently calculated as the difference between TDP and SRP, it is more common to see values reported for DOP and not for POP. Organic P forms found in detrital material in STA-1W include sugar phosphate, glycerophosphate, polynucleotides, and phospholipids (Pant and Reddy, 2001). In soils, organic P forms include phospholipids, nucleic acids, inositol phosphates (phytin), glucose-6-phosphates, glycerophosphate, phosphoproteins, and polymeric organic P of high-molecular weight compounds (Steward and Tiessen, 1987).

While the terminologies SRP, DOP, POP, PIP, and PP (surface water and porewater), and labile versus recalcitrant P fractions (soils) have been widely used in characterizing the species of P in the STAs, these are operationally defined based on sample preparation and analytical procedure. Because the chemicals used in extraction and analytical procedure can actually interfere with the speciation through hydrolysis of organic material, there has been no accurate assessment of the actual distribution of DOP or POP forms in the STAs. Other experts have also pointed out the limitations of the common P speciation methodologies (Baldwin, 2013; Denison et al., 1998; McKelvie, 2005). Despite the known limitations, these methods of P speciation in water and soil remain popular due to the convenience and ease of procedures, and in providing coarse estimations of the different P pools.

Similarly, P species in soils are categorized according to their reaction with extractant salt, weak acid, or weak alkali solution. Depending on the extraction procedure, P species in the STAs have been categorized into microbial, labile, moderately labile, and resistant P pools (Ivanoff et al., 1998). Previous soil P fractionations on STA soils indicate that inorganic P pool comprise the bulk of stored P in SAV-dominated areas while organic P comprise the bulk of soil TP in EAV areas. A recent study on limited samples from STA-1W and STA-2 found that reactive P comprise up to 70 percent of soil TP while 30-40 percent is in the highly refractory pool (Bhomia et al., 2012). Similar to the limitations of wet chemistry methods in accurately quantifying the different P pools, the analysis of the extracted materials can also result in breakdown of DOP and PP and may potentially bias the results. An assessment of the P pools in the soil by extraction method, complemented by a more accurate speciation according to molecular structure, could provide an indication of the short-term and long-term stability of the P currently stored in the STAs and what will be accreted in the future.

The likely sources of P detected at STA outflows are (1) residual P from inflow that has not settled (PP) or has not been converted via biogeochemical transformations (PP and DOP), and excess SRP that was not consumed by microorganisms, periphyton, or SAV or has not reacted with cations such as Ca; (2) the release of P from organic matter decomposition and upward flux within the STA (SRP, DOP, PP); and (3) P associated with suspended particulates including suspended solids, plankton, bacteria, and detritus (Kadlec and Wallace, 2009). To date, only minimal attention has been focused on phosphorus speciation (Kadlec and Wallace, 2009). The composition and origin of the remaining P observed at the outflow of STAs, and the actual composition of PP (e.g., biomass, necromass, and inorganic matter), DOP (e.g., monoesters, diesters, or more refractory forms such as phytic acid), and more complex molecules in STA outflow water are unknown. Understanding the speciation of P is a precursor in knowing the sources and fate of the different P species. This information will be useful in enhancing our understanding of the P reduction performance along the treatment flow-way and in developing strategies for further lowering outflow TP concentrations.

3.3.3 Phosphorus Transformations and Flux

Phosphorus species and characteristics change in natural systems as a result of various biotic and abiotic processes. A schematic of the general P cycle in wetlands is presented in **Figure 3-2**, and a more detailed conceptual model describing the key processes involved in these transformations is presented in **Appendix A**. Transformations that occur within the water column, the soil-water interface, or within the soil column can affect the P concentrations measured at the STA outflow locations. Phosphorus is transported with flows in dissolved and particulate forms. A bulk of the inorganic P in the water column is rapidly absorbed by bacteria, periphyton, and floating aquatic vegetation, while a fraction of the inorganic P may also adsorb on particles containing metallic ions such as calcium (Ca), magnesium (Mg), iron (Fe), and aluminum (Al) or co-precipitate with Ca or Mg. The relative importance of each ion that can control these sorption or co-precipitation reactions depends on other factors such as pH, alkalinity, and redox potential.

Soluble reactive phosphorus (SRP) in the water column or porewater can be removed efficiently by plant uptake, while stored phosphorus in plants can be released back into the water column in the forms of SRP, DOP, and PP (Reddy et al., 1999; Corstanje et al., 2006; Mitsch and Gosselink, 2007). Rapid but short-term retention, i.e., assimilation into vegetation and periphyton and incorporation into detrital tissue, should be distinguished from long-term nutrient burial, i.e., soil assimilation and accretion of organic and mineral matter (Reddy and DeLaune, 2008). Although initial short-term nutrient retention can be high, these storage compartments are often quickly saturated, especially in systems that have not previously received elevated nutrient loads (Johnston, 1993).

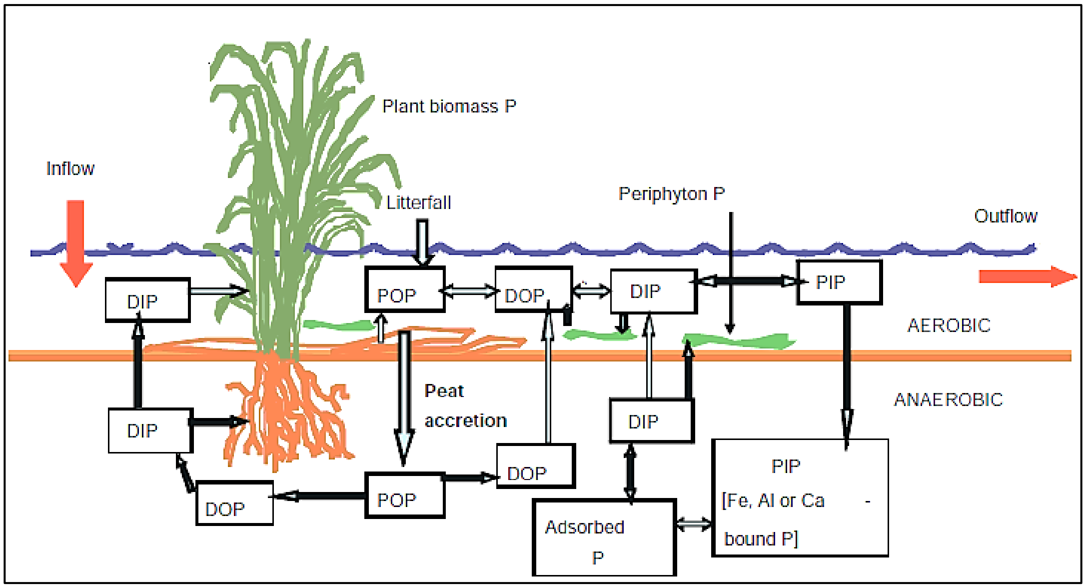


Figure 3-2. Schematic of the general phosphorus cycle in wetland systems (Source: University of Florida Wetland Biogeochemistry Laboratory).

Whether driven by changes in water column properties due to SAV productivity, or direct chemical interactions, precipitation of P with metal cations such as Fe, Al, Ca, or Mg, results in formation of amorphous or poorly crystalline precipitate solids (Dunne and Reddy, 2005; Reddy and DeLaune, 2008). These reactions typically occur at high concentrations of either phosphate or the cations (Rhue and Harris, 1999). In the STAs, Ca supply is abundant, coming from both the inflow water and the underlying substrate (Ivanoff et al., 2012). In the natural Everglades, the rate of Ca accumulation explained 97 percent of the P accumulation rate along the nutrient gradient in WCA2A (Reddy et al., 1993). Gu et al. (2005) and Ivanoff et al. (2012) closely examined the influence of Ca concentration on poor performance of STA-5. They determined that despite Ca levels approximately 10 mg L^{-1} lower than the other STAs, the Ca saturation index indicates a sufficient supply of Ca for Ca-P co-precipitation to occur. Subsequent dissolution and solubilization can also occur when the concentration of any of the reactants fall below the solubility product of that compound, resulting in P release (Dunne and Reddy, 2005). Similarly, Diaz et al. (1994) demonstrated that the ratio of Ca:Mg can influence the solubility of Ca phosphates.

Retained P within the STA cells is distributed among the accreted floc, soil, microbial, and vegetation biomass. The floc and surface soil in the STAs generally have higher P concentration than the deeper soil layer due to enrichment from recent deposition, biomass decomposition, and translocation of P from subsurface soil layer through vegetation uptake. Long-term retention is dependent upon the stability of the organic matter formed (Reddy and DeLaune, 2008, Hamdan et al., 2012), the sustainability of vegetation, and the formation of amorphous Ca-P precipitates. Transfer of P from or to the soil/sediment occurs primarily through deposition or resuspension of particulate matter (Reddy et al., 1999) that can return stored nutrients to the water column and reduce wetland treatment efficiency (Chimney and Pietro, 2006). Reddy et al. (1999) indicate that above-ground plant parts return leached P to the water after death and decomposition and deposit refractory residuals on the soil/sediment surface. However, dead roots decompose underground, therefore adding refractory compounds to subsurface soils and leachates to the pore water root system. Soils in many STA cells have high organic matter content and, therefore, the characteristics and factors regulating the decomposition of organic matter in organic soils determine the long-term storage of organic P pool. Also, organic matter oxidation under anaerobic condition is generally slower than under oxic condition and, consequently, release of P via decomposition is slower (Reddy et al., 2005). However, during periods of drawdown, organic matter decomposition is accelerated, resulting in rapid mineralization of P-containing compounds and subsequent P release into the water column upon re-flooding.

As a result of the gradient between the water column and P-enriched floc and surface soil layers, P flux occurs within the STA cells. This process is influenced by various chemical, physical, and biological processes including diffusion and advection via wind or flow, bioturbation, vegetation-mediated flux, redox conditions, diagenetic processes at surface of sediments, and abiotic and biotic processes in the water column (Reddy and DeLaune, 2008). Higher EPC_o values and greater amount of labile and potentially mineralizable and porewater pools of P in P-enriched areas than in unenriched areas indicate a greater potential for P flux in enriched soils ((McCormick et al., 2002; Reddy and DeLaune, 2008; Reddy et al., 2011; Wright et al., 2009). If surface water has a higher concentration of P than the concentration of the existing amount of mobile P in the soils and sediments then there is still capacity for further P accumulation; however if surface water concentrations are lower than this threshold concentration, P may be released from the sediments**.** In STA-2 cell 1, which achieves the lowest outflow TP concentration among full-scale STA flow-ways, Dierberg et al. (2012) reported SRP flux rates of 0.05 to 0.07 mg m^{-2} d^{-1} at the inflow and negligible flux at the outflow. Using in situ enclosures (Community Watershed Fund and DB Environmental Inc., 2009) measured average flux rates during 2005-2008 were 2.08, 2.98, 3.28 and 2.78 mg P m^{-2} d^{-1}, at enriched sites in WCA2A. Fluxes as high as 6.5 and 10 mg m^{-2} d^{-1} were recorded at highly enriched sites using cores and in situ benthic chambers, respectively (Fisher and Reddy, 2001). Reddy et al. (1999) indicate that soluble P may be entrained (carrying material in a current) by leaching of dissolved organic and inorganic P from detrital macrophytes and periphyton or release from surface sediments. Where flowing water has low soluble P concentration rapid replacement of the water column creates a large gradient and increases diffusive flux of P from sediments.

Another P transformation mechanism in wetlands is plant translocation from the deeper soil layer to the soil surface. This process is termed P mining. The overall effect is the removal of phosphorus from underground storages, and re-deposition of phosphorus on the top layer of sediments. This upward transfer is counteracted by at least two very important processes: the transpiration of phosphorus flux, and translocation of phosphorus from senescing leaves to the rhizomes (Kadlec and Wallace, 2009).

Phosphorus release from historically P loaded soil/sediment to overlying water is typically a result of diffusion-controlled process due to a P concentration gradient between underlying soil and overlying water (Dunne et al., 2010). However, P can also flux from soils to overlying water via advective processes. Advective P release can be caused by bioturbation, the feeding activities of invertebrates in underlying soil and it is the bulk movement of soil porewater to the overlying water column (VanRees et al., 1996). The dynamics of P exchange at the sediment-water interface is regulated by pH of the water column, oxygen status, sorption/precipitation reactions, plant uptake and the physicochemical properties of the underlying sediment (Reddy et al., 1995).

Since previous analyses have indicated that factors other than P loading and inflow concentration may be controlling P concentrations in the lower reaches of the treatment train, investigating P transformation mechanisms in these areas and the factors controlling those mechanisms can provide useful information when selecting and developing management strategies for these flow-ways. In order to understand and manage the long-term effectiveness of the STAs in removing P there is a need to determine the dynamics of P exchange (flux) between the soil and water column, including the P sorption and desorption properties of the floc and the underlying soil layers, and the various biotic and abiotic factors that influence P removal and storage in the STAs.

3.3.4 Microbial Pathways

The life cycle of aquatic microorganisms is short so turnover of essential nutrients such as phosphorus is rapid, particularly under the nutrient limiting conditions observed in the Everglades (Noe et al., 2003). Microbial communities decompose organic material and remobilize nutrients and thus play a major role in metabolism and nutrient cycling of the entire ecosystem (Prenger and Reddy, 2004). Moderate to low P conditions are frequently observed in the STA flow-ways, hence the importance of microbial phosphorus cycling in achieving the ultra-low phosphorus concentrations essential to meet the WQBEL needs to be investigated.

Organic molecules are mineralized or broken down into simpler compounds via microbial metabolism which will influence P bioavailability, e.g., release of inorganic P from DOP forms. This process is catalyzed by various enzymes; two enzymes responsible for catalyzing the breakdown of DOP are phosphomonoesterase (phosphatase) and phosphodiesterase enzymes. Phosphomonoesterases catalyze the breakdown of sugar phosphates and mononucleotides (Turner et al., 2002; Turner and Newman, 2005). Diesterases are responsible for the breakdown of phospholipids and nucleic acids which are typically a small portion of soil organic P due to their fast turnover rates (Anderson, 1980). In earlier studies, phosphomonoesterase was more frequently measured in water and sediment (e.g., Kang et al., 1998; Wright and Reddy, 2001; Frencoeur et al., 2006) under the assumption that phosphomonoesters are the dominant organic P form in wetland ecosystems. However, recent studies reported that both STA1W and WCAs 1 and 2A had large portions of phosphodiesters (Pant et al., 2002; Turner and Newman, 2005), and high phosphodiesterase activity could be a critical determinant of SRP release that subsequently affects water quality. Similarly, preliminary evidence suggests phosphomonoesterase and phosphodiesterase enzymes will be important in P turnover within the STA-3/4 PSTA cell (Zamorano et al., 2014).

Enzyme activities are influenced by various factors, such as temperature, hydrology, soil chemistry, substrate availability, and vegetation once the enzymes are released into the soil (Sinsabaugh, 1994). Chemical composition of plant litter also alters enzyme activities depending on the nutrient requirements of microbes (Kourtev et al., 2000; Güsewell and Freeman, 2005). For example, Typha and Cladium had different nutrient turnover rates and enzyme activities, mainly due to litter quality differences (Corstanje and Reddy, 2006). Similarly, Kourtev et al (2002) demonstrated individual enzyme activities differed significantly among litter from four species as well as among different soil characteristics.

The limited data available of microbial activity and the magnitude of its influence in P cycling elsewhere in the STAs is scarce, thus additional investigations of important microbial pathways that result in P turnover are essential. Determining different enzyme-induced hydrolyses activity could indicate the potential bioavailability of P compounds in the system components, e.g., water column, floc, and soils.

Breakdown of the remaining DOP and POP, particularly at the lower reaches of the treatment train, will further reduce the outflow TP concentration. An understanding of the mechanisms and drivers for microbial decomposition will provide useful information for choosing strategies for STA improvement and assessment of the ecological significance of the P composition of water discharged to the Everglades. The role of microbial activity in internal P cycling in wetlands can be assessed with measurement of the enzyme activity in surface water, floc, and soil (Newman and Reddy, 1993). Since phosphomonoesterase and phosphodiesterase activities contribute to inorganic P release in aquatic system, assessing their activities in the STAs will provide information on the bioavailability and stability of P along the STA flow-way. Also, since enzyme production and activity are influenced by substrate enrichment and limitation (Sinsabaugh, 1994), it will also be important to assess carbon availability.

3.3.5 Periphyton

A benthic periphyton community can obtain nutrients directly from the water column as well as via diffusion from the sediment, and therefore will influence the exchange of nutrients across the sediment water interface (Hansson, 1989; Newman et al., 2004). Periphyton assemblages can play several roles that lead to increased retention of nutrients, including removing nutrients from the water column (epiphytic dominant versus epibenthic) and cause a net flux towards the sediments, slowing water exchange across sediment/water column boundary thus decreasing advective transport of P away from sediments, intercepting P diffusing from sediments or senescent macrophytes, cause biochemical conditions that favor P deposition, and trap particulate material from the water column (Dodds, 2003). Periphyton can assimilate both organic and inorganic forms of P (Bentzen et al., 1992). A chemical analysis of periphyton from the Everglades indicated that less than 20 percent of the total P was associated with Ca and Mg and the remaining P was in organic forms (Scinto and Reddy 1994), suggesting active biological uptake by periphyton. McCormick et al. (2006) indicated that the rate of algal decomposition, and subsequently P return to the water column, is generally regarded as being relatively fast compared to macrophyte decay. However, P precipitates onto periphyton mats as insoluble inorganic complexes are relatively stable pools of P (Scinto and Reddy, 2003). While there have been several publications documenting the importance of periphyton in nutrient reduction in wetlands, the biogeochemical factors affecting the establishment and effectiveness of periphyton in the STAs for sustainable achievement of ultra-low level P at the outflow has not been studied.

3.3.6 Particulate Processes

A major P removal mechanism of constructed wetlands is the removal of suspended and settleable particulates moving through the wetland. Low velocities, coupled with vegetation resistance, promote fallout and settling of the particulate matter that may or may not contain P (Reddy and DeLaune, 2008; DeBusk, 1999). The dominating retention process was sedimentation, and a large proportion of inflowing particles settled immediately upon entering the wetland, i.e., 78 percent of the P load was found in the sediment near the inlet (Johannesson et al., 2011). Particulate P in the STAs can include P sorbed on soil particles and organic matter eroded during runoff from cultivated lands (Sharpley, 1999), P incorporated in detrital matter, and P in resuspended floc or soil particles. In the STAs, PP in floc and the water column accounts for a large and highly active pool of P and roughly half of outflow P (Dierberg and DeBusk, 2008). Therefore, STA performance is likely dependent on the processes controlling particulates. The production, resuspension, transport, and settling (including burial) can vary as a function of a variety of factors: flow velocities (Harvey et al., 2009), vegetation density and biovolume (Larsen et al., 2009), microtopography (Choi and Harvey, 2014), standing stocks and density of particles (Choi et al., 2013), bioturbation, and other ecological variables. In their evaluation of PP transformations in STA-1W and STA-2, Dierberg and Debusk (2008) reported differences in PP transformation patterns between the two systems and suggested that the STA P loading rate history is a prominent factor in dictating both the total mass of PP, as well as bioavailable fraction of PP, discharged from the STAs.

Resuspension, although not viewed as critical in shallow wetlands with low water velocities, can be significant in the STAs during certain events, e.g., storm events or during operation of water control structures. Reddy et al. (1999) stated that resuspension and transport of settled sediment is unlikely in wetlands, except under high flow velocity, which may occur during an extreme weather event. However, Kadlec (1999) suggested that the potential for erosive velocities exists in highly loaded wetlands, with high length-to-width ratios or large wetlands with long travel distances. In the STAs, wind-driven turbulence and movement of fauna can also cause resuspension. The nature of accrued floc material in both cattail and SAV-dominated cells results in resuspension of the floc material with little turbulence. Generally, the levels of total suspended solids (TSS) observed at STA outflows are at or below detection limit of 3 mg/L. However, high amounts of TSS have been observed in STAs after storm events or vegetation dieoff, such as that observed in STA-1W Cells 2B and 4 in 2004-2006 or STA-1E Cell 6 in 2009-2010 after cell-wide loss of SAV. Gas lift, caused by accumulation of dissolved gases under anaerobic condition, can also cause resuspension.

When particulates containing P stay within the water column, they become part of the measured TP at the outflow structures. Particulates containing P have the potential to re-suspend or be dislodged from vegetation, particularly at the lower reaches of the flow-way, therefore contributing to the measured TP at the outflow. Nonreactive PP (NRPP) is part of the total water column phosphorus and is an important part of the phosphorus fractionation of wetland sediments (Reddy et al., 1993). Anthropogenic P could be transported long distances downstream in particulate form. Previous observations in the STAs indicate that in EAV cells, floc and suspended particulates in the water column are generally more organic and have lower bulk density than those in SAV cells where more mineral matter is accrued. The differences in the characteristics of floc in these different plant communities affect water column P directly and indirectly. The high amount of organic matter in floc in predominantly EAV areas likely allows for soil aggregation into larger particulates, particularly when the water level is drawn down (Pant and Reddy, 2001). Floc formed within predominantly SAV communities, which has less organic matter content and higher bulk density, tend to aggregate less (Yang et al., 2009). This makes this accrued material prone to resuspension even when the water level is drawn down. Another indirect effect of high TSS and dissolved solids is water column turbidity, which can negatively impact the health of SAV communities. Dierberg and DeBusk (2008) found substantial removal of PP in the STAs and that the nature and bioavailability of particles can change during transport through these systems. They found variability in labile PP gradient between STA-1W and STA-2, with STA-2 showing a more definitive gradient from the inflow to outflow locations.

Due to the important role that PP has on the overall P cycling and transport in the STAs, a better understanding of the drivers and factors influencing particle dynamics (entrainment, settling, and resuspension) is critical when developing strategies for further reduction of outflow TP concentrations. Management options aimed at increasing settling rates and reducing resuspension could potentially improve STA performance.

3.3.7 Vegetation Pathways

Each cell within the STAs contains a mixture of EAV, SAV, FAV, and periphyton. The proportion of these vegetation types varies depending on target vegetation community for each cell, microtopography, and vegetation management activities. Generally, most STA flow-ways are configured so that the upstream cell is primarily EAV while the downstream cell is primarily SAV. An exception is STA-2 where three of the original flow-ways are configured as single cell areas with either EAV (Cell 1) or SAV-dominated (Cell 3), while Cell 2 was recently transformed into EAV-SAV community.

Emergent aquatic vegetation uses root uptake as their primary source of P into plant biomass but most plant P is returned to the water column following death and decomposition (Richardson and Marshall, 1986). Phosphorus not released from wetland plant tissues during decomposition is ultimately buried within the peat soil. Davis (1982) reported that only about 2 percent of P is stored in live plant tissue and approximately 30 percent of P removed from water column is stored in decomposing litter and 50 percent of the removed P is buried in sediments and soils for a long-term storage. The decomposing litter produced by emergent species provides a substrate for microbial communities and in the process consuming P from the water column (Kadlec and Wallace, 2009; Richardson et al., 1997).

Generally, since SAV and the associated periphyton community can sequester P rapidly and can store it within SAV biomass, other researchers have stated that SAVs are more effective than EAV at removing P from the water column in wetlands (Brenner et al., 2006; Knight et al., 2003; Gumbricht, 1993a; Dierberg et al., 2002; Knight et al., 2003). Kadlec and Knight (2009) indicate that SAV has been shown to be more effective than EAV for phosphorus removal at low concentrations (about 100 µg/L) in a subtropical climate. However, SAV has been shown to be less effective at higher phosphorus concentrations (about 1000 µg/L). Dierberg et al. (2002) indicate that native SAV communities are more efficient at removing P in lower P environment (<100 µg/L) than EAV communities. The long-term performance of STA-2 Cell 1, a predominantly cattail with sparse sawgrass community contradicts the previous findings, achieving POR outflow TP flow-weighted mean (FWM) concentrations of 14 ppb compared to the adjacent SAV cell (Cell 3) that achieved 18 ppb.

There are several mechanisms associated with P sequestration by SAV. Dense SAV beds may physically filter, detain, and cause settling of suspended solids which may contain organic and inorganic P. SAV may assimilate P directly through their thalli, shoots, and leaves (Graneli and Solander, 1988), a process that extracts P from soil/sediment nutrient reservoir. Dense SAV beds also provide extensive surface area for periphyton colonization which removes nutrients directly from the water column. Also, elevation in pH resulting from intense SAV and periphyton photosynthesis can lead to $CaCO_3$ super-saturation, and therefore facilitate removal of P via co-precipitation (Reddy et al., 2002). Calcite precipitation promotes sedimentation of periphyton and P through adsorption and incorporation into nucleating calcite crystals (Koschel et al., 1983; Kufel and Kufel, 2002; McConnaughey et al., 1994; Pietro et al., 2006), and can serve as a nucleus for the formation of Ca–P compounds (Brown 1980). Non-rooted SAV (such as *Ceratophylllum demersum* and *Chara* sp.) uptake dissolved nutrients directly from the water column (Barco and James, 1997; Wetzel, 2001). Rooted SAV species, such as *Najas* sp. and *Potamogeton* sp. can take up P from the soil and water column through both its roots and above ground biomass, although root uptake from the interstitial water of the soil or sediments is the dominant path due to higher levels than in the water column (Wetzel, 2001; Chambers et al., 1989). A healthy SAV community within the water column allows it to remove nutrients from the water column effectively and minimizes the effects of hydraulic short-circuiting. When P loading into the STAs was controlled such that overloading was prevented, SAV cells and associated periphyton demonstrated outflow P concentrations could be reduced to below 20 ppb (Dierberg et al., 2002; Pietro et al., 2006a).

The form of P taken up readily by plants is inorganic P, i.e., SRP, which is then assimilated and converted to plant tissue, then returned to the water column upon decomposition (Kadlec and Wallace 2009). Decay and translocation processes release most of the P uptake, with the residual accreting with sediment and soil accretion. The net uptake that is eventually buried as recalcitrant residual phosphorus is only a very small fraction of the gross uptake (Kadlec and Wallace, 2009). While there have been a few attempts to compare the performance of EAV versus SAV communities in reducing outflow P concentration, the sparse data set and variable field condition has limited the interpretation of such comparison within the STAs. There has also been limited information to understand the biogeochemical characteristics associated with the EAV and SAV communities' P reduction mechanisms. Understanding these characteristics, particularly in the lower reaches of the treatment train, is important for STA optimization efforts.

3.3.8 Faunal Pathways

Research and strategies for managing for P retention in constructed wetland treatment systems have, so far, focused on plants, soils, and microorganisms. However, there are several studies in other wetland and lake systems that suggest the internal production and consumption of P through faunal pathways, e.g., fish, alligators, and water birds, may significantly influence P cycling in aquatic systems. Aquatic fauna such as fish and macroinvertebrates can influence water column P by three primary routes. First, they may change the trophic structure of the water column of a lake, pond or wetland. For example, the reduction of large zooplankton (i.e., efficient algal grazers) like *Daphnia* by abundant planktivorous fishes can enhance phytoplankton establishment and is believed to recycle P in the water column at a high rate (Vanni, 2002). Furthermore, the shading by phytoplankton may reduce growth and cover of macrophytes in shallow systems and destabilize sediments which may lead to additional wind-generated re-suspension of P. Second, fish and invertebrates excrete SRP and when this P comes from benthic sources (e.g., detritus and algae) it serves as a source of new or translocated P (Havens. 1993; Vanni et al., 2006). Finally, benthic organisms (e.g., alligators, catfish, amphiumas, crayfish) may stir up the sediments in wetlands, lakes or ponds and resuspend organic and inorganic solids (Breukelaar et al., 1994; Roozen et al., 2007).

The effects of fish excretion are considered in empirical and bioenergetic models, while the net effects on water column P levels have been addressed in manipulative experiments (replicated studies in enclosures or ponds) and with whole lake fish removal biomanipulation efforts in the USA and Europe. In the majority of the whole lake studies, fish removal resulted in a reduction of the external loading of P, primarily due to reduced excretion of P and reduced bioturbation.

As with aquatic fauna, birds can alter several pathways leading either to higher or lower TP concentrations in the water column of shallow aquatic ecosystems. Direct effects occur when birds ingest nutrients and either egest them as feces (particulate nutrient flux), excrete them as urine (dissolved nutrient availability and flux), or assimilate them into their bodies (nutrient sequestration) while indirect effects include effects on prey abundance or species composition, modification of the physical environment (e.g., bioturbation), and additional propagated effects on other species (Vanni, 2002). Birds take in P from the fish, invertebrates, or plant material that form their prey base, assimilate some portion of it, and excrete or egest the remainder. The location of P deposition by wetland birds therefore depends on their movements, which are influenced by the stage of the nesting cycle, migratory movements, and prey availability, among many factors. Wading birds are known to concentrate P at nesting and roost sites within the Everglades (Powell et al., 1991; Frederick and Powell, 1994) and elsewhere (Bildstein et al., 1992; Montes et al., 2011) and it has been shown in a variety of studies that carnivorous birds can have strong localized effects on P concentrations in water, soils, and sediments (Marion et al., 1994; Scherer et al., 1995; Mukherjee and Borad, 2001; Ligeza and Smal, 2003; Hahn et al. 2007). Herbivorous waterbirds, including geese and ducks, can also be a major source of nutrient enrichment in wetlands, often translocating nutrients from terrestrial habitats into wetlands or between wetland areas (Gere and Andrikovics, 1992, 1994; Manny et al., 1994; Marion et al., 1994; Scherer et al., 1995; Hahn et al., 2008).

The STAs support a highly abundant and species-rich avian community that is about 35 times the density of birds than that of the natural marsh when averaged across all seasons (Gawlik and Beck, 2010). Among the most numerous are the waterfowl, which can exceed tens of thousands in some areas of the STAs. Coots and many dabbling duck species forage on SAV, e.g., hydrilla, and their presence can be associated with greatly reduced biomass of SAV. Thus, waterfowl may have an important indirect effect on STA functioning by modifying the SAV community. Also common in the STAs are fish-eating birds, especially wading birds such as herons, egrets, and storks. Given their important role in the redistribution of nutrients in wetlands, wading birds are or have the potential to be net exporter of P from the STAs. The TP content of wading bird feces (>10% P) can be high relative to other animals (Marion et al., 1994) and the export, import, and recycling of their feces may play an important role in STA phosphorous dynamics and outflow concentrations. While there is knowledge on different faunal mechanisms that have the potential to control P, the relative importance of these mechanisms in the STAs particularly at low P environment, the different factors controlling these mechanisms, and whether we can optimize them using different management scenarios in the STAs is unknown.

3.3.9 Potential Management Options

As discussed in the preceding sections, net P reduction in the STAs is a product of complex interactions of among many pathways and influenced by many biotic and abiotic factors. While many of the mechanisms and factors influencing P reduction in the STAs may be difficult to control, information gained from studies described in this document may lead to some new or enhanced management strategies. These potential management options that can be evaluated using the studies covered under this plan are described below:

Vegetation Management – Vegetation communities can be manipulated to alter water movements in the STAs. If the study indicates that particulate transport is significantly influencing outflow concentrations, then vegetation could be manipulated to reduce preferential flow paths, reduce particle entrainment, and reduce resuspension, from wind events or high flows events. If certain vegetation communities allow for accumulation of more stable P forms, the flow-ways could be managed for those communities. It could be possible to enhance further breakdown of organic P early in the flow-way by creating greater spatial diversity in EAV and SAV combinations within the flow path. If waterfowl are found to be causing higher P concentrations in open areas, the District could encourage habitat mosaics or manage for SAV species that are less attractive to waterfowl.

Hydrology and Hydraulic Management – If findings show that flow rates, or the absence of flow, affect P concentrations in the STAs, operation may be enhanced to provide favorable rates. Continuous low flow could reduce local hydraulic speed zones, thus reducing particle entrainment. If findings show that water depths affect P concentrations in STAs (due to increased P uptake, decreased flux, particulate settling or UV light penetration), the District could manage depths to optimize stages before and after flow events.

Topography Management – If studies show that topographic variation within the cell results in significant variation in P reduction due to uneven flow distribution, dryout of portions of the cell, or unfavorable vegetation community distribution, improving topography of a cell may result in enhanced P reduction performance. If findings show significant particulate transport at the lower reaches of the flow-way, creating particle settling hotspots in relatively flat areas or deep traps could help reduce PP at the outflow.

Soil Management – If results show that internal P loading is significantly affecting outflow TP concentration, then soil management (e.g., inversion of active soil layer or removal of the active soil layer) may alleviate the internal loading issue.

Fauna Community Manipulation – If preliminary results show significant contribution of fauna on the overall P cycling, particularly at the lower reaches of the treatment train, further studies may be necessary to further understand their role. Manipulation of vegetation community and hydrologic condition, as well as promoting enhancement of hunting and fish removal programs are ways to manage faunal communities. Reduction of water level during cold fronts to may allow water temperatures to drop below 10°C, the critical survival threshold for many exotic fish species. Lower water levels will also attract foraging wading birds such that they can transport aquatic fauna from the outflow cells to breeding colonies.

3.4 HISTORICAL DATA MINING AND ANALYSIS

A large amount of data has been collected since inception of the first STA in 1995 including:

- Hydrologic and hydraulic – stage, flow, rainfall
- Water quality - nutrients (P and associated species), major ions, physical parameters
- Soils – spatial data for C, N, P, and basic soil characteristics; soil accretion and P accretion (limited data); soil P stability (limited data)
- Porewater composition (limited data)
- Vegetation – coverage, density (limited data), biomass (limited data), and tissue composition (limited data)
- Microbial activity and decomposition (limited data)
- Fauna – limited to avifauna and fish species and abundance (literature search)
- Topography – ground elevation
- Operational and disturbance history for each of the STA cells
- Results from biogeochemical studies and processes (limited data)

A majority of these data have been collected by the District and stored either in DBHYDRO or ERDP databases, or individual files. Data are also available from a limited number of private and university consultants who have done work in the STAs. Additional information and data can be obtained from reports. All available data pertinent to the study objectives and questions will be gathered, along with the essential metadata, e.g., sampling location, date and time of day of sampling, method detection limits, sampling method, analytical methods, and study details will also have to be assembled. This important initial step will require a coordinated effort among the project team members, contractual scientist, and data steward. Once all relevant data sets are compiled, statistical summary of the data will be essential to determine data distribution, measure of central tendency, variability (i.e., seasonal or annual) and total number of observations for each location of interest. This summary should also provide information regarding limitation of the datasets to the required statistical analyses.

Based on the conceptual model, the deficiencies in dataset, i.e., missing information for critical components or drivers, as well as variables with greatest uncertainties will be highlighted (e.g., internal flow values, limited information on biogeochemical processes), so that these data can be collected while analyzing existing data and subsequently incorporated into future modeling and analysis. Some of the approaches to the data analysis include (1) examination of temporal and spatial trends, (2) multivariate analysis to assess trends in P uptake performance as a function of different factors, (3) stepwise regression analysis, and (4) a retrospective power analyses to determine if the data sets were sufficient to detect observed differences and trends, if statistical differences are not observed.

Along with data mining, further review of published information, i.e., reports and peer-reviewed journals, will also be conducted to review and summarize the types of studies that have been done in the past related to STA performance or any of the components of interest to this study and their outcomes. The expected products of this effort will include:

- All relevant period of record data that has gone through data quality assessment process and placed in a central location within the District’s database infrastructure;
- A report on temporal trends and spatial distribution of key variables and their relationship with P reduction;

- A summary of data gaps and uncertainties relevant to the project questions and critical components of the draft conceptual model;
- Recommendation on sampling locations based on the spatial distribution of key environmental factors; and
- A publication summarizing the findings of the data mining process.

3.5 CONCEPTUAL MODEL DEVELOPMENT

Given the complexities of P cycling with multiple mechanisms and drivers at multiple scales, e.g., from microbial through STA-level, multi-tiered conceptual models are being developed to guide the study. The current version of Tier 1 conceptual model is presented in **Appendix A**. Specifically, the models will help refine our comprehensive list of potential research questions, prioritize the questions, and identify gaps in our knowledge. The conceptual models encompass processes affecting water P cycling at the landscape (e.g., STA level), sampling area (e.g., cell or flow-way level), and sampling unit scale (e.g., individual transect site, mesocosm, soil core). Development of Tiers 2 and 3 models is underway, including adding microbial community dynamics and processes (e.g., P uptake, cycling rates), mechanisms of P speciation (e.g., size fractionation, molecular organic constituents), fine-scale sediment dynamics (e.g., settling, diffusion, or resuspension), and processes at the individual plant-level (e.g., plant uptake, litter formation, and decomposition). For each tier, a list of state variables, flux variables, drivers, and rate modifiers are identified, and more detailed questions pertaining to the key component of each model are developed and evaluated. Each conceptual model component and related science questions will be evaluated with respect to the following criteria: data availability; testability; degree of understanding and predictability; and sensitivity to influence P concentration, cycling, and transport within the water column. The evaluation process will be done by a sub-team comprised of District scientists, modelers, and consultants. The outcome of evaluation for each model will then be used to prioritize research areas.

3.6 STUDY HYPOTHESIS AND APPROACH

The forms of P and transformation of various P forms in the STAs is a result of complex biotic and abiotic processes as shown by the numerous pathways within the conceptual model (**Appendix A**). The research activities within this study plan have been organized into four key study areas, generally in concert with some of the components within the conceptual model:

I. P Speciation and Transformation
II. Particulate Processes
III. Vegetation Pathways
IV. Faunal Pathways

A summary of the linkages among the study areas I to IV, hypotheses within the individual study areas, and the specific research sub-study (appendices) is presented in **Table 3-1**.

Table 3-1. Summary of linkages between study areas, hypotheses, and specific sub-studies.

Study Area	Hypotheses	Appendices										
		C	**D**	**E**	**F**	**G**	**H**	**I**	**J**	**K**	**L**	**M**
I	**I-1.** The ability of an STA flow-way to remove SRP, convert DOP to SRP, and retain PP early in the flow-way will reduce TP concentrations at the lower reaches of the flow-way and at the outflow.	x	x	x	x	x	x	x	x	x	x	
I	**I-2**. Long-term retention of P in the STAs is a function of the distribution and concentration of labile versus recalcitrant forms of P that accumulate in the soil.	x	x	x			x	x	x		x	
I	**I-3**. Microbially-mediated P transformations are essential to reach low TP concentrations in the lower reaches of the flow-way.		x	x		x				x	x	
I	**I-4**. Internal P fluxes in lower reaches of an STA flow-way significantly increase water column P and has significant impacts on the flow-way outflow P concentrations.		x	x		x	x	x	x		x	
II	**II-1**. Outflow TP in STAs can be reduced by increasing settling and burial of PP.		x	x	x	x	x				x	
II	**II-2**. Outflow TP in STAs can be reduced by preventing or decreasing resuspension of PP into the water column.		x	x	x	x	x					x
III	**III-1**. There is an optimal spatial vegetation pattern of EAV and SAV for P retention in terms of facilitating particulate settling and Ca-P co-precipitation, controlling internal flux, generation of stability of stored P in soils, and microbial activities.		x	x	x	x	x				x	
IV	**IV-1**. Fauna impacts P concentrations in the water column of the outflow cells via consumption and excretion, bioturbation, translocation, and herbivory.		x			x					x	x

3.6.1 Study Area I: P Speciation and Transformations

The ability of the STAs to achieve low outflow TP concentrations depends on the system's capacity to reduce P, by transforming P from one form to another, with P bioavailability and recalcitrance being important controlling characteristics. Understanding the speciation of P is a precursor in knowing the sources and fate of the different P species. However, the quantification of P species and their lability has been hindered by methodological limitations. Both water column and soil methods are evolving such that more accurate distinctions may now be possible and the relationship between chemical structure and biological transformations established. Overall, these hypotheses focus on identifying forms of P, the mechanisms and factors controlling their transformations, and soil to water column flux. The investigation of the microbial community, in particular enzymes associated with the cycling of C and P, is also included as part of this study area.

Hypothesis I-1. The ability of an STA flow-way to remove SRP, convert DOP to SRP, and retain PP early in the flow-way affects TP concentrations at the lower reaches of the flow-way and at the outflow.

Studies will examine and quantify P forms along the nutrient/flow gradient and within different vegetation communities (SAV/EAV) to determine whether there are different forms that were created *in situ* or, less likely, remain unchanged from inflow. The SAV complex, as well as the less abundant EAV associated microbial community, can remove P directly from the water column, however, given the energy required to transform DOP and particulate organic P (POP) to SRP, transformations are likely occurring only in the region of the flow-way where SRP is no longer available. A comparison of P species composition in the water column within the STAs to areas of known P enrichment and limitation in WCA2A will allow an assessment of the transformation potential, or recalcitrance, of P species. Surface water samples will be collected and analyzed for the different P species and other parameters that affects P transformations, e.g., calcium concentration, light penetration, pH, and alkalinity (**Appendices D** and **E***)*. The information gathered from data mining on spatial variability of soil characteristics, vegetation patterns, and other relevant factors will serve as a basis in deciding the final number and location of transect sites.

The quantification of PP transport, an essential component of P cycling, is discussed under Study Area II (Particulate Studies) and **Appendix F**. Results gathered from studies described in **Appendices H** through **M** will also provide for an assessment of factors that affect the concentration of the different species.

Hypothesis I-2. Long-term retention of P in the STAs is a function of the distribution and concentration of labile versus recalcitrant forms of P that accumulate in the soil.

Due to differences in P reduction mechanisms, the characteristics and composition of soil accumulating in EAV areas has distinct characteristics from that accreting in SAV areas. Soil in EAV areas is generally highly organic, while in SAV areas, it is generally highly mineral**.** Plants with greater structural complexity will produce carbon compounds that are more complex, e.g., lignin, while algal derived forms will be rapidly decomposed. In contrast, the SAV facilitated Ca and P co-precipitation will produce forms of P that may be more stable to environmental changes because they are less susceptible to oxidation, turnover is more a function of erosion and dissolution. A controlled vegetation study in WCA2A demonstrated that compared to cattail plots, actively managed enriched SAV plots had increased enzyme activity, increased inorganic P forms, and after several years, significantly lower TP concentrations in the water, floc and surface soils (Newman et al., 2013; DB Environmental, 2014). A similar comparison of inorganic versus organic contents was observed in STA-2 while less consistent results were observed between these communities in STA-1W (Bhomia et al., 2012). It is also anticipated that the concentration of labile P will be higher in the front end of a flow-way where the P loading and concentration are relatively higher than the lower reaches of that treatment flow-way.

Investigations for this hypothesis will involve examination and quantification of the different P forms in the floc and soils along the nutrient/flow gradient and different vegetation communities (SAV/EAV) to determine whether these differences potentially affect the recalcitrance of the accreted P **(Appendix C, Appendix D,** and **Appendix H**; also CHIP Project). A comparison of the EPC_o values along the nutrient gradient will allow a spatial evaluation of the system's ability to be a source or sink for P (**Appendix I**). An assessment of the diffusive P flux rate along the nutrient gradient will also be conducted **(Appendix J)**. An integrated analysis of the total storage of the different P species and the flux rates will allow for an estimation of short-term and long-term potential P release from the soil to the water column at along the treatment flow-way. Vegetation density and biomass will influence P accretion; therefore this study will also be co-located at sites with detailed vegetation surveys **(Appendix L)**. A comparison of these results with those collected along a 35+ year nutrient enrichment gradient will allow the verification of long-term P storage and release **(Appendix E)**.

Hypothesis I-3. Microbially mediated P transformations are essential to reach low TP concentrations in the lower reaches of the flow-way.

Organic P forms are important and often dominant constituents of the total P pool in the floc, soils, litter, and water column. The breakdown of these organic compounds is facilitated by microbially mediated enzymatic activity; either directly to obtain inorganic P, or indirectly through C or N acquisition. Microbial P transformations are influenced by numerous factors, including C and P availability, pH, temperature, hydrologic conditions, and substrate quality. Seasonal and temporal quantification of microbially mediated P transformations will be incorporated into *in situ* assessments along the inflow to outflow gradient, in different vegetation communities, and under different P concentrations and hydrologic conditions to assess conditions that optimize microbially mediated organic P breakdown **(Appendix K)**. Phosphorus transformations are strongly controlled by the P supply rate, therefore enzyme activity and the turnover of water column DOP and POP will be assessed under low and high flow as well as stagnant conditions. Results from studies described in **Appendices D, E, K,** and **L** will also provide input in assessing the influence of the different environmental factors on microbial activities.

Hypothesis I-4. Internal P fluxes in lower reaches of an STA flow-way significantly increase water column P and have significant impacts on the flow-way outflow P concentrations.

Internal P fluxes vary spatially and temporally, and is controlled by a combination of physicochemical and biological processes influencing water column P. Soil biogeochemical factors (e.g., desorption and sorption, mineral content), will be quantified by examining soil biogeochemistry parameters, and P fractionation **(Appendices D, H,** and **I**; also PSTA Study). Because vegetation and bacterial dynamics, e.g., root mining, P uptake from water column, and hydrologic conditions directly influence P flux rates, studies will be conducted within different vegetation habitats, at different points in the growing seasons and under stagnant, low and high flow conditions using a combination of benthic chambers, porewater profiles and soil cores **(Appendix J)**. Results from studies described in **Appendices H, I, J,** and **L** will also provide input in assessing the influence of the different environmental factors on P flux process.

Stop/Go:

1. If no significant differences in P species composition are observed within the STA study sites or selected WCA2A sites, it will be assumed that detailed P speciation data are not necessary to assess P turnover, and further detailed P speciation assessments using the method described in **Appendix C** will be discontinued. If compositional differences are distinct, mesocosm studies to assess factors controlling their degradation will be conducted **(Appendix N).**
2. If the initial results for flow-way measurements (**Appendix D**) show insignificant difference among sites, the number of sites will be reduced for remaining measurements.
3. Flow regime imposition is dependent upon water availability and the District's operational stormwater flow needs. If target flow condition, particularly the desired high flow event cannot be achieved during the target study period for a given flow-way, study under high flow event will be delayed until such time that high flows are possible.
4. Based on the results of initial microbial activity measurements, if results for either the water column, floc, porewater, or periphyton remain stable over time, the frequency will be reduced.
5. If significant differences in microbial activities are observed along the transect and between flow-ways, a more in-depth analysis of the microbial communities (i.e., taxonomic identification and controlled substrate study (**Appendix N**) will be conducted.
6. The data mining and chemical P fractionation results will provide information on properly locating sites for the P flux studies, based on the relative distribution of the different forms of P in the soil. Flux sampling or field measurements will focus on transect locations where soil has the greatest potential to release P based on the size of the labile P pool relative to total P.
7. If the baseline soil characterization revealed no significant differences in the spatial distribution of various P sorbing components within a flow-way, the number of soil samples for this study will be reduced accordingly. If significant differences exist, a more focused sampling and analysis to capture true spatial variability in P sorption and EPC will be conducted.
8. If benthic chamber study results are comparable with the results from soil core flux studies or the porewater results, benthic chamber measurements will be discontinued.

3.6.2 Study Area II: Particulate Processes

Particulate P accounts for about half of the P measured in STA outflows; therefore, it is important to understand the processes controlling water column particulate concentrations through STAs. These processes include production (e.g., periphyton sloughing, litter fragmentation), settling (i.e., settling out of the water column or burial into soil), resuspension via entrainment or bioturbation, and horizontal transport. In some cases, management options such as herbicidal treatments or canal-plugging may influence these dynamics to favor greater settling and burial of PP or reduce particle resuspension.

To promote particle settling and reduce resuspension, water flow must be maintained below a critical entrainment threshold (CET), the velocity required to entrain and re-suspend particles, and mechanical disturbances to sediment (bioturbation) must be minimized. In Everglades wetlands, the CET velocity of benthic floc particles typically ranges from 0.6 to 3 cm s^{-1} (Larsen et al. 2009; Sklar and Dreschel, 2014), low enough for strong winds, pulsed or local discharges from structures, or movement of floating mats to re-suspend particles into the water column. In both

Everglades and STA wetlands, the magnitudes of particle settling and resuspension rates remain poorly quantified, though it is well known these processes are controlled substantially by vegetation density and microtopography (Harvey et al., 2009; Choi and Harvey, 2014). Bioturbation by fauna also can remobilize large pools of PP stored in the floc, soil, and standing vegetation. STA management should aim to minimize such disturbances or ensure that other portions of STA flow-ways can effectively settle out resuspended particles.

This research study focuses on factors controlling PP settling and resuspension and their net effects on water column TP, from inflow to outflow. The information gathered from this study will provide useful information when evaluating specific management options, e.g., optimal flow operation, desirable vegetation patterns, and soil management, to enhance particulate settling and entrainment and reduce resuspension, particularly at the lower reaches of the treatment flow-ways. Specific studies that look at these are in **Appendix F**.

Hypothesis II-1. Outflow TP in STAs can be reduced by increasing settling and burial of PP.

Reducing PP in the water column, particularly at the lower reaches of the treatment flow-ways and the outflow locations, requires optimal hydrologic and environmental conditions conducive for settling particles or favoring processes that bury surficial sediments. Factors that could favor settling and reduce resuspension are: i) flow velocities below the CET for benthic floc, ii) low water levels, iii) optimal vegetation density and/or vegetation patterns, iv) microtopographic variation; v) better soil aggregation, i.e., higher density particulates; vi) mechanically disking in surface sediments, or, if feasible, removal of the topsoil layer containing easily resuspended particles. Quantifying particle dynamics along selected flow-way transects will be the first step to evaluate some of these factors (**Appendix F**). As part of the transect study, water column particulate concentrations, particle settling rates, and water velocities will be quantified from inflow to outflow locations to determine the factors most effective in promoting settling and most amenable to manipulation for potential management purposes. For instance, variation in vegetation density within a flow-way could be used to evaluate its effectiveness in promoting settling and reduction in water column particulates. Sampling will focus on outflow portions of flow-ways, but will include central and inflow areas to provide context for understanding outflow dynamics. Data collected from Study Areas I and III, specifically those described in **Appendices D, E, F, and L** will also feed into this study.

Hypothesis II-2. Outflow TP in STAs can be reduced by preventing or decreasing resuspension of PP into the water column.

Factors that could increase resuspension in the STAs include: i) vegetation die-offs, ii) floating mats which scour benthic floc, damage vegetation, and potentially release their own particulates, iii) high velocities near flow structures, iv) preferential flow paths such as remnant canals, v) wind events, vi) bioturbation, vii) low SAV and EAV cover; and viii) presence of highly organic floc, unaggregated inorganic floc, and unconsolidated soil with low CET. Similar to the studies focusing on particulate settling (above), particle dynamics measurements along the study transect sites will be used as a first step to evaluate resuspension as a function of variation in these factors (**Appendix F**). In some cases, event-based sampling may be required, e.g., sampling after faunal disturbances, wind events, or extended draw-downs. Data collected from Study Areas I, III, and IV, specifically **Appendices D, E, and F** will also feed into this study.

Stop/Go:

1. Results from data mining and preliminary field measurements will be used to prioritize studies for key factors controlling settling and resuspension and in determining the study sites and number of samples/measurements.
2. Results of the initial transect sampling will provide STOP/GO points for follow-up controlled studies (e.g., using test cells or flume tests, **Appendix N**) to isolate and evaluate the direct effects of specific factors (e.g., flumes with different vegetation density or SAV type on particle settling; dry-down experiments in test cells to evaluate particle settling and burial).
3. If velocities measured in these areas of high flow are found to be consistently below the CET of the underlying floc and soil, then no further CET measurements will be conducted.
4. If PP is not elevated in any region along the transects, then particulate studies will be discontinued.

3.6.3 Study Area III: Vegetation Pathways

Vegetation is one of the primary mechanisms in STA P removal, either through physical and hydraulic resistance that helps with particulate settling, direct P uptake and eventual burial, or by providing a surface for periphyton and microbial colonization and activity. Previous studies suggest that the mode and scale of net P removal, as well as P storage in accreted floc and soil, differ between EAV and SAV cells, particularly by P species, as different forms tend to accumulate in different vegetation communities. Additional vegetation surveys are needed to make a more accurate assessment of the influence of vegetation type, species, coverage, density, condition, and tissue composition on short-term and long-term P removal.

The focus of this study is on the influence of spatial configuration of vegetation communities (community composition, coverage, density, health, stability, etc.) in flow-way P reduction performance. Specific studies that look at these are in **Appendix L**.

Hypothesis III-1. There is an optimal spatial vegetation pattern of EAV and SAV for P retention in terms of facilitating particulate settling and Ca-P co-precipitation, controlling internal flux, generation of stability of stored P in soils, and microbial activities.

EAV and SAV work differently for P retention, so examining spatial relationships is essential to understanding how a vegetation mosaic can contribute to optimal STA performance. The primary focus of these studies is to provide spatial vegetation data (configuration, density, cover, and tissue nutrients) and relate them to environmental factors (hydrology, hydrologic history, soils, soil history, light penetration, community decline, soil P stability, phosphatase activity, epiphyte growth, etc.) in cells with varying performance to determine how vegetation communities and spatial configuration affect P retention (Study Area I), and to provide spatial vegetation data for other sub-sections of this project (Study Area II - Particulate Processes, and Study Area III – Faunal Pathways). Data acquisition will be via the existing aerial imagery efforts (at ~13,500 feet above the mean terrain), low-altitude remote sensing using an unmanned aerial vehicle (UAV) or helicopter-mounted UAV equipment, ground surveys, and biomass sampling (**Appendix L**).

Stop/Go:

1. This task is anticipated to continue throughout the duration of this study at a frequency defined in **Appendix L**. Although there are no stopping points anticipated, the spatial and temporal frequency may change depending on seasonal condition or if there are no observed temporal or spatial changes after the first year.
2. If initial comparison of vegetation cover assessed from field photos versus actual field vegetation cover assessment (1 m^2 quadrat counts) determine comparability between the two methods, field photos will continue to be taken, but photo analysis for estimated vegetation cover will be discontinued.

3.6.4 Study Area IV: Faunal Pathways

Recent studies show that faunal populations play an important role in the redistribution of nutrients in aquatic environments and can increase nutrient contents in the water column. Animals can have direct and indirect effects on P concentrations. Direct effects occur when animals ingest nutrients and either egest them as feces (particulate nutrient flux) excrete them as urine (dissolved nutrient availability and flux), or assimilate them into their bodies (nutrient sequestration). Indirect effects include effects on the aquatic community (e.g., top down effects on vegetation or herbivores), and modifications of the physical environment (bioturbation). Animals, especially birds, can also be important vectors of P-transport, and are known to translocate large quantities of P within and among wetlands.

The STAs support a highly abundant and species-rich faunal community, and there is a strong possibility that consumers influence our ability to achieve the desired low (<13 ppb) P concentrations in outflows. However, the direct and indirect effects of faunal assemblages on P concentrations in the STAs are not known. The following hypotheses articulate the potential mechanisms by which fauna might influence P-cycling in the STAs.

Hypothesis IV-1. Fauna impacts P concentrations in the water column of the outflow cells via consumption and excretion, bioturbation, translocation, and herbivory.

Herbivorous animals such as waterfowl and fish may increase P concentrations in the water column directly by excretion/egestion or indirectly by reducing SAV cover, biomass and sustainability. Animals that interact with the benthos can move stored P from the sediments back into the water column via modification of the environment, e.g., bioturbation, or via consumption of detritus and excretion. This serves as a new or translocated source of P.

Highly mobile animals, i.e., birds, may translocate nutrients to, from or within STAs. This may be particularly relevant when foraging is focused in the STAs and other activities such as nesting occur elsewhere. Long-lived and large animals, e.g., alligators may act as nutrient sinks, storing large amounts of P in bones and other tissues.

For an initial assessment of the potential impacts of faunal assemblages on P cycling and SAV consumption in the STAs, the following information will be collected: 1) the biomass/community composition of aquatic fauna (e.g., fishes and crayfish), birds and alligators in the SAV cells, and 2) Mass-specific P excretion rates of common fauna, especially benthic and detritivorous aquatic species (excretion rates for birds and alligators will be obtained from published sources). The biomass and excretion results will then be combined to estimate areal P excretion by the entire faunal assemblage, i.e., rates of P released to the water column through excretion. Specific studies that address these hypotheses are detailed in **Appendix M**.

Stop/Go:

1. If the biomass of fish/invertebrates/birds results in significant net P excretion rates in relation to P inflows and other sources of P (Study Area I), then additional manipulative studies may be needed to determine if this consumer derived P contributes significantly to TP (potential new RS Science Plan study). That is, whether the excreted P substantially contribute to TP at outflows or is recycled by autotroph) with little overall effect on TP. If net P excretion is not significant, then the assumption is that faunal excretion is not influencing P loading and further studies will not be necessary.
2. If the community composition of fish/invertebrates contains a large biomass of detritous/benthivorous or burrowing species then additional manipulative studies may be needed to 1) quantify how much ‘translocated’ P contributes to outflow TP (Study Area III); and 2) to test biomanipulation scenarios for controlling such species (potential new RS Science Plan study). A relatively small biomass would preclude such studies.

3. If the biomass of herbivorous animals (waterfowl, fish, crayfish, apple snails) is found to have the potential to impact SAV, then manipulative studies may be necessary to (1) determine their effects on SAV sustainability and biomass (Study Area III), and (2) to test biomanipulation scenarios for controlling herbivory using experimental enclosures/exclosures (potential new RS Science Plan study).

3.7 DATA MANAGEMENT

Due to the complexity of this project and the fact that it involves many project participants, including District scientists and external contractors, it is critical to have a centralized location for all data gathered during the data mining process, method development, environmental and ecological surveys, transect studies, and controlled experiments. The project team will work closely with the designated data steward.

Water quality data produced by the District’s laboratory will be loaded into DBHYDRO. The District’s laboratory staff performs the necessary data verification prior to loading to DBHYDRO. Ecological data, other than water quality, will be loaded into ERDP database. The study Project Manager or designated scientist will perform data validation prior to loading into the database. Any data, metadata, figures, maps, or other related information that is not compatible with either ERDP or DBHYDRO databases will be loaded into MORPHO. The data steward will be in charge of controlling the data loading and distribution.

3.8 DATA SYNTHESIS, INTEGRATION AND ANALYSIS

Comprehensive understanding of internal processes functioning in STAs is essential in identifying the key P removal pathways and biogeochemical processes identified in the conceptual models. The objectives of this task are to synthesize results from previous studies and the different sub studies within this present project, analyze data to quantify rates and constants, e.g., flux rates and P transformation rates, and develop parameter equations for critical components, i.e., key P removal pathways and biogeochemical processes, of the conceptual model. In addition, data gathered from this study will help enhance current mass balance model formulations and may be used to refine model algorithms to capture the effects of important ecological and biogeochemical processes that are not currently represented in models. Development of any new models or refinement of any existing models is outside the scope of this project.

Statistical analyses to determine significance of trends, relationships, and differences in effects of different variables will be done using SAS and JMP software. Spatial analyses of results, where applicable, will be done using ARC GIS software.

3.9 REPORTING

The project Study Leader will meet with the project team and contractors on a routine basis to help ensure that the project stays on time and on budget. Monthly reports will be required from each of the task leads and the contractors. The Study Leader will summarize the findings and status on a quarterly basis and provide quarterly progress report to management. Presentations about the study will also be delivered when requested, e.g., for the District's Science Plan team, Restoration Strategies workshops, and Long-Term Plan meetings.

An update on the study progress and findings will be prepared in the annual South Florida Environmental Report (SFER). At the conclusion of the project, a final report also will be prepared describing all the studies performed, presenting all the data gathered, and summarizing the findings, conclusions, and recommendations.

3.10 STUDY SCHEDULE

• DSP development	Initiate FY2013	Complete FY2015
• Workshop and peer review	Initiate FY2014	Complete FY2015
• Data collection	Initiate FY2015	Complete FY2018
• Data integration synthesis	Initiate FY2017	Complete FY2019

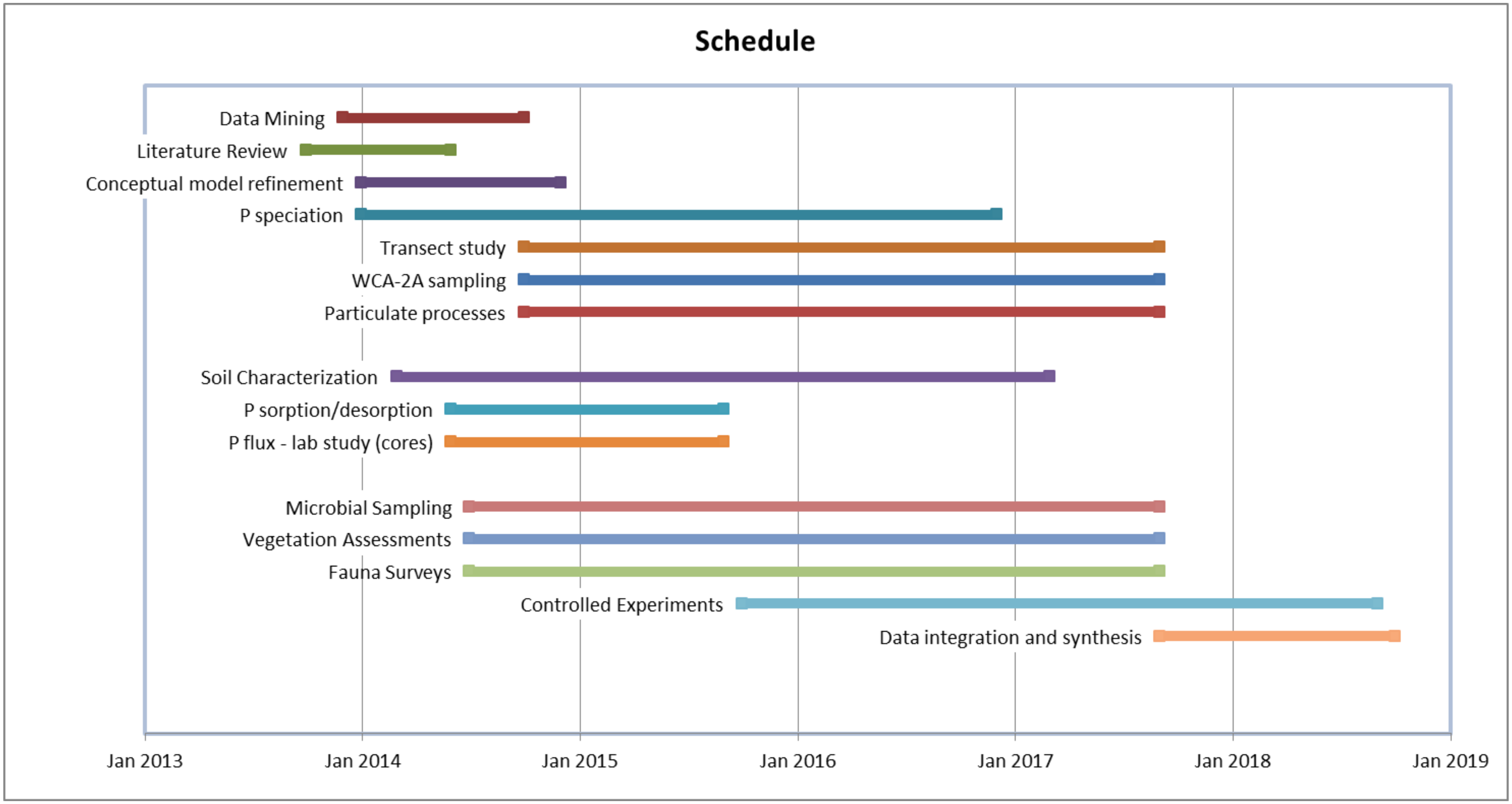


Figure 3-3. Schedule for key project activities.

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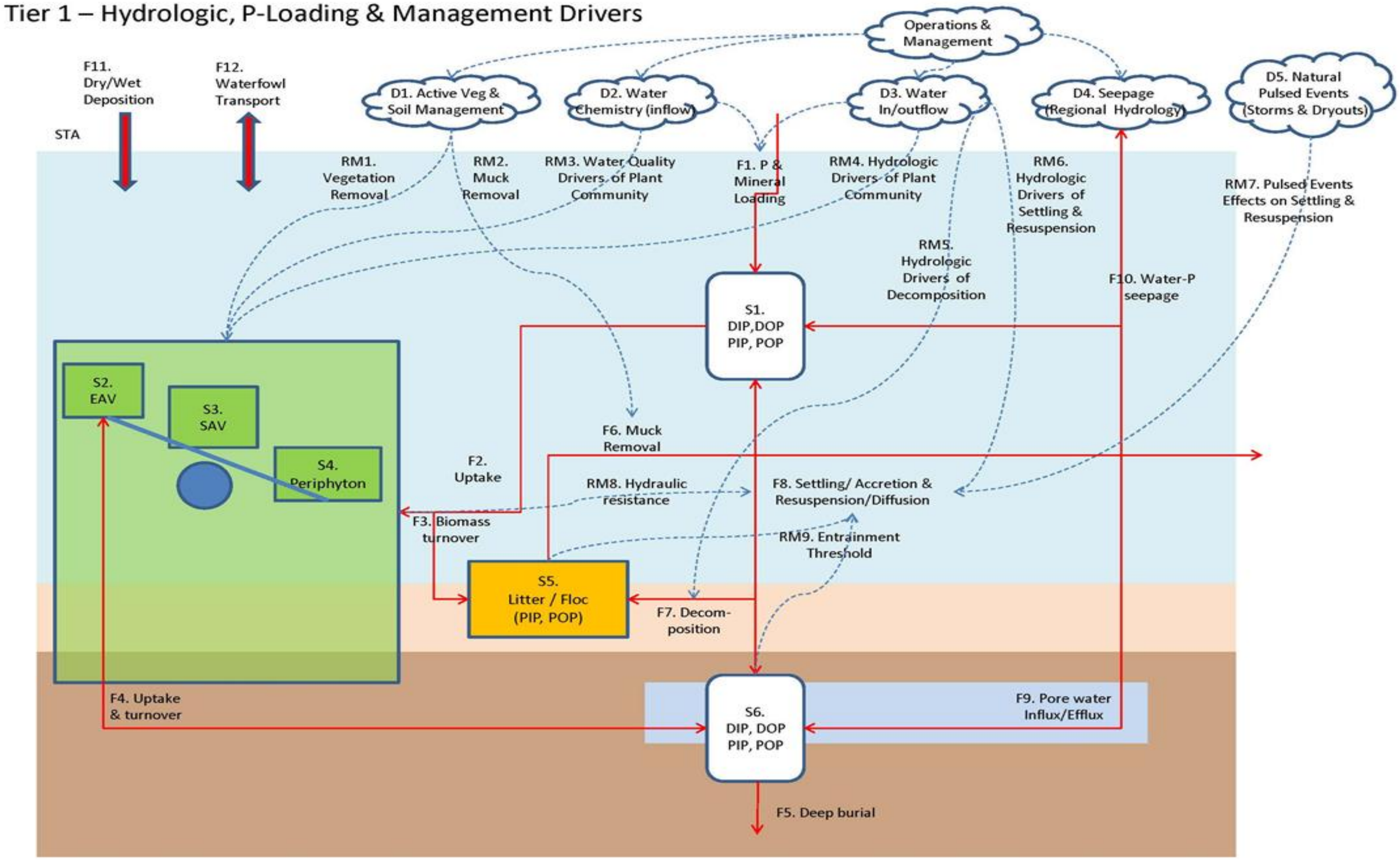
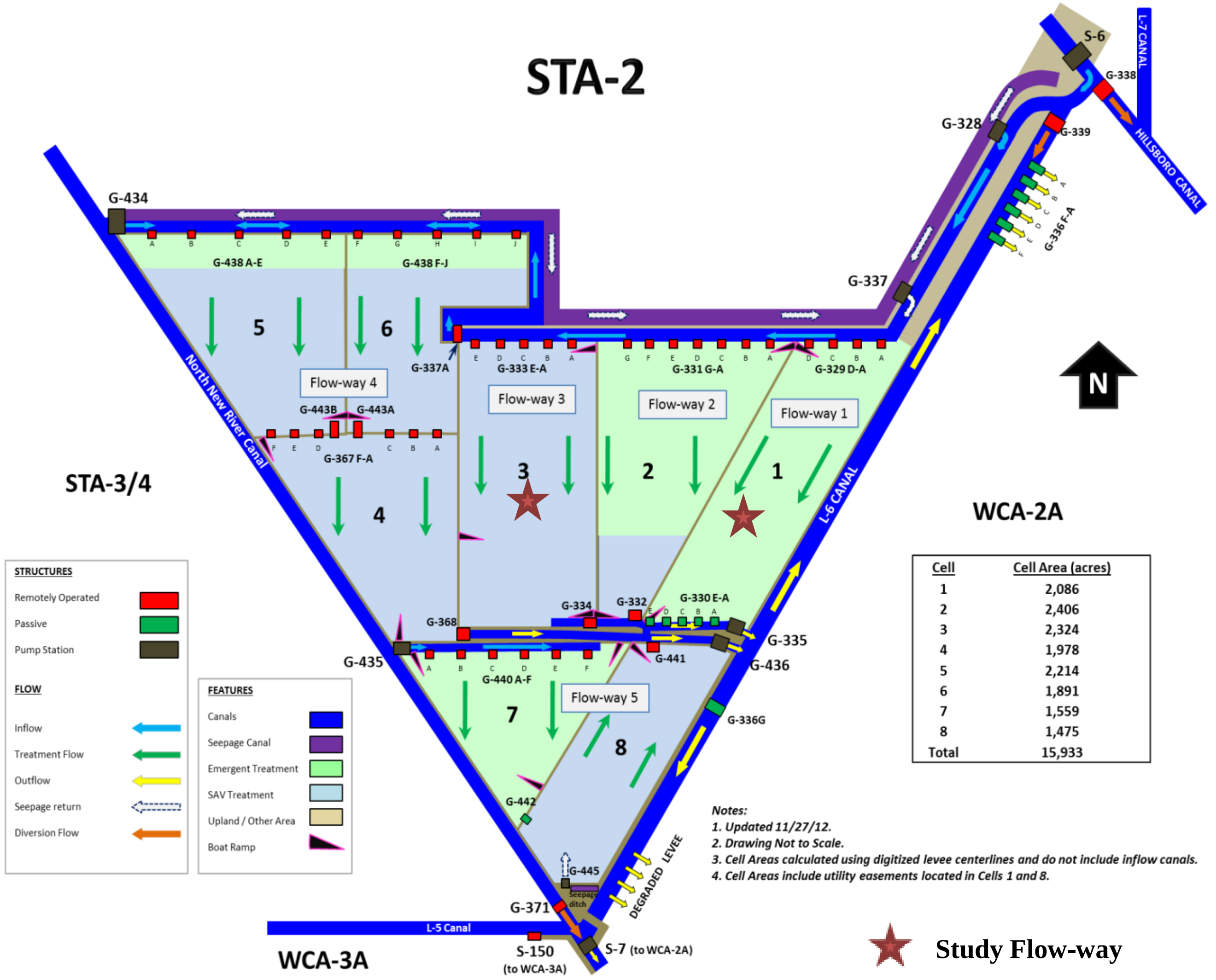
APPENDIX A: DRAFT CONCEPTUAL MODELS FOR PHOSPHORUS CYCLING AND TRANSPORT AT LANDSCAPE, AREA (SAMPLING UNIT) AND SAMPLING UNIT LEVELS

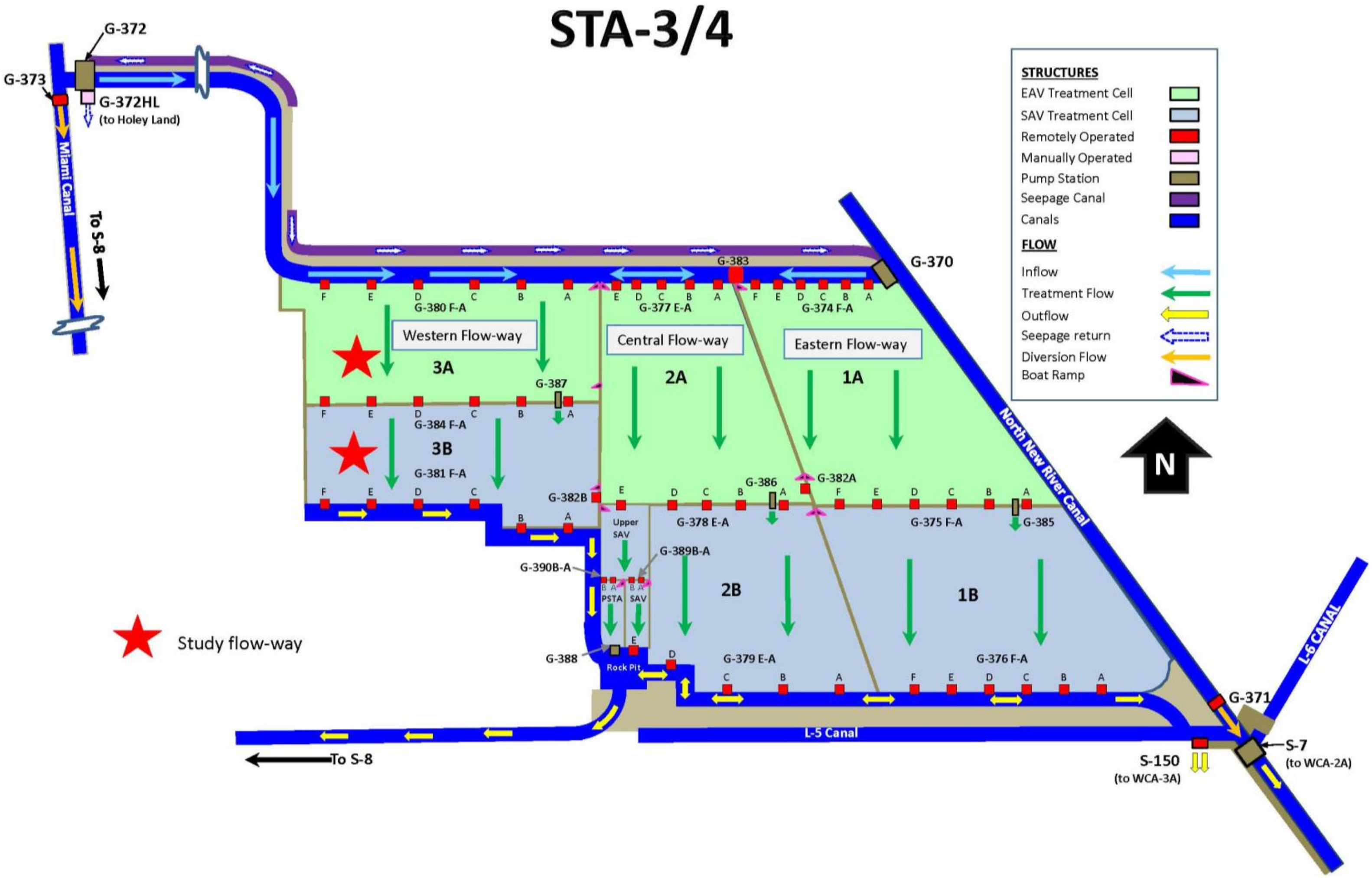

Tier 1 – Hydrologic, P-Loading & Management Drivers

APPENDIX B: SCHEMATICS OF PLANNED STUDY FLOW-WAYS (STA-2 CELLS 1 AND 3, AND STA-3/4



Cell	Cell Area (acres)
1	2,086
2	2,406
3	2,324
4	1,978
5	2,214
6	1,891
7	1,559
8	1,475
Total	15,933

Notes:
1. Updated 11/27/12.
2. Drawing Not to Scale.
3. Cell Areas calculated using digitized levee centerlines and do not include inflow canals.
4. Cell Areas include utility easements located in Cells 1 and 8.


STA-3/4
STRUCTURES
EAV Treatment Cell
SAV Treatment Cell
Remotely Operated
Manually Operated
Pump Station
Seepage Canal
Canals
FLOW
Inflow
Treatment Flow
Outflow
Seepage return
Diversion Flow
Boat Ramp
N
G-372
G-373
G-372HL
(to Holey Land)
Miami Canal
To S-8
G-370
G-383
G-380 F-A
G-377 E-A
G-374 F-A
Western Flow-way
Central Flow-way
Eastern Flow-way
3A
2A
1A
G-387
G-384 F-A
3B
G-381 F-A
G-382B
G-386
G-382A
G-378 E-A
G-375 F-A
G-385
North New River Canal
Upper SAV
G-389B-A
G-390B-A
PSTA
SAV
2B
1B
G-388
Rock Pit
G-379 E-A
G-376 F-A
L-6 CANAL
G-371
L-5 Canal
S-7
(to WCA-2A)
S-150
(to WCA-3A)
To S-8
Study flow-way

APPENDIX C: PHOSPHORUS SPECIATION

Specific Objectives: Identify and quantify the different forms of phosphorus that comprise the organic phosphorus pool in water, floc, and soils using direct and detailed analytical methods. Develop an improved sampling approach for organic P in surface water samples that increases sensitivity and accuracy of all P form evaluations. This will allow a more accurate assessment of which forms of organic P may be limiting the ability of the low performing STAs to reduce TP concentrations to those observed in high performing STAs and in the natural system. This goal will be accomplished by a combination of field sampling and laboratory analyses.

Hypotheses Linkage: I-1 and I-2

Methods: Samples will be collected by the District and sent to a contract laboratory specializing in detailed P speciation. The laboratory will:

1. Optimize ^{31}P nuclear magnetic resonance (NMR) spectroscopy methods for the identification and quantification of organic P forms in floc and soils from high and low performing STAs;
2. Evaluate and optimize various sampling strategies for surface water samples necessary to improve P speciation ability, including utilization or development of extraction and concentration techniques that are specific for organic P;
3. Develop mass spectroscopy (MS) methods for distinguishing organic P forms in the water column from high and low performing STAs, focusing on selective ionization of organic P;
4. Assess sources and transformations of organic P using complementary carbon data, such as stable isotopes and ^{13}C NMR spectra; and
5. Compare the forms and concentrations of the different forms of organic P within the STAs to those measured along the nutrient gradient in WCA2A to assess whether STAs have maximized the degradation/mineralization of organic P forms.

Samples that will be used for testing will be collected at monitoring stations that are part of the Transect sampling sites and will represent organic and mineral conditions, enriched and unenriched conditions and EAV versus SAV conditions. This will provide all potential possibilities for differentiating P composition and any issues associated with differences in biogeochemistry.

APPENDIX D: FLOW-WAY ASSESSMENTS AT DIFFERENT FLOW CONDITIONS (INFLOW TO OUTFLOW TRANSECT STUDY)

Specific Objectives: The objective of this assessment is to evaluate the biogeochemical response of the different regions along selected STA flow-ways to three different flow scenarios; stagnant, low flow, and high flow events. Previous data analyses and experience in operating the STAs have indicated that controlling inflow concentrations and load help achieve concentrations at or around 20 ppb; however, recent data analyses have shown that at moderate inflow concentrations and loading, the STA's ability to achieve concentrations closer to the 13 ppb QBEL may be influenced by other factors. This study is anticipated to determine what those factors are and the relative magnitude of influence of each of those factors, particularly those related to P sources, P flux, and P species transformations.

Hypothesis Linkage: I-1, I-2, I-3, I-4, II-1, II-2, III-1, and IV-1

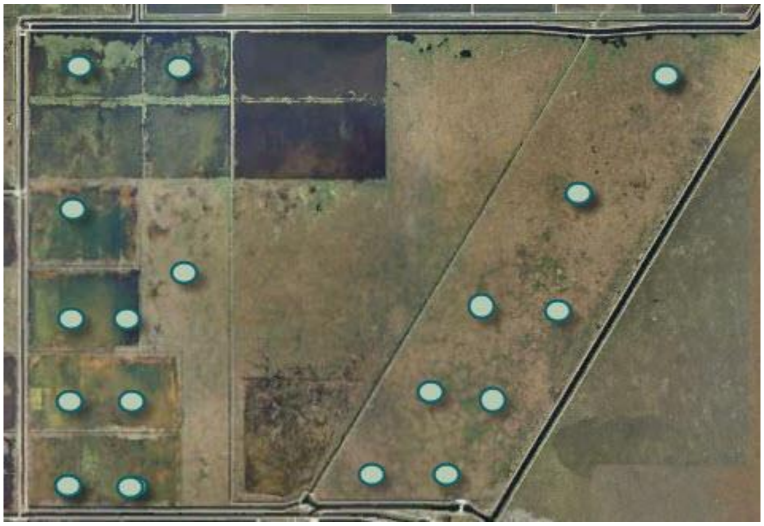
Methods: The selected flow-ways are STA-2 Cell 1, STA-2 Cell 3, STA-3/4 Western flow-way (Cells 3A and 3B) (**Appendix B** and **Table D-1**). STA-2 and STA-3/4 were selected because the inflow concentrations and loading rates are comparable at moderate levels. The specific flow-ways in these two STAs were selected to represent flow-ways that have achieved 13 ppb or better (good performing flow-way) versus areas that achieve between above 13 and less than 30 ppb (less effective flow-way), different cell configurations, different vegetation communities, and different soil condition (**Table D-1**).

In addition, the STA-3/4 PSTA, which currently collects similar information under a separate project, will be considered in the overall evaluation (see PSTA Detailed Study Plan). A schematic of study transect site concept is presented in **Figure D-1**. The sites will be selected along the inflow to outflow gradient using professional judgment on the actual location, based on the results of data mining and analysis, and representation of the flow-way region (i.e., avoiding non-representative condition). Although the focus of investigation is the lower reaches of the treatment train, understanding the biogeochemical conditions in the front end region of the flow-way is critical in identifying the source and potential management strategies that could help reduce the water column P in the lower reaches of the treatment train.

To maximize the data collection and minimize disruption of the site being characterized, the project team will take advantage of equipment that can be deployed remotely, including remote P analyzer at the inflow and outflow locations, autosampler (for collecting TN and TP samples), multi-parameter field meters (for measuring pH, DO, Special Conductivity, temperature, dissolved organic matter (DOM), and turbidity), water level loggers, and light meters. These equipment and meters allow for data collection at short time steps (e.g., every three hours). Samples will be collected before, during, and after an imposed flow regime (stagnant, low flow, and high flow). Grab samples will also be collected once before, during, and after the imposed flow/no flow event and submitted for other analytes (e.g., TP, TKN, SRP, Ca, Mg, NH4, NOx, DOC, Fe, sulfate, chloride, alkalinity, color, and chlorophyll). A close coordination with the control room will be necessary to coordinate resources and deployment of the equipment and instrumentation and allow for measurements of the different parameters at least two weeks prior to the flow/no flow treatment, during flow event, and two weeks after flow/no flow event.

A more detailed examination of the nature and composition of organic P is being done separately using NMR/MS analytical technology (**Appendix C**). At a lesser number of sites along the same flow-way transects, other samples will also be collected for specific purpose: (1) particulate (floc) transport, settling, and resuspension (**Appendix F**); (2) soil porewater to allow for estimation of P flux from the soil column to the water column (**Appendix J**); (3) surface water, floc, periphyton, and soil for microbial activity assays (**Appendix K**); (4) periphyton for biomass and nutrient composition (**Appendix K**); (5) vegetation cover, density, and biomass (**Appendix L**); and (6) water velocity measurements (**Appendix G**). Data for stage and flows into and out of the flow-ways will be obtained from the existing routine STA monitoring programs. Data for PSTA evaluation will be obtained from the existing PSTA research project.

STA-2



STA-3/4 Western Flow-way

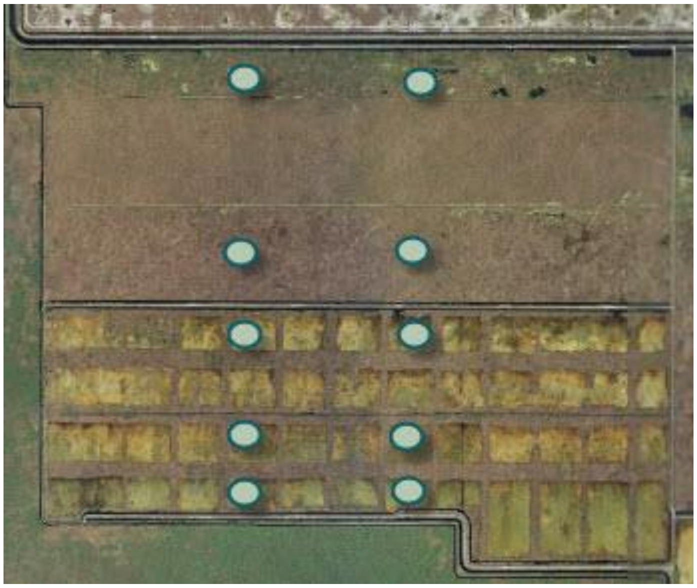


Figure D-1. Planned transect study for STA-2 (top) and STA-3/4 (bottom) flow-ways. Automated equipment and instruments will be deployed at these sites to capture information before, during, and after the imposition of target flow/no-flow conditions. Sampling for particulate dynamics, microbial, flux, and vegetation will be conducted at a lesser number of sites along these transects. The actual location of sites will be determined upon completion of data mining and initial site surveys.

Table D-1. Selected flow-ways for evaluating P forms, flux, and transformations in the STAs. The number of transect sites pertains to locations that will be equipped with automated sampler and field meters. Approximate number of sites for other measurements, such as sediment traps, porewater (flux), and microbial, will be 50 percent of the total.

Location	Treatment Area (acres)	Period of Record Outflow TP Conc.* (ppb)	Configuration	Estimated FW Length (miles)	Vegetation	Soil Characteristics	# Transect Sites	
							EAV	SAV
STA-2 Cell 1	2,086	15	single-cell flow-way; pre-existing wetland prior to becoming an STA	3.3	EAV – mixed cattail and sawgrass	Highly organic pre-STA soil and post STA accreted soil; Floc TP=1172 mg/kg; 0-10 cm soil TP=410 mg/kg	8	N/A
STA-2 Cell 3	2,324	17	single cell flow-way	2.8	SAV, with a patch of EAV cell on the western region	Highly organic pre-STA soil; post STA soil has inorganic floc is deposited over the organic soil layer; Floc TP=827 mg/kg; 0-10 cm soil TP=537 mg/kg	2	8
STA-3/4 Western FW (Cells 3A and 3B)	4,558	14	EAV-SAV cells in series	2.2	Cell 3A – dense cattail Cell 3B - ~60% SAV and ~40% EAV	Shallow (~1 ft of highly organic soil depth); Cell 3A floc TP=1550 mg/kg and 0-10 cm soil=813 mg/kg; Cell 3B floc TP=1003 mg/kg and 01- cm soil TP=645 mg/kg	2	8
STA-3/4 PSTA cell**	100	10.6	100-acre cell where muck had been removed	0.7	SAV with some EAV strips	No muck; some accreted mineral soil with low P concentration	See separate PSTA plan	

*Annual flow-weighted mean concentrations

**STA-3/4 PSTA is included in a separate Science Plan study; data will be included during data integration and data analysis.

APPENDIX E: WCA-2 SAMPLING

Objectives: This objective of this study is to examine existing sites within the WCA2A with known variation in P enrichment to provide additional data that will allow the evaluation of which forms of P and what internal processes may be limiting the ability of the low performing STAs to reduce TP concentrations. The ability of STAs to retain P is based on their capacity to accumulate P in the soils. To what extent this can be accomplished is a function of the lability and recalcitrance of the P forms and how readily they can be cycled internally. It has also been suggested that STA performance decreases as STAs age. The NE section of WCA-2A has experienced 35+ years of P enrichment which has produced a P enrichment gradient extending over 7 km into the marsh, beyond which the system is P limited and surface water TP concentrations are consistently < 10 ug/L. Quantification of the processes, mechanisms, and factors relevant to P uptake along the gradient in WCA2A and the STAs will provide important comparisons on the transformability, or potential recalcitrance of P these systems that may help improve STA performance.

The expected outcomes are:

- Quantification and comparison of short- and long-term lability and recalcitrance P species in water, floc, and soils between WCA2A sites of different nutrient condition with sites along the inflow to outflow gradient of STA flow-ways.
- Comparison of P storage capability of SAV and EAV between STAs and CHIP, where plots receive the same high P inputs.
- Quantification and comparison of P flux and transport between SAV and EAV areas developed and maintained under the same P loading conditions.
- Quantification of rate limiting steps for microbial P cycling as a function of P gradients.

Methods: Five sampling sites were selected from existing WCA-2A long-term transect sites (**Figure E-1**). U3 and E5 were selected as background sites because these sites have very low P concentrations in the water column; U3 represents mineral-enriched soil while E5 represents a more organic soil. The parameters to measure are a limited sub-set of those listed in **Appendix D**; P speciation in water, soil and floc, P flux, and microbial activity. Collection procedures will also be the same as those for STA flow-way transects. In addition, direct comparisons between SAV and EAV processing of P will be made through complementary analyses conducted within open and control plots of the Cattail Habitat Improvement Project, replicated actively managed areas in WCA-2A, where distinct changes in P pools, similar to those in STAs have been observed (**Figure E-2**).

Hypotheses Linkage: I-1, I-2, I-3, I-4, II-1, II-2, and III-1

Sites: F1, F2, F4, E5, U3, and CHIP sites (EO1-3, EC1-3, TO1-3, TC1-3)

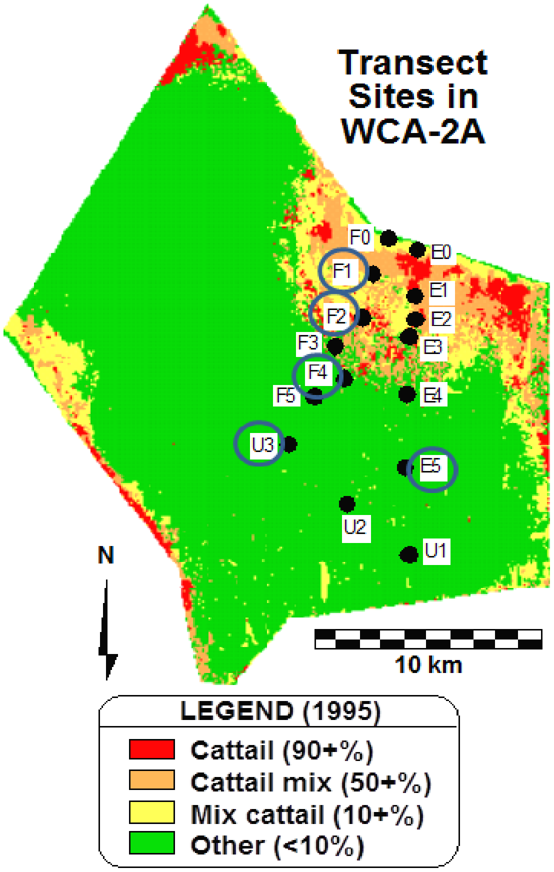


Figure E-1. Location of sampling sites along nutrient gradient in WCA-2A.

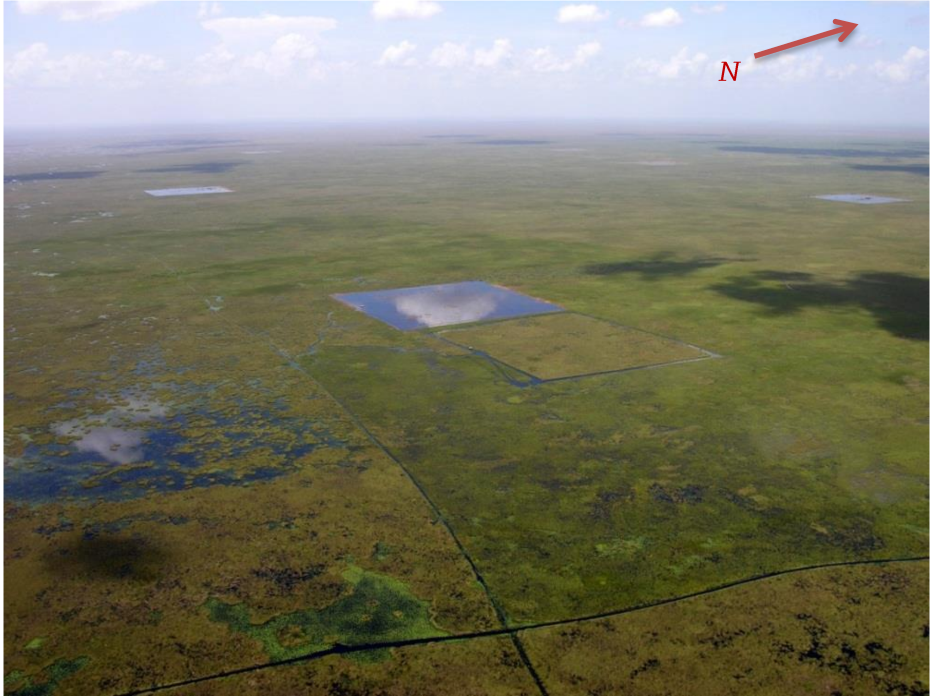


Figure E-2. Aerial view of CHIP paired open and control plots immediately north of cattail/ridge and slough transition.

APPENDIX F: IN-SITU SEDIMENT DYNAMICS

Specific Objectives:

1. To evaluate the effects of specific operational or environmental conditions (e.g., change in flow operation; vegetation community structure; presence of vegetation strips, and presence of deep areas) in altering particulate processes. Particulate P (PP) burial, reductions in PP resuspension, or decreases in water PP and TP will be considered benefits.
2. To identify conditions in which PP resuspension is high, but potentially unmanageable (e.g., wind events) and cost-prohibitive or logistically impractical to fix (e.g., preferential flowpaths caused by large remnant canals).

Hypotheses Linkage: I-1, II-1, II-2, and III-1

Methods: Field measurements will evaluate the extent to which particle settling, resuspension, and transport are driven by hydrologic drivers (flow), ecosystem structure (e.g., vegetation density, community type), particle composition (e.g., density), and disturbances (e.g., bioturbation) over a range of conditions representative of the environmental variation observed in and among STAs. Data mining and STA workshops will be used to prioritize the final studies and in some cases, controlled experiments in test cells or mesocosms may be used to refine these relationships.

Particle Settling/Burial Studies (Hypothesis II-1)

Sampling will be conducted as part of a larger transect study (**Appendix D**) to evaluate settling and burial as a function of vegetation density and patterns, microtopography, particle density, water level reductions, and mechanical burial or covering of sediments. While the exact sampling locations are to be determined, locations will be along the main project transect locations, along the inflow to outflow gradient. Sampling will focus on outflow portions of flow-ways, but must include central and inflow areas to provide context for understanding outflow dynamics. A sampling design will be developed to utilize the spatial and temporal variation in the key independent variables to explain the variation in observed settling rates of PP. The independent variables include some or all the following:

- Vegetation density or pattern (e.g., patterns of SAV and EAV-strips in STA 3/4 Cell 3) (**Appendix L**)
- Topographic variation (e.g., flat areas versus deep areas perpendicular to or parallel to flow directions) (**Appendix G**)
- Water column particulates dominated by mineral-rich versus highly organic particles (**Appendix D**)
- Areas of high and low flow velocities (**Appendix G**)
- Before and after soil amendment (covering) or soil burial (disking) activities

The exact independent variables studied will depend on those prioritized from data mining and STA CEM workshop results.

Particle settling will be quantified using a combination of vertical traps and mass changes in water column and benthic floc particulates (summarized in **Figure F-1**). Vertical traps modified from Kerfoot et al. (2004) have been successfully applied to Everglades deep-water habitats (Sklar and Dreschel, 2014), and additional methods used in Everglades marshes (Leonard et al., 2006) may be applied for evaluating methodological biases (e.g., settling at mid-water column depth versus at the top of the floc layer) and uncertainties. To evaluate changes in particulate standing stocks over time and space, the transect study (**Appendix D**) will measure water particulates, PP and TP concentrations and benthic floc mass and floc-P standing stocks. Vegetation biomass and P stocks will also be provided by transect sampling through destructive harvests, biometric measurements and aerial imagery (**Appendix L**). Hydraulic variables (velocity, direction, depths) will be provided by hydrologic monitoring studies (**Appendix G**) utilizing Acoustic Doppler Velocimeters (ADVs) for small scale, continuous measurements and tracer deployments (e.g., fluorescent dyes or SF6; Ho et al., 2009) for large-scale flow patterns. Lab analysis of particulates collected from sediment traps, water column, floc and vegetation samples will be conducted to determine the composition of mass associated with each attribute, including organic content, and nutrient concentrations. It is possible that additional specialized analyses may be required to fully understand the settling dynamics of particles generated from different sources (e.g., inflow canal-derived particles, SAV- or EAV-derived particles). Available methods include isotopic markers (Stern et al., 2007) or molecular organic markers (Saunders et al., 2006) previously applied in canals, STAs, and Everglades marshes.

Particle Resuspension and Transport Studies (Hypothesis II-2)

These studies will test the degree to which outflow TP in STAs can be reduced by preventing or decreasing resuspension of PP. Resuspension results from two primary mechanisms: (1) particle entrainment when flow velocities exceed a Critical Entrainment Threshold (CET) velocity, and (2) mechanical disturbance such as bioturbation or scouring by moving objects such as floating mats. Here, several factors will be evaluated to determine their impacts on particle resuspension and transport, including:

1. Vegetation die-offs which reduce hydraulic resistance by vegetation and also release particles (**Appendix L**);
2. Floating mats which scour benthic floc, damage vegetation and potentially release their own particulates;
3. High velocities near flow structures (**Appendix G**), especially in areas with high standing stocks of benthic floc or P-enriched floc and soil (**Appendix H**);
4. Preferential flow paths (**Appendix G**) such as remnant canals which often contain P-enriched sediments and remnant sloughs oriented parallel to flow;
5. Extreme wind events;
6. Bioturbation by fauna (**Appendix M**);
7. Extended dry-down periods (**Appendix G**) which increase litter decomposition and fragmentation;
8. Extended periods of stagnant conditions with greater suspended particulates and low SAV and EAV cover (**Appendix L**); and
9. Floc and soils of varying composition of mineral versus organic matter (**Appendix H**) with low critical entrainment threshold (CET) velocities.

Similar to the studies focusing on particulate settling (above), transect sites will be used as a first step to evaluate water column PP concentration and transport as a function of variation in these factors. In some cases, event-based sampling may be required, including sampling after fauna disturbances, wind events, or extended draw-downs. As with settling/burial studies above, sampling will also be conducted as part of the transect study and the final list of studies will depend on those prioritized from data mining and workshop results. Sampling will include selected sites along the selected transects representing inflow to outflow gradient, locations with differing floc composition (mineral versus organic), locations with high and low velocities (e.g., near inflow or outflow areas versus central portions of flow-ways), and areas disturbed by floating mats, wildlife activity, wind, or vegetation die-offs.

Measured dependent variables will include water column PP, particulate transport, and particulate resuspension. Water column PP will be measured as part of the transect study (**Appendix D**) and locations or time periods of very high or low water PP levels may be useful in identifying factors underlying resuspension of PP. However, since entrained particulates can be transported and settled elsewhere in the system, water column PP concentrations alone may not be sufficient to understand entrainment effects. Therefore, sediment transport will be estimated using traps capturing horizontally advected sediment, similar to those currently deployed in Everglades wetlands (DPMST 2010; adapted from Phillips et al., 2000). Alternatively, sediment transport will also be calculated using estimates of flow and water particulate concentrations. Lastly, the critical entrainment threshold (CET) velocity will be measured in sites with contrasting floc and soil composition, as these are the most likely factors to affect particle density and in turn the CET. CET velocities will be measured using benthic annular flume deployments (DPMST, 2010). CET data combined with STA velocity data can indicate periods of increased (or reduced) particle entrainment and may be useful to explain or predict increasing (or decreasing) column particulate concentrations (Sklar and Dreschel, 2014). For evaluating resuspension due to bioturbation, CET measurements will not be made as flow is not the primary driver of resuspension in this case.

Synthesis of Particulate Processes Studies

Sampling for settling and resuspension studies will be co-located to the extent possible and integrated with transect sampling (**Appendix D**) and hydraulic monitoring (**Appendix G**). As a result, a PP mass budget can be constructed to account for particle standing stocks (water, floc, and detritus) and particle fluxes (production, resuspension, and settling) as summarized in **Figure F-1**. While production may not be measured directly here, it can be inferred from vegetation biomass and known biomass turnover rates (e.g., Noe et al., 2002; Daoust and Childers, 1998), and, alternatively, settling rates can be used as a proxy for production, as settling is an integrated measurement of both particle production and settling through the water column. It is intended that these results will be integrated within a flow-way level evaluation synthesizing P flux data from studies I, III, and IV evaluating biogeochemical transformations of P, vegetation pathways, and fauna pathways. This larger synthesis is exemplified conceptually in **Appendix A** and will allow for comparisons of the overall effect of particle processes to other studied pathways (P transformations, vegetation pathways, and faunal pathways) in controlling outflow TP.

Finally, it is expected that particulate responses to abiotic and biotic variables measured in similar Everglades studies, such as the Decomp Physical Model (DPM) in WCA3B or Cattail Habitat Improvement Project (CHIP) in WCA2A, may be used as part of the data mining needed to prioritize the sub-studies listed here. Many of the same habitats, plant species and substrate types observed in STAs are observed in these studies. As a result, findings from studies as DPM may also serve to strengthen conclusions from the STA particulate processes or provide similar synthesis approaches (e.g., PP and P budget models) applied to synthesizing STA particulate data.

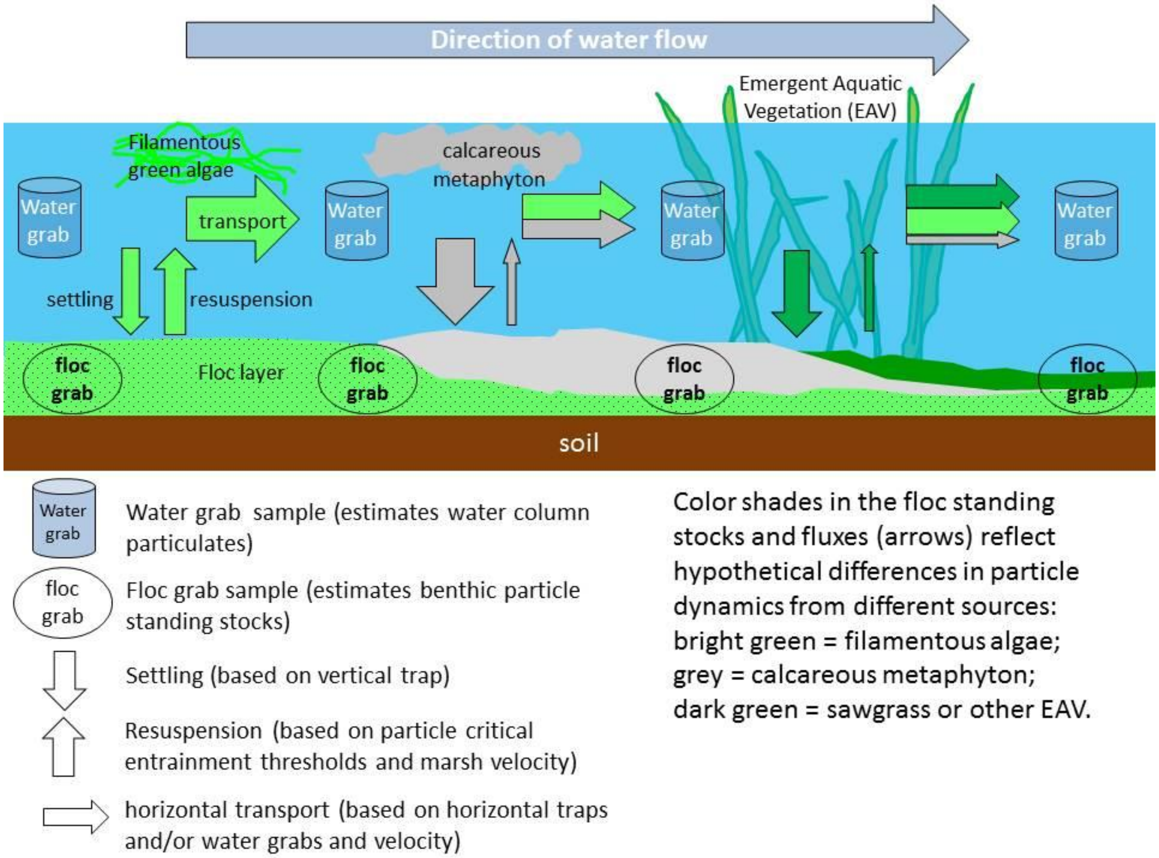


Figure F-1. Conceptual plan for quantifying particulate dynamics using integrated data from particulate sampling, transect studies, vegetation surveys and hydrology monitoring along a conceptual gradient in vegetation or landscape configuration. Settling rates are quantified using vertical sediment traps, and sediment transport rates are quantified using horizontal sediment traps, and secondarily from available hydrologic data and water column particulate concentrations. Resuspension is estimated from floc critical entrainment thresholds (CET, minimum velocity required to entrain or suspend particles) and marsh water velocity data. All data shown in this diagram would be accompanied by TC, TN, and TP analysis, enabling particulate dynamics to be expressed in terms of total mass and PP.

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APPENDIX G: HYDROLOGIC AND HYDRAULIC MEASUREMENTS

Specific Objectives: Hydrologic (stage, water depth) and hydraulic (flow) measurements will be conducted to support the investigations related to P speciation and transformations, particulate dynamics, vegetation pathways, and fauna pathways. Hydrologic and hydraulic measurements will be done in conjunction with the other field tests.

Hypothesis Linkage: I-1, I-3, I-4, II-1, II-2, III-1, and IV-1

Methods: Detailed hydrologic monitoring plans covering the instrumentation, sampling protocols, locations, and duration will be prepared as the specifics of the field tests become available. In some cases, hydrologic monitoring results will be needed to prioritize and generate sampling designs for the field and experimental studies. Anticipated monitoring activities can be divided into three groups based on their spatial extent:

STA/Flow-Path Scale: Data from the surface water flow and stage monitoring at STA structures conducted as part of routine STA monitoring will be utilized for this study. Water budgets that are being developed under a separate Science Plan study will be examined to understand the overall treatment performance of the STAs and flow-ways. Surface water stages and flow rates are monitored at water control structures where flow rates are controlled. Precipitation can be estimated from nearby rain gauges or NEXRAD data. Evapotranspiration will be computed by a model based on lysimeter experiments. Seepage will be estimated using seepage coefficients and head differentials between the cells and adjacent water bodies. For investigations described in Appendices D, F, J, K, and L, the impacts of target flow scenarios (stagnant, low flow, and high flow conditions) will be investigated by controlling the flows at the flow-way inflow structures.

Flow-way Scale: Internal water movement is critical in understanding P cycling and transport. Spatial variability in the hydrogeological parameters influences vegetation pattern and density, which in return may cause significant variations in the local velocities and flow depths. Initial assessment will be done by examining existing aerial images and cell ground elevation data. Visual inspections will also be conducted to verify potential preferential flow paths or flow obstructions. If necessary, tracer studies will be used at the transect scale to provide information about flow velocities and preferential flow-paths and flow pattern through vegetation communities or landscape features, as needed for field studies. Methodologies may include the use of tracers, e.g., rhodamine, fluorescein, or lithium.

*Study Site Scale***:** Water depths and velocities in the vicinity of transect sampling stations will be monitored continuously throughout the field tests. The final number and deployment locations for sampling stations will be determined as these studies become more refined. These measurements will likely entail continuous deployments of pressure transducers and Acoustic Doppler Velocimeters (ADVs) with specifications appropriate for STA wetlands. Finer time and spatial resolutions may also be needed for some hydrometeorological data (e.g., precipitation, ET, wind direction).

APPENDIX H: SOIL CHARACTERIZATION

Objective: Collect and analyze soil samples for the different P species and factors affecting P uptake and release from soil. Results will be used in evaluating effects of different vegetation communities, location with regard to the inflow to outflow gradient, hydraulic and nutrient loading, and inflow concentration. Results will also be used in evaluating the influence of soil characteristics in flux rates and water column P chemistry.

Hypotheses Linkage: I-1, I-2, I-4, II-1, and II-2

Methods: The purpose of this sampling is to obtain baseline soil data for the field studies and address gaps and uncertainties that may be identified during the data mining process. While ample spatial soil data have been collected in the past, the analyses have been limited to basic chemistry and very little information is available for soil processes, including microbial activities and flux potential. Soil samples will be collected within the selected study flow-ways listed in **Table D-1**. Triplicate intact cores will be collected at pre-determined depths (representing floc, STA accrued layer, and pre-STA soil layer to a maximum depth of 30 cm), based on examination of existing data (**Table H-1**). Core section samples will be sent to the laboratory for analysis. Samples are maintained in ice during transport and in refrigerated condition prior to analysis.

As part of the baseline assessment, soil cores will be collected in triplicate from STA-2 Cell 1, STA-2 Cell 2, STA-3/4 Cell 3A, and STA-3/4 Cell 3B. A 4-inch stainless steel corer will be pushed into the soil column and retrieved for sectioning (**Figure H-1**). The standing water column will be slowly removed by pouring, and then soil will be extruded using a PVC extruder**.** Soil section depth, parameters, and target frequency of collection is shown in **Table H-1**.

Table H-1. Soil profile sampling depth, parameters, and frequency.

Section	Test Category	Parameters	Frequency
Floc layer*	Basic chemistry	TN, TP, TC, AFDW, BD, pH, TCa, TFe	Quarterly
	P fractionation	Microbial, Labile, moderately labile, resistant P forms	Quarterly
0-5 cm	Basic chemistry	TN, TP, TC, AFDW, BD, pH, TCa, TFe	Annual
	P fractionation	Microbial, Labile, moderately labile, resistant P forms	Beginning and end of study
5-10 cm	Basic chemistry	TN, TP, TC, AFDW, BD, pH, TCa, TFe	Annual
	P fractionation	Microbial, Labile, moderately labile, resistant P forms	Beginning and end of study
10-20	Basic chemistry	TN, TP, TC, AFDW, BD, pH, TCa, TFe	Annual
	P fractionation	Microbial, Labile, moderately labile, resistant P forms	Beginning and end of study
20-30	Basic chemistry	TN, TP, TC, AFDW, BD, pH, TCa, TFe	Annual
	P fractionation	Microbial, labile, moderately labile,	Beginning and end of study

*See also Floc Dynamics study

Figure H-1. Soil sample corer and extrusion process.

When evaluating the potential role of stored P in soil to the overlying water column P concentration, knowing soil TP concentration is not sufficient. It is important to know the forms of the stored P and determine what portion can be readily released (labile P pool) and the portion that is resistant to breakdown (stable P pool). A detailed soil P fractionation can provide the speciation based on extractability using weak salt, acid, and alkali solutions. This will be complementary to the analysis that is planned using NMR and Mass Spectrophotometric methods (**Appendix C**).

Approximately 50 percent of the samples collected from the soil chemistry core sampling will be used for soil P fractionation**.** Fractionation will be done following the method developed by Ivanoff et al. (1998). The procedure involved sequential chemical extraction with 0.5M $NaHCO_3$ (representing readily labile fraction), 1 M HCl (representing moderately labile inorganic P, e.g., P bound to Ca, Mg, Fe, and Al), and 0.5 M NaOH (representing moderately and highly resistant organic P associated with fulvic and humic fractions, respectively). Phosphorus remaining in the residual sediment after the sequential extraction was measured by the ignition method and is called residual P, non-reactive P that includes both organic and inorganic P.

APPENDIX I: PHOSPHORUS SORPTION AND DESORPTION CHARACTERISTICS

Specific Objectives: This study will evaluate the P sorption and desorption characteristics of the soils in the STAs and the various abiotic factors that influence them. Specifically, this study will: i) compare the P sorption and release properties of floc and soil layer from a good-performing and a poor-performing STA under aerobic and anaerobic conditions, ii) evaluate the relationships among P sorption parameters and selected soil properties, and iii) identify soil variables that best predict P sorption/desorption.

Hypothesis Linkage: I-1, I-2, I-4

Methods: Sites will be selected from the study transects listed in **Appendix D**. The actual number of sites will be decided upon examination of the historical data. Soil samples will be collected at a fixed depth to include at a minimum, the floc and upper 10 cm layers, using intact core sampling method. Phosphorus adsorption isotherms will be determined on fresh soil samples using the batch incubation technique. The method involves mixing a known amount of soil with a solution containing known P concentrations. The mixtures are equilibrated for 24 hours at a constant temperature and under continuous shaking (Nair et al., 1984). Phosphorus not recovered in solution after 24-hour equilibration represents the fraction of P sorbed by the soil. Mathematical descriptions of P sorption in soils will be made by fitting sorption data to Langmuir or Freundlich equations:

Linearized simple Langmuir:

$$C/S = (1/bS_{max}) + (C/S_{max})$$

Where C is the concentration of P remaining in solution after equilibration, mg/L
S is the total amount of P sorbed by the solid phase, mg/kg
b is a constant related to bonding energy, L /mg P
Smax is the adsorption maximum, mg P/kg

The plot of C/S versus C should yield a straight line with intercept and slope equal to 1/bSmax and 1/Smax, respectively.

Linearized Freundlich:

$$\log S = \log K + n\log C$$

Where K is phosphate adsorption coefficient, L/kg
C is the concentration of P remaining in solution after equilibration, mg/L
S is the total amount of P sorbed by the solid phase, mg/kg
n is empirical constant

A plot of logS versus logC provides a straight line with intercept and slope equal to logK and n, respectively.

These equations will generate curve-fitting parameters such as sorption maximum, energy of adsorption and equilibrium P concentration (EPC_o). EPC_o represents the P in soil porewater that is in equilibrium with P in the solid phase, at which there is no net movement of P from soil to water or vice versa (Reddy et al., 1999).

The EPC_o values determined by adsorption isotherms have been used to indicate whether adsorption or desorption occurs in the soils or sediments, when added P alters the concentration of P in the porewater. If the water entering a wetland has P concentration below EPC_o, theoretically, that soil would release P to overlying water. Conversely, if the water entering a wetland has a P concentration greater than the EPC_o, then that soil would retain P. While EPC_o values provide useful information regarding the ability of the soils/sediments to either retain or release P as a function of floodwater P concentrations, further field verification and data collection will be required to account for the combined physicochemical and biological processes that control the amount of P that is in solution at any given time. Under field condition, the soluble fraction is taken up by plants, sequestered in soils/sediment or disperses in the surrounding environment.

In addition, soils will be analyzed for other properties, and chemical composition that influence P sorption and desorption, e.g., organic matter content, pH, and extractable metals (**Appendix H**). The expected outcomes are (1) a summary of P sorption parameters (e.g., sorption maximum, energy of adsorption, equilibrium P concentration (EPC_o), (2) information on the spatial distribution and seasonal variability of measured sorption parameters, and (3) correlation between soil properties such as organic matter, pH, and extractable metals with sorption parameters. These parameters will then be considered during final data synthesis and evaluation to determine the ability of the individual flow-way to achieve the desired outflow concentrations.

APPENDIX J: PHOSPHORUS FLUX

Specific Objectives: The objectives of this study are to (1) measure maximum P flux rates from enriched and unenriched areas of the STA, with and without vegetation, under stagnant water conditions using intact soil cores; (2) determine P flux rates under different flow conditions, using porewater equilibrators or piezometers; and (3) identify soil variables that exert influence on P flux. The expected outcome for this investigation include maximum P release rates for enriched, unenriched, vegetated, and unvegetated areas of the STA; phosphorus release rates under stagnant and flow conditions; P speciation in the water column; and relationship between water chemistry (e.g., calcium) and P flux rates.

Hypotheses Linkage: I-1, I-2, and I-4

Methods: The diffusive flux of P from the soil to the overlying water column, with and without vegetation, will be determined using intact soil core incubations (**Figure J-1**). Site water, obtained along the inflow to outflow gradient, with a range of initial water column P concentrations typically seen in the STA will be used to determine the effects of floodwater P concentrations on the ability of selected STA soils to either retain or release P. Floodwater exchanges using STA site water will be performed over a minimum of four 7-day hydraulic retention cycles. Samples will be collected on days 1, 3 and 7 and analyzed for soluble reactive P (SRP), total dissolved P (TDP), total phosphorus (TP), and calcium. In-situ measurements of pH, dissolved oxygen (DO), temperature, and specific conductivity will be taken with a YSI on days 1 and 7 of each of the four incubation periods. The soil cores will be equipped with platinum electrodes for redox measurements. At the end of the 7-day cycle, the water column will be removed and replaced with a new 30-cm of STA site water. These steps will be repeated until the last floodwater exchange is completed. The rate at which P is released to the water column will be calculated by determining the slope of the concentration versus time curve through linear regression, then multiplying by the floodwater volume to soil surface area ratio of the soil core (Fisher and Reddy, 2001).

$$Ji = \frac{dC}{dt} x \frac{V}{A}$$

Where Ji = flux of P (mg m^{-2} d^{-1}), C = P concentration in floodwater (mg L^{-1}),

A = soil surface area (m^2), V = volume of floodwater (L), and t = time (d).

The use of benthic chambers will also be evaluated for a limited number of sites to help evaluate the actual flux in field condition (Fisher and Reddy, 2001).

Porewater samples will also be collected along the STA flow-way (**Appendix D**) and WCA transects (**Appendix E**) during the treatment flow regimes described in **Appendix D**. The goal is to collect porewater profiles that are representative of the period of stagnant, low flow and high flow conditions. Porewater equilibrators or peepers will be used to obtain vertical profiles of porewater within the soil and water columns (**Figure J-2**). The peepers will be inserted into the sediment and the chemical constituents in the porewater will diffuse across the cell membrane until equilibrium is achieved. Equilibrium studies have shown that two weeks is sufficient time for dissolved constituents in the sediment porewater to equilibrate with deionized water inside the equilibrator cells (Newman and Pietro, 2001; Fisher and Reddy, 2001). Upon retrieval, samples will be withdrawn with a syringe and analyzed for dissolved constituents. The sediment porewater data and the published diffusion coefficients will be used to estimate the flux of P across the soil-water interface using Fick's First Law, which states that flux is proportional to concentration gradient.

$$J = -\Phi D_s \, (dC/dz) \;\; (8.64 \times 10^5)$$

Where J = diffusive flux in mass per unit area per unit time, mg m^{-2} d^{-1};

Φ = porosity, cm^3 cm^{-3},

D_s = whole-sediment diffusion coefficient in terms of area per unit time, cm^2 s^{-1};

C = dissolved reactive

P concentration, g cm^{-3}; and

z = depth, cm

In-situ measurements of pH, DO, temperature, specific conductivity, and redox will be taken under different flow conditions.

Figure J-1. Intact soil core incubations.

Figure J-2. A photograph of porewater equilibrators.

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Reddy, K.R., R.H. Kadlec, E. Flaig and P.M. Gale. 1999. Phosphorus retention in streams and wetlands: A review. *Critical Reviews in Environmental Science and Technology*, 29:83-146.

APPENDIX K: MICROBIAL ACTIVITY

Specific Objectives: Evaluate the patterns and trends in microbial activities and relate those with the P species distribution and P cycling along a phosphorus gradient within the different STA flow-ways. Under mineralized P limiting conditions, the phosphorus associated with dissolved organic phosphorus (DOP) can be released through organic matter breakdown catalyzed by monoesterase and diesterase enzymes, which are found within living cells as well externally-released enzymes (exoenzymes). When SRP is present, the production of monoesterase and diesterase enzymes can be repressed. Thus, the activity of these enzymes can serve as an indicator of available phosphorus. Specifically, we will be conducting monoesterase and diesterase enzyme assays for the water column, periphyton, floc, and soil (0 – 5 cm fraction).

Hypothesis Linkage: I-1 and I-3

Methods: The study will be conducted in two phases. During the first phase, the water column, periphyton, floc, and soil samples (components) will be collected from selected flow-ways (**Appendix D**) to gain an understanding of the spatial distribution of enzyme activities and to refine the methodology used for the microbial collection and assay procedures for monoesterase and diesterase enzyme activity. Periphyton biomass will be collected using either acrylic dowels or periphytometers (**Figure K-1**). At the time of sample collection, water column temperature, dissolved oxygen, TP, SRP, DOC, and light readings through the water column will be measured (**Appendix D**). Water column and periphyton productivity will be estimated in situ using the light/dark bottle technique. The data collected in this phase will provide the baseline information for the microbial assays and periphyton sampling. In the second phase, the same components will be sampled and analyzed for microbial enzyme assay analysis at the sites selected for the transect study. Because the transect monitoring will be conducted to quantify P flux at stagnant, low flow, and high flow events, the components will be sampled immediately before the events, during the event, and 1 week following the flow/no flow event. This microbial sampling will be in concurrence with the measurements and sampling discussed in **Appendix D**. Specifically, the water column temperature, dissolved oxygen, and light penetration through the water column will have to be measured at the time of sample collection and samples will have to be collected for TP, SRP, and DOC analysis. Water column and periphyton primary productivity will be measured in situ using light/dark bottles. Additional data collected during the flow regime transect measurements and vegetation surveys (**Appendix L**) will be correlated with enzyme activity results. The activity of monoesterase and diesterase enzymes will be determined fluorometrically using 4-methylumbelliferyl phosphate as the substrate and measured at various intervals over an hour to estimate the activity potential and rate. The enzyme activity for the periphyton will be normalized by surface area, biomass, and primary productivity while the other matrices will be normalized by weight.

Figure K-1. Dowel (left) and periphytometer (right) deployment.

APPENDIX L: VEGETATION ASSESSMENTS

Specific Objectives: Survey and/or sample plant communities and relate those to water column P concentration, floc and soil P storage and stability, soil characteristics, soil accretion, and UV radiation/light penetration.

Hypotheses Linkage: I-1, I-2, I-3, I-4, II-1, III-1, and IV-1

Methods: This project will build on existing information from the data mining contract combined with new data and techniques to test the vegetation related hypotheses listed above. A detailed spatial component will be included by the use of high-resolution aerial photography and linked with field data to provide a more complete data set (temporal and spatial) for analyses.

Remote Sensing: Photo transects of the flow-ways will be flown with either an unmanned aerial vehicle (UAV) or the UAV equipment mounted on the District Bell 407 helicopter. These photos will be mosaicked together (AgiSoft Photoscan, AgiSoft LLC, St. Petersburg, Russia) to provide landscape coverage of the flow-way at 1.7 cm resolution. We know from recent test studies, density and identification can be determined at a species level. A map of the flow-way will be created (Feature Analyst, Overwatch Systems, Ltd, Austin, Texas) at sufficient detail (i.e., number of vegetation communities) to answer the hypotheses. Random photo samples will be taken outside the study flow-way to determine vegetation community distribution across the entire STA and provide information for STA-wide analyses. This data will also be used to answer related hypotheses such as faunal use (see **Appendix M**). This assessment will also utilize the aerial imageries acquired annually through the routine STA aerial flights. These images, acquired at 13,500 feet ± 300 feet above the mean terrain, will be analyzed for EAV coverage and density, to obtain estimates of the EAV's role in P cycling.

Emergent Aquatic Vegetation: Vegetation surveys will be conducted according to the frequency specified in **Table L-1**. These surveys will be conducted at strategic locations within each study flow-way. Percent cover for each species within a 1 m^2 quadrat will be recorded. The quadrat will also be photographed at 1 m above the vegetation using a digital SLR camera (**Figure L-1**). Percent cover from photos will be recorded by two observers and compared to field data. If these methods do not differ after the first two sample events, photographs will continue to be collected for future comparisons, but cover/species estimates from photographs will not be recorded. Above ground vegetation materials will be harvested within a 1 m^2 quadrat and analyzed for biomass, TC, TN, TP, and Ash content. EAV vegetation communities, community fitness/configuration, biomass, and nutrient composition will be analyzed as a function of hydrology, hydrologic history, soils, soil history, light penetration, phosphatase activity, and epiphyte growth along the flow way using multivariate statistics.

Submerged Aquatic Vegetation: Quantitative vegetation surveys will be conducted four times per year in the selected flow-way along an inflow to outflow transect. The timing of the surveys will bracket the stagnant condition and target flow regimes for the transect study. These surveys will determine the relative density and coverage of dominant SAV species, using two methods: a rake method approach in which a rake is used to retrieve representative plant communities and the sampler identifies the species and its relative density (**Figure L-2**) and a percent cover approach in which percent cover of species are estimated in 4 replicate samples of a 1 m^2 area. The combination of these two methods provides temporal overlap with existing data (rake method) and a more quantitative measure of vegetation coverage/density for future analysis and remote sensing field data. Photo sampling (as above) will be attempted with SAV communities but may not be possible due to reflection off the water's surface. Spatial maps of the SAV species distribution will be constructed using Arc GISv9 Spatial Analyst (Environmental Systems Research Institute, Redlands, CA).

SAV vegetation communities, community fitness/configuration, biomass, and nutrient composition will be analyzed as a function of water column P, hydrology, hydrologic history, soil characteristics, light penetration, and epiphyte growth along the flow way using multivariate statistics. On a coarser spatial scale, vegetation samples will be collected along the inflow-to-outflow transect to assess standing crop biomass, and tissue TN, TP, TC, and TCa concentrations. Sampling locations are shown in **Appendix B**. Sampling will be done four times during a period that brackets the flow regime treatments (stagnant, low flow, and high flow events). Tissue P storage results can then be integrated with P storage in floc, soil, and water column to assess the internal nutrient budget and determine how this is affected by flow condition, vegetation communities, and relative location along the inflow to outflow gradient.

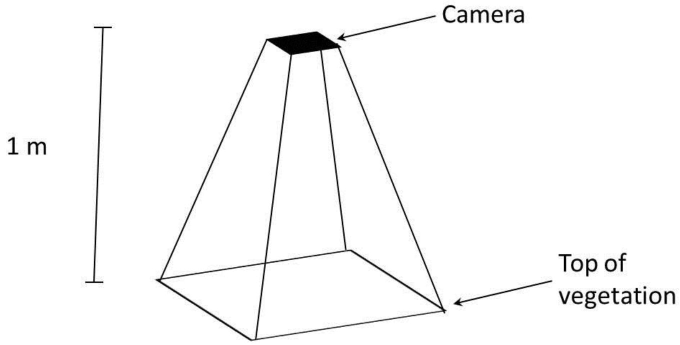

Figure L-1. Camera set up for ground photo samples.

Figure L-2. Rake method of identifying species present at specific sites and their relative density, e.g., 0=absent, 1=sparse, 2=dense, 3=very dense.

Table L-1. Method description and frequency of survey/sampling to assess vegetation condition in study flow-ways.

Parameters	Method	Frequency
Emergent vegetation coverage (entire flow-way)*	Aerial imagery remote sensing at 13,500± 300 feet above the mean terrain	Annual
Emergent vegetation coverage/density (transects)	Low-altitude (~300-500 ft above the mean terrain) aerial remote sensing	Quarterly
Emergent vegetation biomass (above ground)	Quadrat sampling	Semi-annual
Emergent vegetation tissue nutrient composition (above ground)	Quadrat sampling	Semi-annual
SAV relative density	Rake method, species relative density (0=no vegetation, 1=sparse, 2=moderate, 3=dense	Quarterly
SAV biomass	Quadrat sampling	Quarterly
SAV tissue nutrient composition	Quadrat sampling	Quarterly

*This aerial imagery for the all the STAs is conducted annually under routine monitoring program.

APPENDIX M

QUANTIFYING FAUNAL ASSEMBLAGES AND EXCRETION

Specific Objectives: The direct and indirect effects of animals on nutrient cycles in the STAs are unknown. This project will quantify animal communities and their effects on water quality and vegetation by surveying the density, biomass and composition of aquatic fauna, i.e., fishes and large macroinvertebrates, and birds in the outflow cells, and determine the mass-specific P excretion rates of common aquatic fauna.

Hypotheses linkage: II-2**,** IV-1

Methods:

Aquatic Faunal Survey*:* Extensive sampling will occur in two selected SAV cells. The exact study locations are yet to be finalized but will include areas with different types of habitats and P levels. The first step in the sampling process will be to develop a sampling scheme, e.g., a stratified or random design that suitably addresses spatial heterogeneity in fish and invertebrate biomass related to variation in macrophyte habitat (e.g., Chick & McIvor, 1994). The data will be examined for significant variation by SAV habitat using univariate or multivariate statistics, and the results will be used to develop the sampling design (number and location of sampling sites).

Two techniques will be used to quantify areal biomass. Throw traps (1 m^2) will provide estimates of density, biomass and composition of smaller fishes and macroinvertebrates that are generally less than 4 cm total length (Dorn et al. 2005). All collected animals will be euthanized with MS222 and formalin and then transported to and processed in a suitable lab. Throw traps are inappropriate for sampling larger fish so a block-net sampling method (Chick et al., 1999) will be used for fish larger than 4 cm. Block-net sampling is a removal technique that uses Rotenone in an enclosed area to quantify fish per unit area. Fish euthanized with this procedure will be collected, identified, measured and buried on site.

Aquatic Faunal Excretion Rates*:* Nutrient excretion rates will be measured directly using short-term incubations of recently captured individuals (Schaus et al., 1997) of the most abundant species. Water samples will be collected in the field, filtered, and used to calculate dissolved P before and after the addition of a given individual for 1-2 hours. These will be compared with controls that contain no animals. The exact methodology is yet to be developed.

Waterbird Survey*:* Birds influence water column P through excretion (recycling) and the translocation (import or export) of nutrients at relative large spatial scales (within or among wetlands). In order to understand their role in nutrient cycling in the STAs, it is necessary to get quantitative estimates of species abundance, as well as the forms and availability of P in their excreta. Due to the potential for extreme variability in spatial and temporal abundance of birds, the survey methodology will be determined after an initial pilot study. A likely candidate approach is double-observer, fixed interval point counts (Reynolds et al. 1980, Nichols et al. 2000) combined with aerial photography (**Appendix L**). Nutrient excretion rates of birds will be obtained from published data.

Calculating Areal P Excretion Estimates*:* Areal biomasses of species from the throw trapping, block net sampling and avian surveys will be combined with the species-specific and mass-specific excretion rates to generate estimates of total community excretion estimates for multiple locations inside each of the STA cells. These estimates, in $\mu g \text{ P} \bullet ha^{-1} \bullet h^{-1}$, will be compared with concentrations in the water as well as uptake across the STA cells (input and output values) to consider the magnitude of the source of P internal recycling compared with external sources and net uptake rates. The P excretion rates calculated here can be used to parameterize P-budgets for the STAs.

APPENDIX N: CONTROLLED STUDIES

More detailed scope, including design and methodologies for the controlled studies will be developed in the future after the data mining, literature review, and initial studies have been completed. It is anticipated that a smaller subset of factors and mechanisms will emerge as potential key influencers to P reduction processes, information gaps, and uncertainties related to these factors and mechanisms will be identified. Isolating certain variables and studying these in a more controlled environment may provide results to establish cause and effect relationship between key variables and water column P.

Some initial concepts for controlled studies are as follows:

1. Use of in-situ benthic chambers to measure P flux under field conditions (**Appendix J**).
2. To isolate the variables influencing microbial P release, there may be a need to control other variables, e.g., supply of SRP. Conducting this study in the field (**Appendix K**), where P loading varies and flux of SRP from the soil occurs, will not be sufficient to fully address questions related to the role of microbial activities in P cycling. This study may be conducted using microcosms (cores or cylinders).
3. A mesocosm study using the elongated tanks at the STA-1E Flying Cow facility is also planned to complement the in-situ sediment dynamics study (**Appendix F**). The tentative experimental design concepts are as follows:
    a. Different flow velocities (using different flow-through rates) – below and above the CET
    b. Controlled vegetation and substrate conditions; i.e., through planting & transplanting soil monoliths)
    c. Controlled floc sources, i.e., transplanting floc from specific plant communities or cells within the STAs
    d. Altered water quality – TP, alkalinity, $CaCO_3$

The research plan will be updated in early FY2015 to include more information on the controlled studies.

SECTION 4:
INVESTIGATION OF STA-3/4 PSTA PERFORMANCE, DESIGN AND OPERATIONAL FACTORS

4.1 OVERALL STUDY PLAN SUMMARY

Implementation of the STA-3/4 Periphyton-based Stormwater Treatment Area (PSTA) Project has been underway since WY2008. Beginning in FY2012, additional research and evaluation efforts were initiated for the PSTA Project and these efforts are continuing as part of the Restoration Strategies Science Plan.

The purpose of this study is to assess the chemical and biological characteristics and the design and operational factors of the PSTA Cell in STA-3/4 that contribute to the superior performance of this technology. Key factors thought to enable the PSTA Cell to achieve ultra-low outflow total phosphorus (TP) levels will be examined through experiments conducted at a variety of scales, including laboratory bench-scale studies, soil core studies, flow-through mesocosms, and field-scale investigations, with the goal of informing design decisions for successful full-scale replication of this technology. The operational ranges under which the PSTA Cell achieved ultra-low outflow TP levels will be determined, and the management practices required to sustain good performance in the PSTA Cell will be identified.

4.2 BASIS FOR THE PROJECT

Key Science Plan Question Study Addresses

- Key Question 2: How can internal loading of phosphorus to the water column be reduced or controlled, especially in the lower reaches of the treatment trains?
- Key Question 4: How can the biogeochemical or physical mechanisms be managed to further reduce soluble reactive, particulate, and dissolved organic phosphorus concentrations at the outflow of the STAs?

Science Plan Sub-Question Study Addresses

- What are the treatment efficacy, long-term stability, and potential impact of floc and soil management?
- What are the key physical-chemical factors influencing P cycling at very low concentrations?
- Are there things that can be done in the STAs to enhance settling, filtering, and treatment of DOP and PP in the water column?
- What are the sources (internal/external, plants, microbial, wildlife), forms, and transformation mechanisms controlling the residual P pools within the different STAs and are they comparable to what is observed in the natural system? [Note: This sub-question will be integrated with work being performed under the P flux study; see Section 3 of this document.]

It should be noted that while selected components of this research project address the above Science Plan questions, this effort primarily is designed to elucidate critical factors that will guide evaluation of the feasibility of full-scale implementation of the PSTA technology. The questions and hypotheses related to these factors are provided in Section 4.3, *Study Plan Objectives*.

4.3 BACKGROUND/LITERATURE REVIEW

The concept of using periphyton to cleanse stormwater prior to entering the Everglades has been investigated by South Florida Water Management District (SFWMD) scientists and other researchers for over 20 years. Periphyton communities are complex assemblages of cyanobacteria, eubacteria, diatoms, and eukaryotic algae and are found in lakes, streams, and wetlands, including the marshes of the Everglades (McCormick and O'Dell 1996). Several characteristics of periphyton communities make them well suited for biological treatment of surface waters in wetlands. Periphyton growth is associated with surfaces (i.e., on macrophytes or the sediment surface). Periphyton typically has a high affinity for P and responds to P inputs more rapidly than other wetland components (macrophytes, soils) and thus is important in the uptake and storage of P (McCormick et al., 1996). The presence of highly productive periphyton communities in P-limited systems has been linked to the increased uptake efficiency and rapid recycling of nutrients, due to the close association of autotrophic and heterotrophic microbial components (Wetzel, 1996). Floating and benthic calcareous periphyton mats are a key component of the ultra-oligotrophic Everglades marshes (Browder et al., 1994), and are thought to be capable of reducing water column TP to extremely low levels.

A field-scale periphyton-based STA was constructed from 2004 to 2005 for the purpose of addressing uncertainties associated with large-scale implementation of the PSTA treatment technology. The PSTA Project is located in STA-3/4, on a total of 400 acres and is comprised of a 200-acre Upper SAV Cell, a 100-acre Lower SAV Cell and a 100-acre PSTA Cell (**Figure 4-1**). The PSTA Cell is unique among STA treatment cells in that the extant peat was scraped to expose the underlying rock.

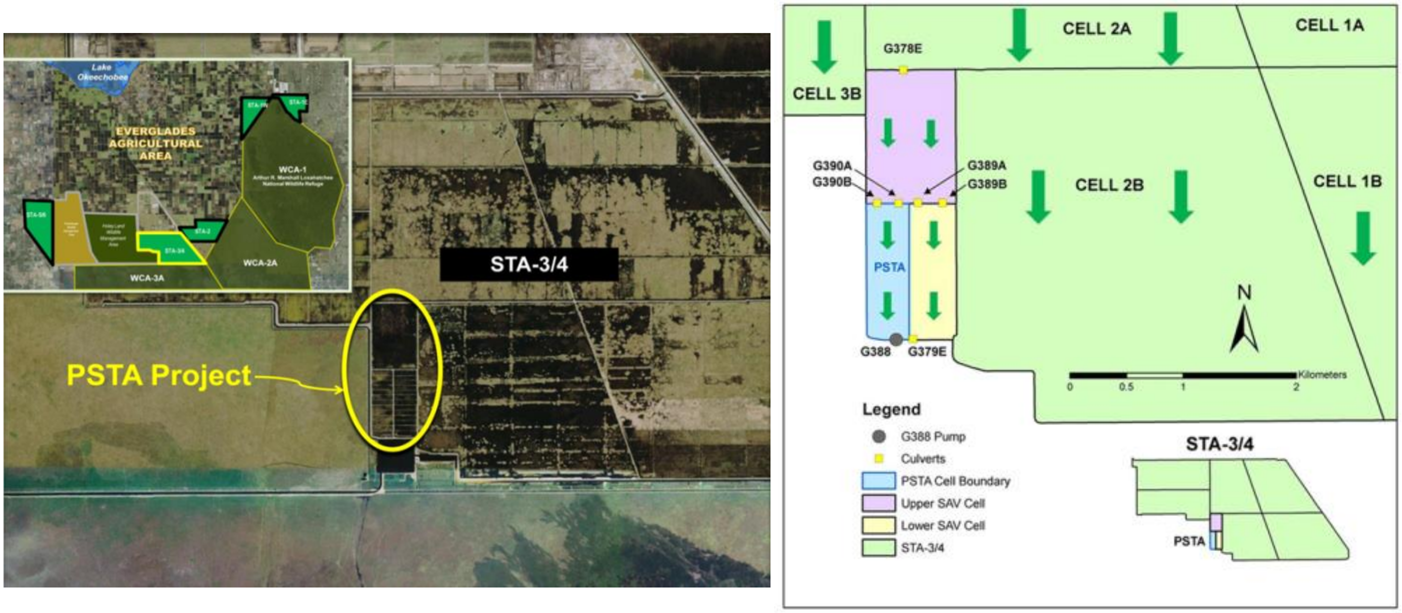

Figure 4-1. Location map of STA-3/4 and PSTA Project (left), including Upper and Lower SAV cells, PSTA Cell, and related water control structures (right); green arrows show flow direction.

The PSTA Cell has successfully developed into an oligotrophic, periphyton-rich wetland treatment cell. Due to conducive growing conditions, several species of submerged aquatic vegetation (SAV) have also colonized the PSTA Cell since it began operation. Over the years, visual observations suggest that the SAV community in the PSTA Cell has fluctuated in terms of species composition, relative density, and coverage. Periphyton colonization of the benthic surface and surfaces of submerged and emergent macrophytes has also fluctuated, but appears robust in many parts of the cell (**Figure 4-2**).

Figure 4-2. Periphyton growing on *Chara* sp. and *Eleocharis* stems in the STA 3/4 PSTA Cell.

The STA-3/4 PSTA Cell has shown promising performance by discharging outflow waters with ultra-low TP concentrations. During the first four years of operation (WY2008–WY2011), the PSTA Cell's flow-weighted mean (FWM) TP outflow concentrations ranged from 8 to 12 microgram per liter (µg/L), or parts per billion (ppb), with an overall average of 10 ppb. This treatment was achieved with an average annual inflow TP concentration of 20 ppb, and annual P loading rates of 0.14–0.42 g P/m^2/yr (Andreotta et al., 2014). Replicating the success of the PSTA technology across other STA flow ways would improve the SFWMD's ability to meet Water Quality-based Effluent Limits established to protect downstream marshes. However, several questions remain regarding the factors contributing to the success of the project. These questions need to be addressed before the PSTA technology can be implemented in other STAs.

Soils can be an important source of P to the water column in wetlands, and soil P flux typically increases with soil P concentration (Fisher and Reddy, 2001). Organic soil removal to expose a limerock substrate is a key design element that differentiates the PSTA technology from other STA flow ways. However, the magnitude of internal P flux has not been quantified for the PSTA Cell either in the original design condition (exposed limerock) or in the current operational condition (with newly accrued sediments).

In shallow lakes, SAV can obtain P exclusively from the sediments when bioavailable P is low in the water column (Barko and Smart, 1980; Carignan and Kalff, 1980). Under such circumstances, the macrophytes act as a nutrient "pump" by using sediment P for tissue growth, then releasing DOP and PP upon senescence. What is less clear is the effect of organic soil removal on macrophyte biomass and tissue P contents. High biomass or tissue P contents would suggest that macrophytes were an important source of internally recycled P.

Cultivation of a periphyton community that is well suited for ultra-low water column P concentrations is another central element of the PSTA technology. The community composition and standing biomass of macrophytes and periphyton within the PSTA cell may differ from STA flow ways with the organic soil intact, but this has not been investigated in sufficient detail. The spatial distribution of SAV species in the PSTA Cell was quantified in August 2008 and February

2009, and indicated dominance by *Chara* sp., with moderate coverage by *Najas guadalupensis*, and lesser amounts of *N. marina*, *Potamogeton* sp., *Hydrilla*, and *Ludwigia repens* also present (Pietro et al., 2010). *Chara* and *N. guadalupensis* are common to STA flow ways with muck soils, and isolated patches of *N. marina* and *Potamogeton* sp. have also been reported (Andreotta et al., 2014). However, little information exists on the periphyton community that occurs in the PSTA cell, or the macrophyte community that acts as a natural substrate for periphytic growth. If the macrophyte and periphyton communities are not different between the PSTA cell and muck-based cells, muck removal may not have had the anticipated effect of minimizing macrophyte growth in favor of periphyton development.

The PSTA Cell is operated to minimize the presence of emergent vegetation across most of the treatment area, in favor of benthic periphyton mats and epiphytes on SAV. The STA-3/4 PSTA Cell was constructed with twelve vegetated strips oriented perpendicular to flow to manage the hydraulic efficiency by impeding water conveyance under prevailing shallow-water conditions (DBE 2009). The strips are also thought to reduce wind-driven suspension and transport of newly-accrued flocculent sediments. However, the function of the emergent vegetation strips and the impact of management activities (such as herbicide application on the emergents) on the overall P removal performance of the PSTA Cell have not been carefully evaluated.

Over the first four-year operational period, there were numerous complications in collecting and interpreting the PSTA Project's hydraulic data. First, the accuracy of the flow data at all of the PSTA Project's water control structures was in question. Second, the amount of seepage entering the PSTA Cell from surrounding water bodies (i.e., the surrounding PSTA Project cells, STA-3/4 treatment cell, and discharge canal) was not known but was assumed to be quite large as evidenced by higher outflow than inflow volumes. Third, the quality of the seepage water was not known, making it difficult to calculate the P budget for the PSTA Cell. As a result of these various hydrologic and hydraulic issues, interpretation of the PSTA Cell's excellent performance over the first four water years has been challenging. Despite these challenges, the best estimates of P loading to the PSTA Cell indicates P loading rates have been modest as compared to other STA flow ways (Andreotta et al., 2014).

Examination of outflow P speciation data associated with the PSTA Cell suggests that ultra-low outflow TP levels are achieved through slight reductions in both particulate P (PP) and dissolved organic P (DOP), relative to outflow values in adjacent SAV cells. Reductions in these constituents may be the result of processes that are optimized by shallow water conditions. For example, shallow waters increase light penetration to the sediment (benthic) surface. The difference in light penetration to the benthic surface between depths of 1.5 ft and 3 ft can be dramatic in highly-colored surface waters typical of the STAs. McCormick et al. (2006) noted that nearly all photosynthetically active radiation (λ = 400-700 nm) was attenuated within the upper 30 cm (1 ft) of the water column in STA 1W Cell 4. Shallow conditions also increase contact between surface waters and benthic periphyton. If the PSTA technology is to be applied broadly, however, tolerance to deeper water conditions may provide the greatest operational flexibility to the SFWMD.

An important limiting factor in P removal is that a portion of the TP in surface water is DOP, a form less available for biological uptake or chemical sorption than SRP. Under certain conditions, however, DOP molecules are broken down through hydrolysis reactions into more bioavailable forms of P. Phosphatase enzymes are naturally-produced proteins that catalyze these hydrolysis reactions, effectively increasing the transformation of DOP into more bioavailable P forms. Phosphatases are found internally within organisms and as extracellular moieties within the aquatic environment (Wetzel, 1991).

Performing enzyme assays to estimate the activity of phosphatase enzymes in the PSTA Cell has two potential applications. First, the transformation of DOP to SRP may be an important mechanism operating in areas that achieve ultra-low TP concentrations. The coupling of algal photosynthesis with bacterial metabolism in periphyton mats has been shown to enhance bacterial enzyme production, compared to conditions where photosynthesis was inhibited (Espeland and Wetzel, 2001). Thus, photosynthetically productive periphyton mats may be able to achieve higher levels of enzyme production than systems where photosynthesis is reduced, either by deep colored water or macrophyte shading. A second application of enzyme assays is based on the fact that phosphatase activities can be repressed by the presence of bioavailable SRP, a property that makes activity measurements suitable indicators of increased P availability. Alkaline phosphatase activities in surficial sediments and on the roots of emergent macrophytes (*Eleocharis cellulosa* and *Typha domingensis*) were suppressed by (negative correlated to) P availability in wetland soils (Rejmánková and Macek 2008). Since the PSTA Cell and STA outflows are very low P environments, it is useful to have an alternate, sensitive indicator of increased P availability that can detect limits to PSTA Cell performance sustainability before outflow P concentrations increase (Newman et al., 2003).

Enzymes can become deactivated when bound to DOM compounds, but the process is potentially reversed by UV photolysis of the complexing organic matter (Boavida and Wetzel 1998). Organic soils can be important sources of DOM to wetland surface waters, including monoesters and diesters of organic phosphorus (Turner and Newman, 2005). Photolysis by UV radiation can degrade DOM and stimulate bacterial metabolism (Wetzel et al., 1995). Because UV radiation is strongly absorbed by DOM in surface waters, photolysis is likely to occur only in shallow water. Therefore, the interactions between water depth, light availability, periphytic growth, and enzyme activity are important to DOP processing in STA surface waters.

The primary objective of this study is to determine design and operational factors that contribute to the PSTA cell's superior performance, in order to facilitate large-scale replication of the success of this technology. Data gathered from the PSTA Cell will provide needed information on the effects of muck removal, water depth and hydraulic and P loading on treatment performance. Alternate approaches to immobilize soil P, and specific mechanisms contributing to sustainable low-level TP removal will be tested in replicated mesocosm studies. These data will be critical to any future feasibility analysis of using PSTA technology to improve STA back-end P removal performance.

4.4 STUDY PLAN OBJECTIVES

In addition to the primary objective described above, the following questions are posed to address gaps in the current understanding of the PSTA technology related to design, operations, and sustainability. For each question, several research hypotheses are provided that will be tested by the experimental and monitoring efforts described in this Study Plan. While these questions and hypotheses are in part related to the SFWMD Science Plan questions and sub-questions noted previously, they are specifically targeted to elucidate critical factors that will guide evaluation of the feasibility of full-scale implementation of the PSTA technology.

What are the important design elements that enable the PSTA Cell to achieve ultra-low outflow TP levels? (muck removal, compartmentalization)

Hypothesis #1: Removal of muck soils in the downstream portion of an STA flow-way reduces or eliminates the flux of P from the soil to the water column, which in turn contributes to conditions favorable to low outflow TP concentrations.

Hypothesis #2: The removal of muck soils, or the reduction of bioavailable P levels in muck soils through the use of limerock or other soil amendments, will decrease soil P availability to macrophytes, resulting in reduced growth rates, decreased tissue nutrient content and biomass turnover, and in turn, a shift towards periphyton dominance and lower outflow surface water TP concentrations.

Hypothesis #3: The compartmentalization provided by vegetated strips has resulted in strong internal gradients in sediment accumulation by decreasing turbulence in the water column, and minimizing resuspension and transport of flocculent sediments

What are the key operational ranges that enable the PSTA Cell to achieve ultra-low outflow TP levels? (P Loads, HLR, and water depth)

Hypothesis #4: Low outflow TP concentrations have resulted from the moderation of hydraulic loads and P loading to the PSTA cell. Higher P loads will compromise treatment efficacy and result in increased outflow TP concentrations.

Hypothesis #5: Stable and shallow water depths have contributed to the superior performance of the PSTA cell, due to factors such as enhanced UV penetration throughout the water column; increased operational water depths will result in higher outflow TP concentrations.

Hypothesis #6: Shallow water depths (and a low surface water level compared to surrounding water levels) increase groundwater interaction, which in turn has led to low outflow TP concentrations for the STA-3/4 PSTA cell.

What management practices are required to sustain the PSTA Cell's good performance? (sediment management, vegetation management)

Hypothesis #7: Over time, accrued sediments in a PSTA Cell become a source of P to the water column and result in increased outflow TP concentrations compared to the initial condition when the cell bottom was mainly comprised of limerock substrate. Accumulation of sediment will result in elevated outflow TP concentrations, compared to a bare limerock substrate.

Hypothesis #8: Periodic drawdown will benefit PSTA cell performance by consolidating sediments, and helping maintain low macrophyte densities.

4.5 DETAILED STUDY PLAN AND EXPERIMENTAL DESIGN

4.5.1 Study Plan Description

This Detailed Study Plan, when implemented, will produce a review of existing data on the PSTA technology, the development of a conceptual model for PSTA P removal pathways, an assessment of treatment performance under a range of operational conditions, spatial analysis of the PSTA Cell's surface water, periphyton, vegetation and sediment characteristics, and experimental research to evaluate key processes and design assumptions. The main study area is the PSTA Cell, while comparisons will be also made to muck-based SAV cells (i.e., STA-3/4 Cells 2B and 3B, the Upper SAV Cell and the Lower SAV Cell). Water quality will be monitored at inflow/outflow structures and internal transects by multiple approaches [grab samples, interior auto-samplers, Remote Phosphorus Analyzers (RPAs)] to evaluate performance under different operational (flow and stage) conditions. Because of the inherent limitations to conducting multiple assessments with a single field-scale treatment cell, bench-scale and mesocosm studies are also planned to investigate in replicate the influence of key factors on P removal performance. These factors include the type of substrates (limerock versus muck), the influence of macrophytes versus periphyton on P uptake, the influence of water depth on community characteristics and outflow P levels, and the influence of different inflow P concentrations and nutrient loading rates on outflow P levels.

A preliminary review of existing data on the PSTA technology is provided in Appendix A. A conceptual model for the PSTA concept is under development, and details are provided in Appendix B. In the following section, the methods for each study are presented in relation to the research hypotheses listed above. For the sake of continuity, the components of each study are summarized together under the primary hypothesis to be addressed, though most studies will provide data for testing several of the hypotheses outlined above. A complete list of monitoring activities, frequencies, and parameters is summarized in Appendix C. The overall duration for the research and monitoring effort, including sample collection and data analysis, is FY2012 through FY2016.

Hypothesis #1: Removal of muck soils in the downstream portion of an STA flow way reduces or eliminates the flux of P from the soil to the water column, which in turn contributes to conditions favorable to low outflow TP concentrations.

Organic soils in wetlands can be a source of P to overlying waters or act as a P sink when waters are P-enriched. Intact soil core studies are useful for determining relative soil P flux rates under controlled conditions, and compared favorably to flux measurements using in situ benthic chambers (Fisher and Reddy, 2001).

Several approaches are being undertaken to examine this hypothesis. First, intact core studies were conducted to compare short-term P flux from PSTA sediments into the overlying water with P flux from muck soils. Second, a survey of sediments in the PSTA cell was conducted to compare P concentrations between the PSTA cell and muck soils from adjacent flow ways. Finally, a longer-term (~9 months) study compared water column TP concentrations in outdoor, flow-through columns established on exposed limerock and muck soils.

PSTA Intact Soil Core Studies

To improve understanding of the roles of soils, macrophytes and periphyton on the ability of the PSTA Cell to achieve ultra-low-level outflow TP concentrations, a series of intact core studies were carried out. The first (Core study #1) compared P flux or uptake by newly-accrued sediments from the PSTA Cell to areas where the organic muck soils remained intact, under stagnant, un-vegetated conditions. This study addresses whether existing muck soils are more or less stable with respect to sequestered P, than the newly-accrued sediment material in the PSTA Cell. Follow-on core studies (#2 and #3) examining soil effects on macrophytes and periphyton are described under Hypothesis #2.

Intact soil cores were collected from STA-3/4 Cells 2B and 3B, and PSTA soil cores were constructed by placing 5 cm PSTA Cell sediments on top of 10 cm limerock. Water was collected from the PSTA Cell inflow and the Cell 2A outflow to achieve a target reflood water TP concentration of ~20 ppb. The water in the cores was sampled twice weekly: after 4 days for TP, and after 7 days for TP, TSP, SRP. After each Day-7 sampling, the water column was exchanged with fresh reflood waters. Core Study #1 encompassed 4 water exchanges for a total of 28 days.

Total P and SRP concentrations changes over time were used to determine P flux rate from each soil type. Phosphorus speciation was used to characterize surface water P as SRP, DOP (TSP-SRP), or PP (TP – TSP). Additional parameters (DOC, Alkalinity, dissolved Ca, alkaline phosphatase activity, and UV-Visible absorbance) were monitored to examine the effects of soil type on DOM in the water column (Hypothesis #5). Data collected from these studies is currently being analyzed and the interpretation and summary of results is underway.

PSTA Cell Sediment Chemistry

Sediments were collected in May 2012, along inflow, middle and outflow transects, to estimate sediment P storage in the system. Sediments were analyzed for bulk density, TP, TN, TC, ash-free dry weight and Ca content. Sediment samples will be collected again in WY2014 at the same locations to calculate P accumulation rate in the sediments, and look for increased P concentrations as an indicator of diminished sustainability of low sediment P flux conditions (**Figure 4-3**). Together with a survey of accrued sediment depth (see Hypotheses #3 and #8), the sediment chemistry survey provides a basis for comparing P storage by PSTA sediments to accrued sediments in muck-based treatment cells.

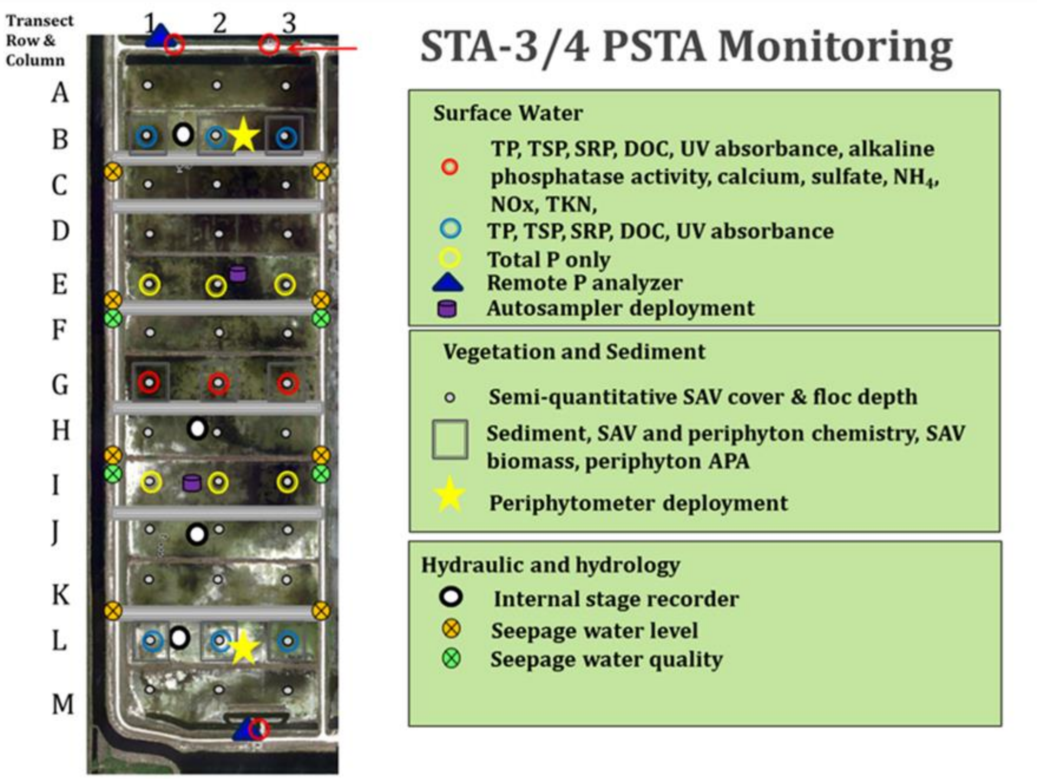

Figure 4-3. PSTA Cell monitoring and sampling locations.

Flow-through Columns

A survey of sediments in the PSTA Cell indicated that some areas contain residual amounts of muck overburden that either were not removed during construction, or gradually sloughed into the cell from the vegetated strips or adjacent levees (see *Sediment Depths and Accretion Rate* section under Hypothesis #3, and 2013 SFER – Volume I, Chapter 5). This effort will evaluate the long-term effects of soil characteristics on P flux and P removal performance under flow-through conditions. Whereas surficial muck layers remained intact in the Core Studies described above, in this Column Study the surficial soil layer will be removed from muck treatments to simulate partial removal of the organic soil during PSTA Cell site preparation.

The Column Study consists of nine 30-cm diameter cylinders, containing sediments with an overlying water column. There are three substrate treatments: Exposed limerock (LR) from the

PSTA Cell; muck soil from the Lower SAV Cell with the accrued layer removed and the upper muck layer exposed to water column (UPPER); and muck soil from the Lower SAV Cell, with accrued layer and upper muck removed so that the lower muck layer is exposed to water column (LOWER). All treatments will be established in triplicate columns at 1.5 ft water depth using PSTA Cell inflow water collected from G-390B. Chara and periphyton will be collected from the PSTA Cell and placed in each column. Water column TP concentrations and phosphatase enzyme activities will be monitored over time to assess substrate effects on soil P flux, water column TP concentrations, and bioavailable P. Additional parameters (DOC and UV-Visible absorbance) were monitored to examine the effects of soil type on DOM in the water column (see Hypothesis #5). In a follow-on study, columns with exposed limerock will be compared to columns established on intact soils (surficial layer retained) from muck-based STA-3/4 Cell 3B.

Hypothesis #2: The removal of muck soils, and/or the reduction of bioavailable P levels in muck soils through the use of limerock or other soil amendments, will decrease soil P availability to macrophytes, resulting in reduced growth rates, decreased tissue nutrient content and biomass turnover, and in turn, a shift towards periphyton dominance and lower outflow surface water TP concentrations.

To address this hypothesis, the condition of macrophytes in the PSTA Cell will be surveyed, including species distribution, standing crop biomass and tissue P contents. Areal biomass, growth rate, taxonomy, tissue P contents and enzyme activity of periphyton will also be determined at stations within the PSTA cell. The characteristics of the macrophyte and periphyton communities in the PSTA cell will be compared to previous findings from other STA flow ways.

The above hypothesis will also be evaluated by measuring macrophyte growth response to different sediment conditions (accrued sediment from the PSTA Cell compared to muck soils) in Core Studies #2 and #3. The changes in plant and periphyton biomass and nutrient content will be assessed as a function of soil type. A similar comparison will be made under flow-through conditions in the Column Study. Finally, the vegetation response to sediment management will be explored in an outdoor mesocosm study where several alternatives to muck removal are being evaluated. Details of each of these experiments are provided below.

PSTA Cell Macrophyte Survey, Sampling and Analysis

A semi-quantitative vegetation monitoring scheme will include surveys conducted twice per year within the PSTA Cell and adjoining muck-based SAV cells. These surveys determine the relative density and coverage of dominant SAV species, using an approach developed by DBE for SAV-dominated STA cells (Andreotta et al., 2014). The presence or absence of various macrophyte species is recorded, along with a density or cover ranking on a scale of 1 to 5. Spatial maps of the SAV species distribution will be constructed using Arc GISv9 Spatial Analyst (Environmental Systems Research Institute, Redlands, CA). Examples from two surveys conducted in WY2013 are provided in Andreotta et al. (2014). Chara dominates the SAV community in the middle and outflow regions of the PSTA Cell. Therefore, this species was selected for inoculation into PSTA Core Study #2 and the Column Study.

On a coarser spatial scale, macrophyte vegetation samples were collected in August 2012 from the PSTA Cell along the inflow-to-outflow gradient to assess standing crop biomass, as well as tissue N, P, C and Ca concentrations. Sampling locations are shown in

Figure 4-3. Sampling will be repeated twice per year to determine temporal trends in tissue P concentrations and biomass P storage. These values will be compared to data from previous studies in other STA flow ways (e.g., Vegetation Biomass and Nutrient Analysis for STA-1W (DBE, 2002). If the standing crop of macrophyte biomass is lower in the PSTA Cell than is reported for muck-based cells, then the organic soil removal may be responsible.

PSTA Cell Periphyton Survey, Sampling and Analysis

The effects of removing muck soils as an internal nutrient source may include the development of unique periphyton communities in the PSTA Cell, relative to other muck-based systems. On two occasions annually (wet and dry seasons) samples of periphyton will be collected at inflow, middle and outflow stations in the PSTA cell and analyzed for biomass per unit area and nutrient concentrations. Periphyton samples were collected on April 24, 2013 and August 8, 2013 from glass slide periphytometers deployed in the PSTA Cell for a 6-week incubation period.

A linkage between P removal performance and the species composition of the biological communities operating in the STAs would greatly improve the ability to identify key mechanisms that create and maintain ultra-low P concentrations in the surface waters. Therefore, periphyton community composition will be classified according to dominant species by biovolume. Phosphatase enzyme activity will be also assayed in conjunction with the periphyton taxonomic effort.

PSTA Core Studies: Sediment Effects on Macrophyte and Periphyton Communities

The water columns in Core Study #1 were un-vegetated, yet many parts of the PSTA cell are colonized by SAV. Submerged aquatic vegetation can reduce apparent sediment P flux rates by absorbing inorganic P that diffuses from sediments into the water column. Alternatively, SAV can act as a nutrient pump by incorporating sediment P into plant tissue and then releasing P compounds during senescence (Carignan and Kalff, 1982).

To examine the role of SAV on sediment P flux, a second study (Core Study #2) was conducted using the intact sediment cores from Core study #1. Half of these cores were planted with *Chara* sp., the dominant SAV species found in the PSTA Cell. The other cores remained un-vegetated. Vegetated cores received 4 pieces (each 15 cm length) of lightly epiphytized *Chara* collected from the PSTA Cell. The water in each core was sampled once weekly, and then exchanged with fresh reflood waters, for 4 cycles over a 28-day period. Total P concentrations and enzyme activity in the water column were determined for each core. Changes in P concentration over time were used to calculate a P flux rate. Differences in soil P flux between PSTA sediments and muck soil were evaluated simultaneously under both vegetated and un-vegetated conditions. Enzyme activity was measured as an alternate indicator of P bioavailability in the water column, as a function of soil type and presence/absence of SAV.

Chara exists in many areas within the STAs. However, *Chara* found within the PSTA Cell often appears heavily colonized by periphyton, relative to *Chara* in other STA flow ways. In the 3rd phase of the intact core studies (Core Study #3), *Chara* and periphyton communities were compared. To the un-vegetated cores from Core Study #2, periphyton from the PSTA Cell was added. The water exchange frequency was extended to once every 14 days, while sampling for surface water TP and enzyme activity continued on a weekly basis for 40 days. These data were used to assess the effects of the presence of *Chara* and periphyton on sediment P flux, surface water TP concentrations and alkaline phosphatase activity. Data collected from these studies is currently being analyzed and the interpretation and summary of results is underway.

Mesocosm Study: Alternatives to Muck Removal

A review of the performance of limerock-based treatment systems and related research efforts, including previous PSTA platforms, indicated that the PSTA Cell in STA-3/4 is unique in its ability to achieve TP outflow concentrations less than or equal to 10 ppb for multiple years (see Appendix A). Removal of organic soils above the limerock substrata clearly will increase costs associated with STA construction, however, and the benefits of organic soil removal remain unclear.

An outdoor mesocosm study was established in 2011 to investigate the ability of soil amendments (applied onto muck soils) to achieve low outflow TP concentration consistently over time. Two treatments were established using soil amendments applied as a cap over existing muck soils. These include crushed limerock and aluminum -based water treatment residuals (WTR), a stable material with high affinity for P adsorption. A third treatment consists of WTR mixed into the surficial muck soils. These treatments are being compared to a control treatment on un-amended muck soils. The mesocosms are being operated with inflow water from the discharge canal of STA-1W, with additional pre-treatment as necessary when the STA performance is not adequate to provide inflow TP levels in the range of 15-40 ppb. The inflow and outflow TP concentrations will be monitored for a three-year period.

Dry-out can cause short-term increases in water column P from senescent vegetation or soil P mineralization. The soil amendment alternatives examined in this mesocosm study may mitigate sediment-P or biomass-P release following dry-out by increasing P uptake potential of the sediments. During the course of the study, one dry-down event is planned to assess the potential effect on P removal performance. Prior to dry down, the macrophyte biomass standing crop in each treatment will be assessed by collecting aboveground plant tissues from a portion (e.g., 0.5 m x 0.5 m quadrat) of each mesocosm. The effect of soil amendments on macrophyte standing crop and water column P will then be evaluated to support or refute the above hypothesis (#2).

Hypothesis #3: The compartmentalization provided by vegetated strips has resulted in strong internal gradients in sediment accumulation, by decreasing turbulence in the water column and minimizing resuspension and transport of flocculent sediments.

When the PSTA Cell was constructed, although most of the peat was scraped and removed, a portion of the scraped peat material was left on site and reconfigured into twelve vegetation strips (each 18”- 24” tall) oriented perpendicular to flow (**Figure 4-4**). The emergent vegetation strips were created within the PSTA Cell to improve hydraulic efficiency (Burns & McDonnell, 2003). *Eleocharis* was planted along these strips during the initial construction, and herbicide was applied periodically to confine these emergent macrophytes to the strips. The importance of the emergent vegetation strips and the impact of management activities (such as herbicide application) on the overall P removal performance of the PSTA Cell have not been scientifically evaluated.

A tracer analysis of cell hydraulics performed in July 2009 under flowing conditions (15-28 cfs) found a hydraulic retention time of 4 days, low dispersion and efficient hydraulic performance (DBE, 2009). In WY2012, in preparation for pulse events (short-term increased hydraulic loads to the cell of about 60-90 cfs), the vegetation on six of the twelve emergent vegetation strips was sprayed and compacted to reduce the amount of hydraulic resistance in the cell. The modified vegetation strips will be maintained with herbicide during the remainder of the PSTA Study.

Several pressure transducers have been installed at various locations throughout the PSTA Cell to support an evaluation of the PSTA cell’s hydraulics before and after the vegetation strip modifications (see Hypothesis #7). In addition, a spatial assessment of the depth of sediment accretion will be conducted annually. Data sondes will be deployed at several internal locations to record short-term fluctuations in water column turbidity. These efforts are described in more detail in the following section.

Sediment Depth and Accretion Rate

Sediment accretion depth was measured in the PSTA Cell during WY2012 (1/18/2012) and WY2013 (11/8/2012) along the inflow to outflow gradient. During each survey, depth of organic material above bedrock was determined *in situ* using a tape measure and tile probe. Intact cores were retrieved to verify accrued sediment depths. The measurements were made along 13 transects within each compartment of the PSTA Cell defined by the vegetated strips (**Figure 4-3**). Thickness of the underlying residual muck (where present) was determined as the difference between total sediment depth to bedrock and the accrued sediment depth. One additional survey will be completed in WY2014. Evidence of strong longitudinal gradients in accretion depth would support the hypothesis that vegetated strips provide stability to the movement of flocculent sediments within the cell. Changes over time in the depth or spatial distribution of accrued sediments within the PSTA Cell will be evaluated in relation to the removal of the vegetation from six vegetated strips in April-May 2012.

Figure 4-4. Aerial photo of the outflow region of the PSTA Cell in STA-3/4, showing several emergent vegetation strips and the outflow pump structure G-388. The vegetation on six of the twelve strips in the PSTA Cell was treated with herbicide in April 2012, then compacted to minimize hydraulic resistance during hydraulic (pulse) events conducted in July–August and October 2012.

In-situ diel dynamics of pH, temperature, conductivity, dissolved oxygen (DO), and turbidity have been recorded during “typical” and “pulse” flow conditions (see Hypothesis #4 for details), using data sondes installed at PSTA Cell interior locations. Data sondes have been deployed in the PSTA Cell during a period of low flow conditions (May – June 2012), and during two hydraulic pulses (July 2012 and October 2012). Sondes were deployed in the PSTA Cell in May 2013 for turbidity only. Sondes were recently deployed alongside periphytometers in the PSTA Cell, Cell 3B of STA 3/4, and Cell 1 of STA 2 in March and April 2013, and December 2013–January 2014. These data will provide a means for evaluating differences in sediment physical stability or resuspension as a function of flow rate and position within the PSTA cell.

Hypothesis #4: Low outflow TP concentrations have resulted from the moderation of hydraulic loads and P loading to the PSTA cell. Higher loads will compromise treatment efficacy and result in increased outflow TP concentrations.

Surface water quality and flow rates at the PSTA Cell inflow and outflow structures are routinely monitored by the District. This ongoing effort provides the fundamental information necessary to assess PSTA Technology P removal performance under normal operational conditions, including storm-driven hydraulic pulses, quiescent periods and potentially during post-drought reflooding. To supplement these data and test the above hypothesis, the PSTA study team has conducted intensive monitoring of the field-scale PSTA cell under induced high-flow conditions (managed “pulse” events), and is conducting mesocosm-scale experiments to examine water TP and enzyme response to changes in P loading rate.

Intensive Monitoring of Field-Scale Pulse Events

While the historical (WY2008–WY2012) annual HLR for the PSTA Cell was generally within the ranges observed for Cell 2B, the peak flow rate in the PSTA Cell (as indicated by the pulsing factor, peak flow/average flow) was considerably lower than in Cell 2B. Therefore, the flows delivered to the PSTA cell during its period of high performance may not adequately represent the conditions that occur in other STA flow ways. This PSTA Study includes several pulse events in the PSTA Cell to evaluate the effect of higher flow rates on cell performance.

Surface water samples are collected weekly at inflow structures (G-390A and G-390B) and the outflow structure (G-388) of the PSTA Cell. Inflow and outflow structures of adjacent muck-based treatment cells (STA-3/4 Cells 2B and 3B, and the Upper and Lower SAV Cells) are also monitored for a performance comparison to muck-based cells. In order to obtain inflow and outflow P concentration data at more frequent time intervals, RPAs have been installed at the G-390 and G-388 structures, and at STA-3/4 Cell 2B and Cell 3B outflow locations. The RPAs collect data every three hours thereby capturing any short-term fluctuations in TP and TRP. The units will remain deployed year-round to obtain a clear characterization of the concentrations in and out of the PSTA cell in relation to a variety of operational conditions (high and low flows, different seepage conditions, diel/diurnal measurements, and any natural or controlled pulsed flow events).

Two auto-sampler (A/S) units will be deployed internally within the PSTA Cell to collect water TP samples at 3-hr intervals (**Figure 4-3**). The A/S unit deployments will last approximately one week, and will occur during seasonal flow, no flow, and pulsed flow periods.

In addition, three internal monitoring events are planned in the PSTA Cell each year for three years. The goal is to collect data under both stagnant and pulsed flow conditions, and sample before, during, and after pulsed flow events. Station locations are summarized in

Figure 4-3. Surface water P and N species concentrations will be determined. Nitrogen to P ratios can be used to determine whether P limitation increases with distance through the PSTA Cell. Dissolved organic carbon concentrations and UV absorbance properties of surface water samples will be used to characterize the relative size and recalcitrance of the constituents that comprise the dissolved organic matter (in support of Hypothesis #5). Enzyme activities will be measured to determine the potential role of microbial activities in P cycling under stagnant conditions, and under moderate and high flow conditions. When coupled with vegetation surveys, internal water quality monitoring enables comparison of vegetation cover and health with treatment performance. Spatially interpolated maps and graphics will be constructed to identify water quality trends along the inflow-outflow gradient.

Mesocosm Study: P Loading Effects in PSTA Raceways

A second mesocosm study will be conducted to examine surface water TP concentration response to variable P loading conditions. This will be accomplished by establishing periphyton communities in triplicate raceways, using limerock as the substrate. Each raceway will consist of four tanks plumbed in series so that the total effective treatment length of each raceway is ~10 m (32 ft). Inflow water will be delivered to each raceway by gravity from a head tank at the southern research facility in STA-1W where TP concentrations are expected to be ~20 ppb TP.

Phosphorus uptake through each tank is likely to create a nutrient gradient, such that the water passing to each subsequent tank is lower in P concentration until a “background” concentration is achieved. As the nutrient gradient becomes established, routine monitoring of surface water TP concentrations and phosphatase enzyme activities will provide a basis for comparison to the effective nutrient load to each tank along the raceway. As an alternate response variable, the vegetation in each tank will be characterized periodically through careful observation and periodic tissue P analysis to evaluate competition between periphyton and two SAV species (*Chara* and *Potamogeton*) that are found in the outflow region of well-performing STA flow ways. Efforts for this study will be initiated in early 2014 and will continue through the end of 2015.

Hypothesis #5: Stable and minimal water depths have contributed to the superior performance of the PSTA cell, due to factors such as enhanced UV penetration throughout the water column; increased operational water depths will result in higher outflow TP concentrations.

While the attenuation of light with water depth is a well-established phenomenon, it is not known if algal growth is light-limited or nutrient-limited under the prevailing conditions of the PSTA cell. The interactive effects of water depth and optical clarity will be examined through a combination of PSTA cell monitoring and experimental research. The PSTA cell was operated within a narrow water depth range of about 1.3’ (see discussion below on 2013 survey) for the majority of the period of record. By adjusting the protocols governing outflow pump operations, in mid-2013, the operating depth was increased by 0.5’ to about 1.8’ and will be operated at this increased depth for a period of about one year. Temporal and spatial monitoring of surface water P (described under Hypothesis #4) will continue during the increased-water depth period. Changes in outflow P concentration will be determined to assess the effect of deeper water conditions on PSTA cell performance. Soil core study #1 (described under Hypothesis #1, above) will test whether muck soils contribute DOM and increase attenuation of UV radiation. Monitoring surface water in the full-scale PSTA Cell (Hypothesis #4) will examine the attenuation of PAR and UV radiation under stagnant and flowing conditions. Finally, a mesocosm experiment will measure algal growth response to a range of water depths.

PSTA Core Study #1

In addition to inorganic P flux, organic muck soils can be a source of DOM to the water column. Dissolved organic matter can influence P cycling directly with contributions of P-containing compounds. Effects of soil-derived DOM can also be indirect, for example, by complexing extracellular enzymes, by reducing PAR and UV radiation (affecting photosynthesis and photolysis, respectively), or by providing micronutrient and energy sources for microbial metabolism (Boavida and Wetzel, 1998; Wetzel et al., 1995).

The influence of sediment type on surface water DOP concentration was cross-examined with changes in DOC concentration and UV absorbance properties (indicators of DOM recalcitrance). Differences in autotrophic activity within the water column, as influenced by soil type, were evaluated indirectly by monitoring constituents (i.e., dissolved calcium concentrations, alkalinity, and pH levels) that are commonly affected by photosynthesis in the water column. The phosphatase activities of surface waters within each core were assayed fluorometrically to provide an additional response variable for the effects of substrates on P removal performance. Data collected from this study is currently being analyzed and the interpretation and summary of results is under way.

PSTA Water Depth Mesocosms

In a companion study with the mesocosm effort described in the preceding section, the effects of water depth on P removal performance will also be examined. Using low-nutrient water and limerock as a substrate, periphyton and macrophytes will be established in mesocosms at depths ranging from 9 to 36 inches (23 to 91 cm). Each water depth treatment will be established in triplicate process trains, and consist of multiple tanks in series. The effect of water depth on algal growth rates will be examined by sampling periphyton from the benthic surface and/or artificial substrates placed in the mesocosms. Routine monitoring of TP concentrations in the surface water will be conducted to assess P removal performance under different water depth regimes. Vegetation will be assessed periodically to evaluate relationships between macrophyte species composition and the gradients in surface water and nutrient loading along each process train.

Hypothesis #6: Shallow water depths (and lower water stage compared to surrounding areas) increase groundwater interaction, which in turn has led to low outflow TP concentrations in the STA-3/4 PSTA cell.

Operation of the STA-3/4 PSTA cell has highlighted several challenges associated with constructing an accurate water balance. The cell’s shallow water depth, large head differences from surrounding areas, and a lack of a muck-soil layer each contribute to a high potential for groundwater interaction. Seepage contributions to the PSTA Cell water balance are important to determine if the quality and quantity of seepage water are responsible for the low outflow P concentrations. Such conditions could be site specific, making efforts to replicate the success of the PSTA technology problematic. To test the above hypothesis, this PSTA Study will conduct a thorough assessment of PSTA Cell hydrologic data, including seepage analysis through ion mass balance, refined water depth values with improved ground elevation data and internal stage monitoring, and increased accuracy of flow estimates at structures.

Seepage Analysis through Ion Mass Balance

A mass balance analysis of major ions in the well samples and inflow and outflow surface waters will be used to better quantify seepage and its contribution to the mass balance. The P concentration and chemical composition of surficial groundwater will be determined by sampling wells along the PSTA Cell levees (**Figure 4-5**). Seepage into the PSTA Cell from adjacent treatment cells (Upper and Lower SAV cells, Cell 2B) and the discharge canal will be quantified to improve the water and mass balance for the PSTA Cell.

Depth from top of the well casing to the static water level was measured bi-monthly in all twenty wells, from January 2012 through January 2014. Measurement was conducted using an electronic water depth probe. Groundwater samples were collected quarterly in shallow and deep wells during 2012 and 2013. Groundwater samples are being analyzed for the following parameters to include the major ions, Ca, Mg, Na, K, Cl, SO_4, and alkalinity. TP and SRP will also be analyzed. Water level and water quality monitoring will continue through 2014.

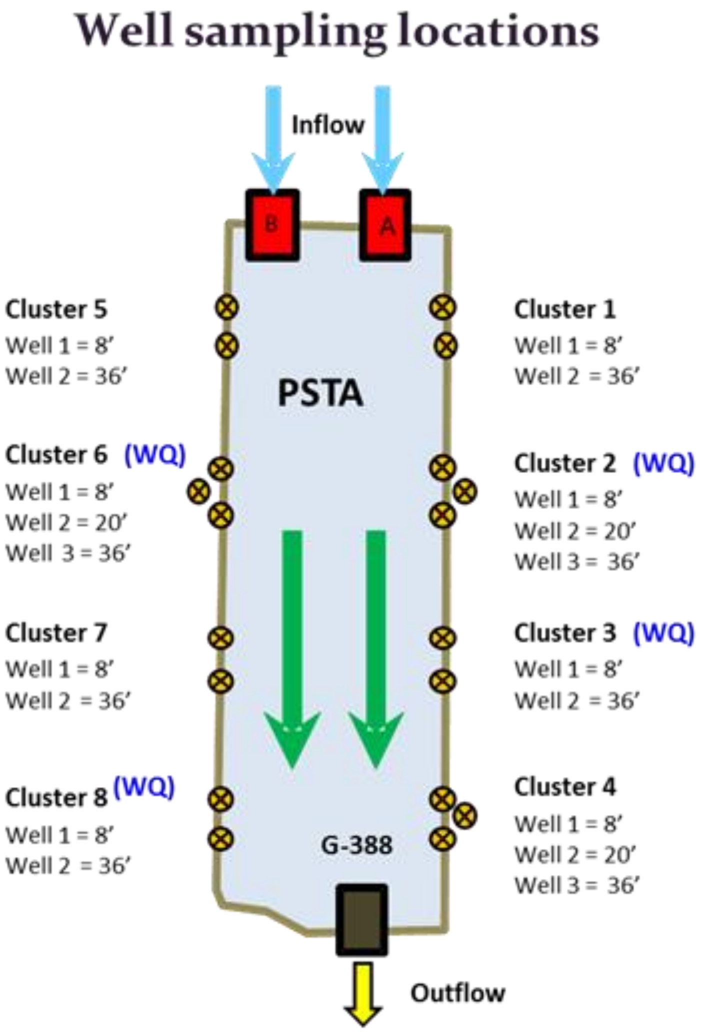

Figure 4-5. PSTA groundwater and seepage monitoring locations.

Ground Elevation Surveys

For the first 5 years of PSTA Cell operations (WY2008-WY2012), the nominal operating depth (2 ft) was based on a caprock elevation of 8.1' NGVD using topographic survey data that was collected before the muck was scraped from the cell. Recent field measurements indicated that water depths in the cell were less than 2 feet. A topographic survey of caprock elevations under current conditions was conducted in 2013 in the PSTA Cell and the adjacent Lower SAV Cell, at stations indicated in **Figure 4-6**. A total of 63 survey points were recorded in the PSTA Cell, including 24 points on vegetation strips and 39 points on bedrock surface between strips. In the Lower SAV Cell, 24 points spaced evenly in the cell were surveyed to establish the average ground elevation. The results of these surveys will be used to evaluate previous performance, update current operation target stages and to derive future operational ranges for the PSTA Cell. Based on the results of the 2013 survey, the average ground elevation in the PSTA Cell is 8.7 ft NGVD, confirming field observations that the water depths were shallower than the originally assumed 2 ft.

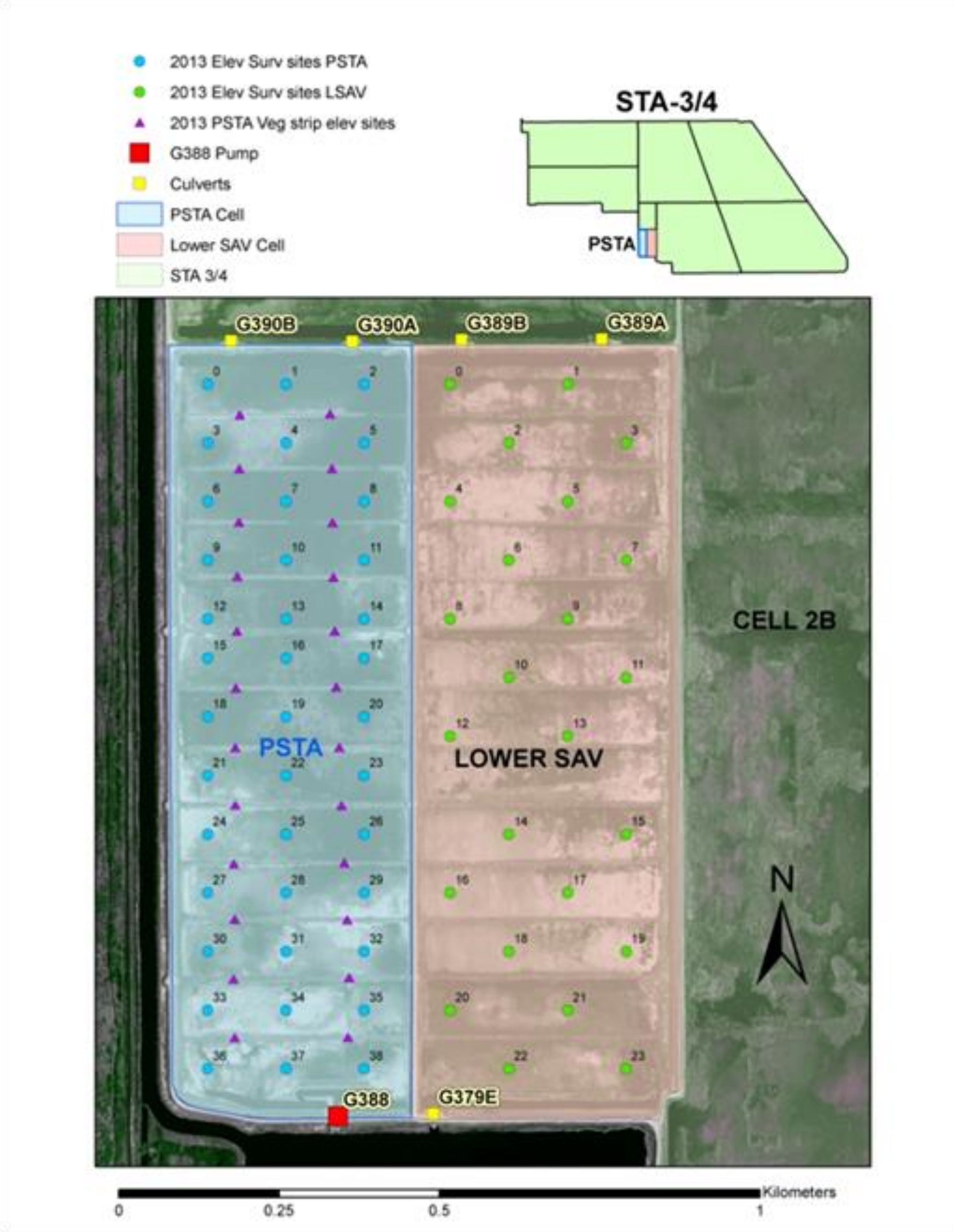

Figure 4-6. PSTA and SAV Cell 2013 elevation survey locations.

Pressure Transducer Deployment (Continuous Water Depth Monitoring)

In addition to the hypothesized effects of the vegetation strips on the PSTA Cell's hydraulics (see Hypothesis #3), previous field observations also suggested that the PSTA Cell outflow structure may have a strong localized effect on water depths in the south end of the PSTA Cell. For this reason, pressure transducers located near the outflow region of the PSTA Cell have been installed to examine the influences of the G-388 pump cycles on internal water depths. Water depth measurements from each transducer are collected every 15 minutes and data is downloaded on a monthly basis. This effort will continue through the end of WY2016. This study will also use stage and flow data collected at the PSTA Cell inflow and outflow structures and stage data at the pressure transducers inside the PSTA Cell during a range of flows before and after the vegetation strip modifications.

Improved Hydrologic Data

Prior to WY2012, assessment of the PSTA Cell's performance and factors influencing its performance was limited due to large uncertainties in flow and seepage data and lack of a comprehensive investigation. In WY2012, several enhancements were implemented in the PSTA Cell to improve structure flow estimates, including modifying the G-390B inflow culvert and modification of the G-388 outflow pump and its operation.

Average water depths inside the PSTA Cell will be estimated using the average of inflow G-390 tailwater (TW) stage and the outflow G-388 headwater (HW) stage minus the updated average ground elevation. The Upper SAV Cell, Lower SAV Cell, Cell 2B, and Cell 3B will serve as comparative units**.** Flows in and out of PSTA Cell are tracked on a weekly, monthly and annual basis. Improved flow estimates for the PSTA Cell will be produced as a result of the modifications of the G-390B inflow culvert and the G-388 outflow pump station; this improved flow data will increase the accuracy of the performance evaluation of the PSTA cell starting at WY2012.

Hypothesis #7: Over time, accrued sediments in a PSTA Cell become a source of P to the water column and result in increased outflow TP concentrations compared to the condition when the cell bottom was mainly comprised of limerock substrate. Accumulation of sediment will result in elevated outflow TP concentrations, compared to a bare limerock substrate.

A PSTA established on a bare limerock surface will accumulate new marl sediments over time (thereby covering the limerock). It is unknown how this marl accumulation will influence performance. Currently, data is lacking on the accrual of new sediments in the PSTA Cell. This study will provide a spatial assessment of accrued sediment depth, an estimate of sediment P storage within the PSTA Cell, and chemical characterization of the sedimentary material, which will define a basis for comparison to existing data from muck-based STA cells, and to further investigations in the PSTA Cell over time. In addition, the effect of accrued PSTA sediment versus exposed limerock on outflow P concentrations will be evaluated in a flow-through column study. The comparison will be made by operating columns with limerock substrate with or without accrued sediments from the PSTA Cell. Low-P waters will be continuously fed to each column, and *Chara* and periphyton will be inoculated into each column. The overlying water column will be monitored weekly for TP concentration and enzyme activity and bi-weekly for P species. If newly-accrued sediments from the PSTA cell provide similar removal performance to exposed limerock, then sediment removal may not be required to sustain performance over time. Efforts for this study were initiated in early 2012 and are scheduled to continue through 2014.

Hypothesis #8: Periodic drawdown (and subsequent reflooding) will benefit PSTA systems through the consolidation of accrued sediments and the reduction of submerged macrophyte density.

Drawdown/Dry-out

Over the years, the STAs have been subjected to severe drought conditions as a result of climate variations. These drought conditions have been detrimental to the health of some EAV and SAV communities. Previous studies by other researchers, however, indicate that desiccated periphyton can recover immediately after rehydration (Thomas et al., 2006; Gottlieb et al., 2005). This component will aim to assess the effects of drawdown/dry-out on the periphyton community in the PSTA Cell and to assess the effect of rewetting after short and prolonged desiccation of the periphyton mats. Because the ground elevation of the PSTA Cell is lower than all of the surrounding areas, it is anticipated that a complete dry-out may be difficult and costly in terms of pumping costs. For this reason, it is anticipated that this component, if implemented at the field-scale, will be conducted opportunistically toward the end of the PSTA study period, and if environmental and hydrological conditions are conducive.

4.5.2 Support of Feasibility Analysis

At or near the completion of this research effort a Feasibility Analysis will be provided on the full-scale implementation of the PSTA concept in one or more STA flow paths. Results of studies included in the current research plan that are designed to address the key questions presented earlier, will provide critical background information in support of this analysis. Based on findings to date, the PSTA systems appear very similar to SAV communities, with a few important differences. First, although the STA-3/4 PSTA system was constructed on a wetland parcel from which muck was removed to expose a limerock base, it is unclear whether this approach is requisite for achieving ultra-low TP outflow concentrations. The first key question of this study plan (*What are the important design elements that enable the PSTA Cell to achieve ultra-low outflow TP levels? e.g., muck removal*) applies directly to the following feasibility questions:

1) Is there indeed a requirement for muck removal, either partial or complete?
2) In selected STA outflow regions, particularly those underlain by deep muck, can a limerock cap serve as an alternative to muck removal?

At present, it is thought that establishing a healthy periphyton community is required to attain low-level P removal performance in PSTA systems. To date, the range of suitable inflow P loading rates under which PSTA communities thrive, and the relationships between outflow P concentrations and P loading rates, are unclear. The second question of this study plan (*What are the key operational ranges that enable the PSTA Cell to achieve ultra-low outflow TP levels? e.g., P Loads, HLR, and water depth*) applies to the following feasibility questions:

1) What is the sustainable P removal rate (k value) for PSTA systems?
2) Under what inflow P loading rates is this P removal performance achievable/sustainable?
3) Is PSTA P removal performance adversely affected by hydraulic pulses, such as those experienced by the full-scale flow paths?
4) What is the required operational water depth range for achieving the desired P removal performance?

From prior research conducted in the Water Conservation Areas and the Everglades National Park, it is anticipated that appropriate water depth conditions, which may include periodic drawdown, are required to support/sustain desirable periphyton communities. The third question of the study plan (*What management practices are required to sustain the PSTA Cell's good performance?*) applies to the following feasibility question:

1) Does allowance need to be made for periodic drawdown of the PSTA cell (while keeping the upstream SAV cell hydrated)?

Additional information will likely be required in support of the PSTA Feasibility Analysis. Including measurements of existing depth of muck (over limerock) at STA outflow regions where PSTA deployment is being considered, along with soil TP profiles (with depth) at these locations.

4.5.3 Data Management

This study will follow the overall data management protocol described in the Restoration Strategies Science Plan and the SFWMD's scientific data management SOP (SFWMD, 2012a). All data collected from this study will be loaded into the different SFWMD databases:

DBHYDRO – water quality data from the inflow and outflow structures; flow data; stage data

ERDP – all ecological data and groundwater well data collected by the SFWMD

MORPHO – all study files, including data deliverables, photos, and drawings

Data loading into the databases will be coordinated with the assigned data steward for the study. All data must be accompanied by clear metadata that would allow reconstruction and understanding of the datasets.

This study component follows the general QA/QC guidelines specified in the Restoration Strategies Science Plan and the SFWMD's Field and Laboratory Quality Manuals. The SFWMD's quality system is outlined in the Quality Management Plan (SFWMD, 2012c), and supported by the Field Sampling Quality Manual (SFWMD, 2011), Chemistry Laboratory Quality Manual (SFWMD, 2012b), and SFWMD Enterprise Scientific Data Management Policies and Procedures (SFWMD, 2007; 2009). The Study Plan Leader will review all data and ensure their accuracy, precision, and completeness prior to loading into the databases. Any data that does not meet the SFWMD's data validation criteria will be qualified using standard SFWMD/FDEP data qualifier codes.

4.5.4 Reporting

The PSTA Study Plan Leader will meet with the study team and contractors on a routine basis to help ensure that the study stays on time and on budget. Monthly reports will be required from each of the component leads and the contractors. The District will summarize the findings and status on a quarterly basis and provide quarterly progress report to management. Presentations about the study will also be delivered when requested, e.g., for the SFWMD's Science Plan team, Restoration Strategies workshops, and Long-term Plan public communication meetings. On an annual basis, the District will prepare a write-up for the SFER, which will include progress and findings that are available at the time of report preparation.

4.5.5 Study Schedule

Previous work done under PSTA study efforts prior to the Science Plan implementation are shown in **Figure 4-7**. This PSTA Study Plan also includes the implementation of various planned components and report deliverables (**Figure 4-7**). Preliminary findings from ongoing and completed efforts will be reviewed periodically to ensure that the data being collected are answering the project questions and determine if other operational conditions or environmental variables need to be studied in greater detail to address those questions. The timing of these Stop/Go decisions is estimated for specific tasks in the figure below.

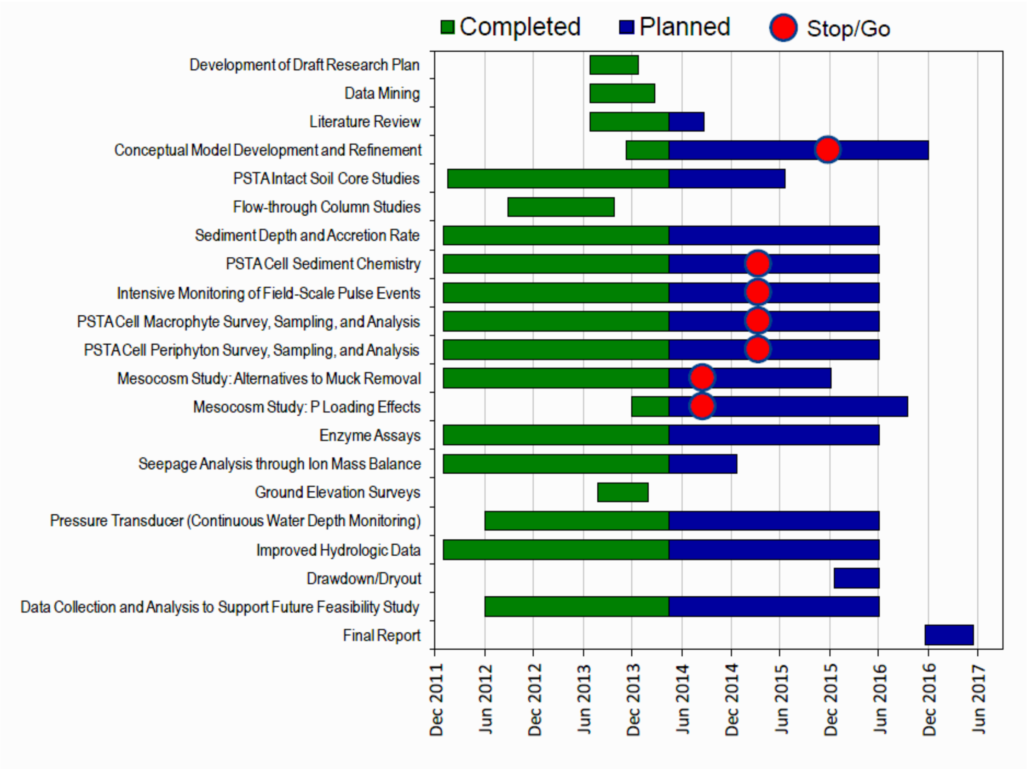

Figure 4-7. PSTA Study schedule through FY2017.

Planned Work:

• Conceptual model development	Continued FY2014	Complete FY2016
• Intact soil core studies	Continued FY2014	Complete FY2015
• Sediment depth & accretion rate studies	Continued FY2014	Complete FY2016
• PASTA cell sediment chemistry	Continued FY2014	Complete FY2016
• Monitoring of field scale pulse events	Continued FY2014	Complete FY2016
• Macrophyte survey& analysis	Continued FY2014	Complete FY2016
• Periphyton survey& analysis	Continued FY2014	Complete FY2016
• Alternatives to muck removal	Continued FY2014	Complete FY2015
• Evaluation of P loading effects	Continued FY2014	Complete FY2016
• Enzyme activity analysis	Continued FY2014	Complete FY2016
• Seepage analysis	Continued FY2014	Complete FY2015
• Analysis to support Feasibility study	Initiate FY2016	Complete FY2017

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APPENDIX A: REVIEW OF PRIOR PSTA RESEARCH EFFORTS

Review of Prior PSTA Research Efforts

The Florida Everglades ecosystem is a peat-based subtropical wetland system, and was characterized by extremely low levels of nutrients such as nitrogen (N) and P. Phosphorus concentrations in the water column of the least impacted Everglades are typically less than 10 ppb and soluble reactive P concentrations are often less than detection limits (Walker, 1999). As part of the Everglades restoration, six large Stormwater Treatment Areas (STAs) have been constructed to treat water coming from agricultural and urban areas to remove excess P before being discharge to the Water Conservation Areas (Chimney and Moustafa, 1999).

Considerable research has been done during the last 15 years to evaluate the capabilities of periphyton-based technology to reduce surface water P concentration. Algal-based treatment, as the final polishing step for agricultural runoff in the Everglades Agricultural Area, was first suggested in 1991 by the Technical Advisory Committee for the Everglades Nutrient Removal Project (Kadlec and Walker, 2003). In 1996, Doren and Jones suggested the acronym PSTA (Periphyton Stormwater Treatment Area) while observing periphyton colonization in association with low TP waters in the Hole in the Donut (HID) project in the Everglades National Park (ENP), where soil and vegetation have been removed down to the calcium-rich soil.

From 1998 to 2003 the SFWMD conducted research on potential advanced treatment technologies to support reduction of P loads in surface waters entering the Everglades (Chimney et al., 2000). Periphyton-based stormwater treatment was one of the supplemental technology concepts considered by the SFWMD for potential application downstream of the macrophyte-based STAs. As part of the Advance Treatment Technology (ATT) Program, the SFWMD initiated a PSTA study in the STA-1W test cells (Chimney et al., 2000). All work associated with the ATT program was completed in January 2002; however, the SFWMD continued monitoring of the three south PSTA cells to document long-term trends in TP removal by the PSTA technology. Substrate tested on these cells included 30 cm of shellrock over 30 cm of peat on two cells (shellrock cells) and only peat substrate on the third cell (peat cell), operated at a constant hydraulic rate of 2.3 cm/day. Results showed that the shellrock treatment outperformed the peat treatment. The overall outflow TP concentrations from the shellrock treatments were significantly reduced from 66 to 15 ppb from May 1, 2002 through April 30, 2006 (Goforth et al., 2005).

In 1998, DB Environmental, Inc. (DBE) used a series of raceways in which periphyton communities were established to reduce surface water P to lowest achievable levels. Shallow (9 cm deep), low-velocity raceways on a limerock substrate were located near the outflow of the ENR project, and were able to produce mean outflow TP concentrations of 10 ppb from an average inflow concentration of 17 ppb (DeBusk et al 2004). In contrast, higher-velocity raceways (2-cm deep) reduced TP concentrations from 17 to 14 ppb (DBEL, 1999).

In 1998, the SFWMD contracted CH2M HILL to evaluate viability, effectiveness and sustainability of PSTA technology at several scales of application (CH2M HILL. 2003a). Phase 1 and 2 were done in fiberglass tanks and three 0.2-hectare cells located in STA-1W. For Phase 3, four field-scale PSTA cells (20,790 m^2) were constructed at STA-2. Results reported that shellrock, sand, and limerock soils were the better options for the development of periphyton biomass. Peat substrate was found to encourage colonization of emergent vegetation and discourage periphyton growth. Inflow TP concentration was found to be important in determining the surface area needed for treatment by this technology. Outflow TP concentrations were reduced from 25 ppb (inflow) to concentrations of 11-15 ppb (outflow). Smaller mesocosms in

Phase 1 and 2 appeared to slightly overestimate removal rates, with a start-up period of 3 to 6 months required to reach stable performance.

In 1998, the SFWMD and the USACE cooperated in testing the PSTA technology. In 2003, the USACE conducted two parallel PSTA technology projects at the Flying Cow Road Test Facility (FCRTF) and at a Field Scale Demonstration (FSD) project in Cell 2 of STA-1E. At the FCRTF, four 10-ft by 100-ft concrete mesocosms were used to test different substrates. These mesocosms, which were monitored from 2006 to 2008, reduced P concentrations of from 25 ppb (inflow) to 9 ppb in Test Cell 3 which had 6-inches of Riviera sand overlaid by a 6-inch layer of local limestone, and to 17 ppb in Test Cell 1, which had a 12-inch layer of Riviera sand. The FSD consisted of three PSTA cells of 46.5 acres in size, with different substrates. Performance of these cells was limited by low flows and low inflow P concentrations for most of the operational period. Inflow P concentrations to the FSD PSTA cells averaged from 8 to 10 ppb, while outflow P concentrations averaged from 8 to 10 ppb. These results concluded that under controlled hydrologic conditions and depending on the inflow P concentration, PSTA can achieve long–term FWM outflow concentrations near 10 ppb. However, scale-up calculations based on the FCRTF project was not recommended due to data quality issues during the study (ANAMAR/WSI, 2011).

In 2002, the Village of Wellington evaluated two aquatic treatment trains, where PSTA cells were lined with 15 cm of limerock. The west side treatment reported average TP reductions from 25 ppb (inflow) to 21 ppb (outflow), while the east side system performance was variable because of extremely high inflow TP concentrations (mean of 118 ppb), achieving an mean outflow TP concentration of 46 ppb (CH2M HILL, 2003b).

History of Operations in the STA-3/4 PSTA Project

In 2004, the SFWMD began construction of the large-scale PSTA project in a portion of Cell 2B in STA-3/4. New levees were constructed to isolate a 400-acre portion in Cell 2 to form a system that included a 200-acre upstream SAV treatment cell, followed by two 100-acre cells, PSTA and Lower SAV. The difference between the downstream cells is that the peat substrate in the PSTA Cell was scraped down to the caprock and removed, while the sediment in the Lower SAV was not disturbed. Using survey data from 2003, the floor elevation of the PSTA Cell was estimated to be approximately 1.8 ft (54 cm) lower than the adjacent SAV cells. Removal of the peat substrate in the PSTA Cell was primarily done to eliminate growing medium for emergent vegetation and to remove a potential source of P to the water column (Goforth et al., 2005). However, as stated above, during construction, emergent vegetation strips were created within the PSTA Cell to improve hydraulic efficiency.).

Preliminary monitoring and start-up operations for the PSTA Project began in WY2006. The mean TP concentration measured at the outflow structure of the PSTA Cell for WY2006 was 15 ppb, however due to a regional drought there were no corresponding inflows or inflow data to compute treatment efficiency. A detailed operation plan for the PSTA Project was developed and implemented in WY2007, stipulating that the Upper and Lower SAV cells would be operated in association with the rest of STA-3/4 Cell 2. It was also stipulated that the PSTA Cell would be operated at an estimated hydraulic retention time (HRT) of 5 days, maintaining an average depth of approximately 60 ± 8 cm (again, based on the estimated average ground elevation using the 2003 survey data). A vegetation survey during 2007 revealed that SAV had become established throughout the PSTA project (Pietro et al., 2008). Difficulties and delays in completing the installation of telemetry equipment needed for remote operation of the PSTA project structures, as well as the continued regional drought delayed routine operation of the PSTA project until W2008.

Originally, the intent was to operate the PSTA and Lower SAV Cells in parallel; i.e., the cells would receive equal hydraulic loads to facilitate a comparison of the treatment efficiency of the PSTA versus SAV technologies. Unfortunately, this plan proved unworkable operationally and the two cells were operated differently. The SFWMD attempted to establish a prescribed HRT in the PSTA Cell, while the timing and quantity of flow into the Upper and Lower SAV cells was dictated by the operation of the remainder of Cell 2B. Discharge from the PSTA Cell, for the most part, was continuous, while outflow from the lower SAV Cell at G-379E was sporadic.

The inflow FWM TP concentrations to the PSTA Cell during the first four years of operation (WY2008 to WY2011) ranged from 14 to 27 ppb, while the outflow FWM TP concentrations ranged from 8 to 12 ppb (Piccone et al., 2013). During this period, large uncertainties in the hydraulic data hindered accurate calculations of HLR, PLR and P retention (mass load reduction and settling rate). Thus, in WY2012, structural, monitoring and operational changes were implemented in an effort to better assess and understand the PSTA Cell P loading rates and performance. At the same time, the level of effort for the PSTA cell was increased.

APPENDIX B: CONCEPTUAL MODEL

A conceptual model will be developed as a vehicle for integration of the information obtained from the literature review, ambient monitoring data, and results from the experimental research to provide a basis for developing an appropriate management model, a qualitative interpretative model, and a quantitative P mass balance model. The conceptual model, built as a graphic tool with one or more diagrams, describes the nutrient and biomass storages and transformations (**Figure B-1**).

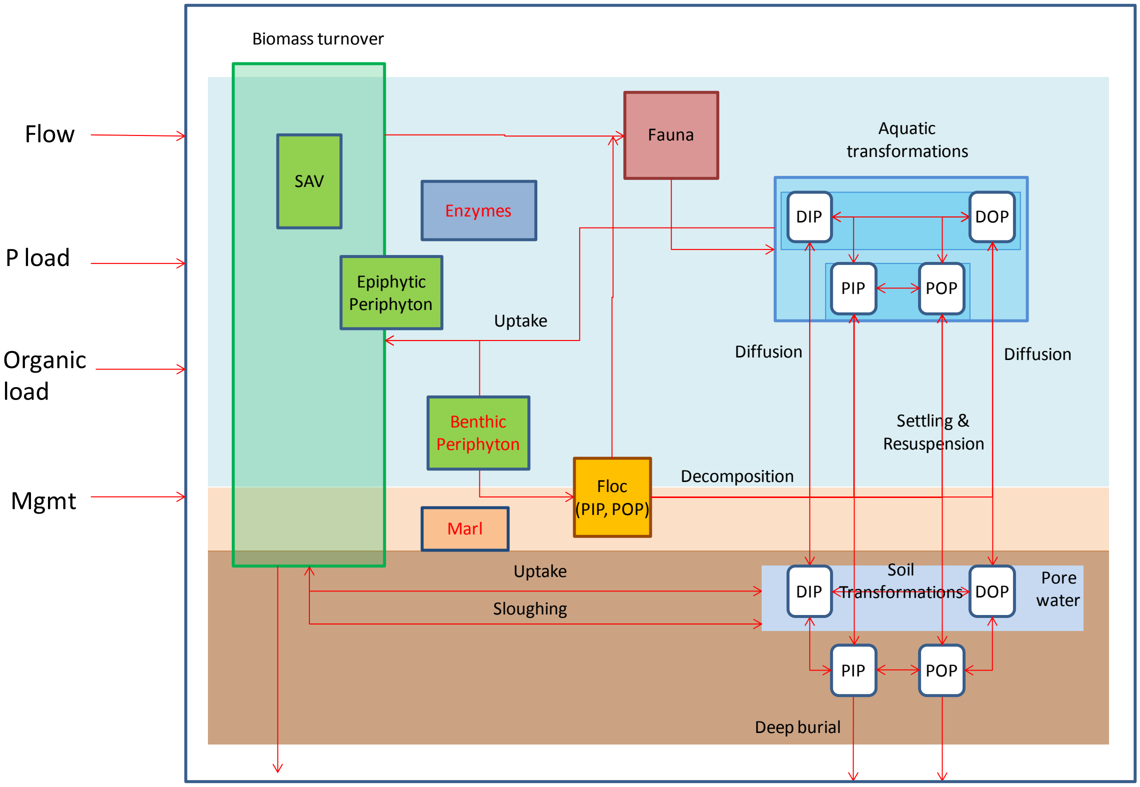

Figure B-1. Overview of conceptual model. [Note: Boxes depict the storages and arrows indicate flows/fluxes between the storages.]

APPENDIX C: MONITORING AND SURVEY TASK SUMMARY

Table C-1. PSTA Study monitoring, stations, frequency and parameters.

Monitoring		Sampling Locations	Sampling Frequency	Parameters
Surface Water Quality	Inflow and Outflow (Autosamplers and Grabs)	G390A, G390B, G388, G379D, and G381B	Biweekly	TP, TDP, SRP, NO3, NO4, TKN, TOC, DOC, Ca, Sulfate, chloride, total suspended solids, Fe? YSI (water temperature, DO, pH, and conductivity)
	Inflow/Outflow Diel Trends (RPA)	G390B, G388, G379D, and G381B	Recording at 3-hr interval	TP, TRP, Temperature
	Internal Gradient and Diel (Autosamplers)	Internal locations between inflow and outflow structures in PSTA	Event and seasonal driven deployment; recording at 3-hr interval	TP
	Internal gradients - Surface water (Grabs)	Internal transects multiple Locations in PSTA and adjacent Cells 2B and 3B	Baseline, then quarterly (covering wet/dry cycle), and or after extreme events	TP, TDP, SRP, NO3, NO4, TKN, TOC,DOC, Ca, Sulfate, chloride, total suspended solids
	Diel/Diurnal dynamics of P	G390B, G388, and Internal Locations in PSTA and adjacent Cells 2B and 3B	Recording at 15-min interval	TP, YSI/Hydrolab (DO, pH, temperature, conductivity, turbidity)
Groundwater	Seepage water quality monitoring	Wells along both sides of PSTA	Water depth: biweekly to monthly ; WQ: quarterly;	water depth, WQ (TP, TDP, SRP, and Major ions; Ca, Mg, Na, K, Cl, SO4, and Alkalinity)
Hydraulic monitoring and hydraulic loading rate analysis	Hydraulic/hydrology measurement	G390A, G390B, G388, Lower SAV, Cell 2B, and 3B	Hourly, daily, and weekly data recorded by the SFWMD	flow rate, stage, and rainfall
	Pressure Transducers (Continuous water depth monitoring)	Four internal locations within the PSTA Cell	Weekly - internal water depths recording at 15-min interval	internal water depths (in)
Sediments	Characterization - floc, soil, and pore water survey	Internal transects multiple locations in PSTA and adjacent Cells 2B and 3B	Baseline, biannually	floc depth, soil accrual, soil bulk density, AFDW, nutrients (TP, TN, TC), pore water
SAV and algae characterization	Characterization - macrophyte and SAV density	Internal transects multiple locations in PSTA and adjacent Cells 2B and 3B	Baseline, then quarterly (covering wet/dry cycle), and or after extreme events	% coverage, biomass, nutrients (AFDW, TP, TN, TC) by species
	Internal gradient periphyton characterization - algal	Internal transects multiple locations in PSTA and adjacent Cells 2B and 3B	Baseline, then quarterly (covering wet/dry season), and or after extreme events	% coverage, biomass, nutrients (AFDW, TP, TN, TC) by species
	Enzyme activity	Internal transects multiple locations in PSTA and adjacent Cells 2B and 3B	Baseline, quarterly, or after extreme events (before will cooperate with quarterly	monoesterase(APA), diesterase (DPA) for water, SAV, periphyton, and sediment)

SECTION 5:
EVALUATION OF THE INFLUENCE OF CANAL CONVEYANCE FEATURES ON STA AND FEB INFLOW AND OUTFLOW TP CONCENTRATIONS

5.1 OVERALL STUDY PLAN SUMMARY

The purpose of this study is to determine if total phosphorous (TP) concentrations or loads change when conveyed through STA inflow or outflow canals. And, if so, what factors influence the changes (e.g., How much sediment and TP have been accumulated in canals throughout the period of operation, and if remedial field work is needed to address this issue?).

5.2 BASIS FOR THE PROJECT

Key Science Plan Question Study Addresses

- Key Question 5: What operational or design refinements could be implemented at existing STAs and future features (i.e., STA expansions, FEBs) to improve and sustain STA treatment performance?(update from Science Plan, SFWMD, 2013)

Science Plan Sub-Question Study Addresses

- What changes in canal management or design improve STA and FEB performance?

The proposed study will be conducted in two phases. In Phase I, a review of existing water quality and flow data will be performed to answer the following questions.

- Do TP concentrations change when conveyed through STA inflow or outflow canals?
- If TP concentrations change along a STA canal, what are the factors that may be influencing the change?

Available water quality and flow data, mass balances for TP, total suspended solids, and other water quality data will be used to determine if each canal is behaving as a TP source or sink, and to provide a preliminary estimate of sediment and TP net accumulation in a canal during the period of operation. Canals that appear to be behaving as TP sources may be candidates for remedial measures, and canals that appear to be behaving as TP sinks may provide insight for future STA/FEB designs or operational strategies.

A summary report documenting the findings from the Phase I study with recommendations for Phase II will be provided. In Phase II, the following activities will be performed:

1. Conduct field investigations to validate the Phase I preliminary findings, i.e., is a canal behaving as a TP source or TP sink. If Phase I suggests that TP is accumulating in a canal, then the estimate of sediment and TP accumulated will be refined based on the field investigations. The field investigations may include sediment core sampling, canal cross section, and centerline profile surveys; laboratory analysis for sediment chemical and physical parameters, etc.;
2. Conduct hydrodynamic modeling to simulate sediment transport and identify optimized operational factors, such as flow rates and stages;

3. Provide recommendations for optimized structural operation and design to improve STA performance based on the findings of this study; and
4. Provide recommendations for potential remedial field work in canals behaving as TP source (e.g., vegetation harvesting, sediment traps, routine maintenance, dredging).

5.3 BACKGROUND/LITERATURE REVIEW

To address water quality concerns associated with existing flows to the Everglades Protection Area (EPA), the South Florida Water Management District (SFWMD or District), Florida Department of Environmental Protection (FDEP), and United States Environmental Protection Agency (USEPA) engaged in technical discussions starting in 2010. The primary objectives were to establish a Water Quality Based Effluent Limit (WQBEL) that would achieve compliance with the State of Florida's numeric phosphorus criterion in the EPA and to identify a suite of additional water quality projects to work in conjunction with the existing Everglades Stormwater Treatment Areas (STAs) to meet the WQBEL.

The Science Plan is being implemented to investigate critical factors that influence phosphorus treatment performance. The Science Plan has been designed to increase the understanding of factors that affect treatment performance; in particular factors that affect performance at low phosphorus concentrations (<20 ppb TP). Results from these studies will be used to inform design and operations of treatment projects which will ultimately improve capabilities to manage P in STAs for achievement of the WQBEL.

As part of the Science Plan, one key area to be examined is how the different FEBs (shallow and deep) can be designed to optimize settling of suspended material, including particulate phosphorus (PP), to prevent their transport into the receiving STA. Such information will be helpful in determining if the design of FEBs should include a settling basin with maintenance access for sediment (and associated PP) removal. Sediment sumps upstream of discharge structures might allow settling and more efficient dredging. For example, the sediment trap recently completed along the C-51 canal that discharges to the Lake Worth Lagoon has been working well (Palm Beach County, 2010). To minimize erosion and thereby further reduce P transport into the STAs, the design of FEB discharge features (e.g., use of weirs) should also be considered. Another potential source of P to the STAs may be from the conveyance canals that extend from the FEBs to the STAs because of potential settling and resuspension of phosphorus-containing particles or release of soluble P due to changes in sediment redox potential. As such, it is pertinent to determine if these conveyance canals, along with the canals entering the FEBs, should be periodically dredged or lined to remove sediment and associated PP. Preliminary information is available regarding the potential for canal sediment management for P removal. A summary of phosphorus speciation data in canals at 11 structures upstream of the STAs from 1974–2012 indicates that PP was approximately 38 percent of TP. A study on particle size distribution and P fluxes at three structures in the EAA indicates that PP ranged from approximately 10-35 percent (Ivanoff, 1993). Collectively, these data suggest that there may be P settling in canals at these locations. It is also important to note that for peat-based farmlands in South Florida, the majority of runoff PP has been found to originate in the drainage canals as a result of biological growth (Kadlec and Wallace, 2009). To better understand the factors contributing to relatively high STA-5 inflow TP concentrations compared to the other STAs, the District conducted a canal sediment study in the STA-5 inflow canal in 2007. Based on these results, in 2008 a $1.79 million canal sediment dredging project was implemented in the L-3 canal immediately upstream of the STA-5 inflow structures. Following the L-3 canal dredging project, a decrease of inflow TP concentrations has been observed, although other factors could also be contributing to these reductions such as BMP improvements in the C-139 Basin.

Surface water TP concentrations have been observed to change from STA inflow pump stations to the inflow structures at the upstream side of the flow-ways. There is also some evidence that TP concentrations may change as water moves from the treatment cell flow-ways to the STA permit compliance discharge structures. For example, along the STA-1E Discharge Canal, TP concentration data measured at S362 always differ from the TP concentration data measured at S369B. Total suspended solid (TSS) is a component of stormwater and is present in STA inflow and outflow canals. Particulate phosphorus may adhere to suspended solids and settle in these canals. High velocities can induce sediment re-suspension resulting in elevated TP in inflow water and/or elevated TP in the outflow collection canals. During severe droughts, water levels in some canals are significantly lowered to the extent that portions of the canal sediments are exposed for periods of time. When re-wetted, the effects of sediment P flux to the overlying water column could also influence the water TP concentrations observed at the inflow and outflow structure sampling locations. Stagnant canal segments may allow excessive phytoplankton growth and settling of organic material that decomposes and removes dissolved oxygen. Low DO at sediment surface would trigger release of soluble P. Seepage of water into or out of STA canals to or from adjacent water bodies might also be a contributing factor in changes in surface water TP concentration. All of these factors may contribute to the TP concentration change along a canal.

Overall, it is important to assess whether the sediments in the canals are an important source of TP to the STAs by characterizing the sediment and TP concentrations in canals flowing to the FEBs and the STAs and determining the factors affecting settling and resuspension in these canal reaches. **Figure 5-1** is a simple diagram showing the potential contributing sources to STA inflow and outflow canals. The inflow and outflow components are generally assumed to include structure flows and seepage flows. For the canals included in this study the initial focus will include structure flows unless there have been historic observations pointing to seepage as likely significant contributing sources. As work on each canal proceeds, seepage estimation (modeling) can be added on an as needed basis.

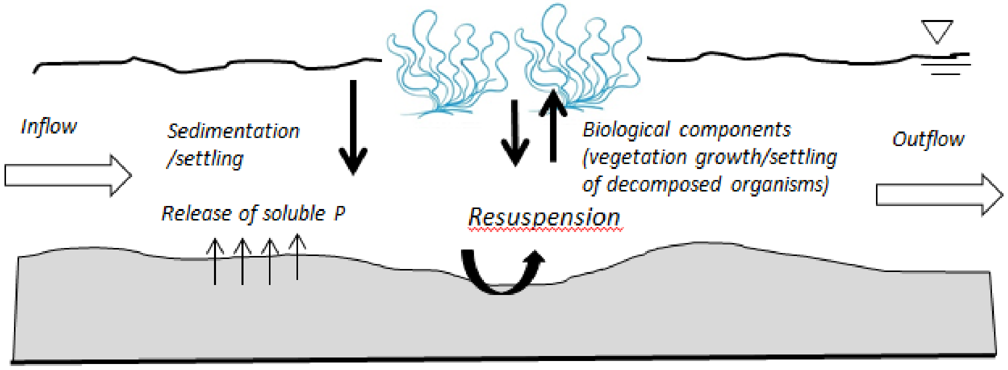

Figure 5-1. Schematic of different P forms transported in a canal.

5.4 STUDY PLAN OBJECTIVES

In this study, the hypotheses to be tested are as follows:

1. Physico-chemical characteristics of canal sediments influence TP concentrations at inflow and outflow waters from the STAs and FEBs.
2. TP concentration in STA or FEB treated water can increase via sediment resuspension or P flux along the outflow collection canal and at the outflow structure.
3. Resuspension and transport of particulates and TP at inflow and outflow canal waters during flow events may affect STA or FEB performance.
4. For STA-2 and STA-1E, seepage to or from adjacent water bodies or groundwater is conveying additional TP to or away from inflow and outflow canals.

The objectives of Phase I are to determine if TP concentrations change when conveyed through STA inflow or outflow canals, and if TP concentrations change, to determine the factors that may be influencing the change. After Phase I is complete, the results will be used to refine the proposed Phase II activities. The objectives of Phase II are to (1) collect field data and conduct detailed analyses necessary to validate Phase I results, and (2) provide recommendations for remedial work to address the impacts or provide recommendations for future STA/FEB canal designs or operations.

Phase I will involve desktop data mining and analysis. Available water quality and flow data, mass balances at different time scales for TP, total suspended solids, and other water quality data will be used to determine if each canal is behaving as a TP source or a TP sink, and to provide a preliminary estimate of sediment and TP accumulated in a canal during the period of operation. **Figure 5-1** is a simple diagram showing the major components to be investigated in this study. The inflow and outflow components are generally assumed to include structure flows and seepage flows. For the canals included in this study, the initial focus will include structure flows unless there have been historic observations pointing to seepage as likely significant contributing sources. As work on each canal proceeds, seepage estimation (modeling) can be added on an as needed basis. The biological factors and activities may also affect nutrient and sediment process in a canal. These biological components will not be investigated in the Phase I study. During Phase II, the biological impacts will be further evaluated by conducting field investigations, soil core sampling, and laboratory analysis of sediments chemical and physical characterizations.

The findings from Phase I will be compiled in a summary report that will include recommendations for Phase II. A flow chart is provided in **Figure 5-2**. If the Phase I results suggest a canal is behaving as a TP sink, the Phase II investigations will be focused on confirming the Phase I results and determining the factors that cause the canal to behave as a TP sink. If the Phase I results suggest a canal is behaving as a TP source, then the Phase II investigations will be focused on confirming the Phase I results, and determining the factors that cause the canal to behave as a TP source. Phase II activities will generally include a cross-section and centerline field survey, sediment core sampling, and laboratory analysis for sediment sample chemical and physical characterizations. Sediment chemical and physical characterizations will provide basis for evaluating the biological factors and activities affecting canal water quality and quantifying the mobile solids in a canal. If additional data collection is needed, then this can be defined after Phase I is completed for inclusion in Phase II. For example, sediment traps or turbidity probes could be installed, etc. Hydrodynamic modeling to simulate sediment and nutrient transport is also planned for Phase II. The data needed for the modeling work will be evaluated as work proceeds.

Phase II activities are planned to include the following:

1. Conduct canal cross section and centerline profile survey, canal vegetation coverage survey, and sediment core sampling.
2. Laboratory analysis of sediment sample chemical and physical characterizations.
3. Refine the estimate of sediment and TP accumulated in the studied canals based on the field investigations.
4. Conduct hydrodynamic modeling to simulate sediment transport and identify the optimized operational factors;
5. Provide recommendations for optimized structural operation and design to improve STA performance; and
6. Provide recommendations for remedial work (e.g., vegetation harvesting, sediment traps, routine maintenance, dredging).

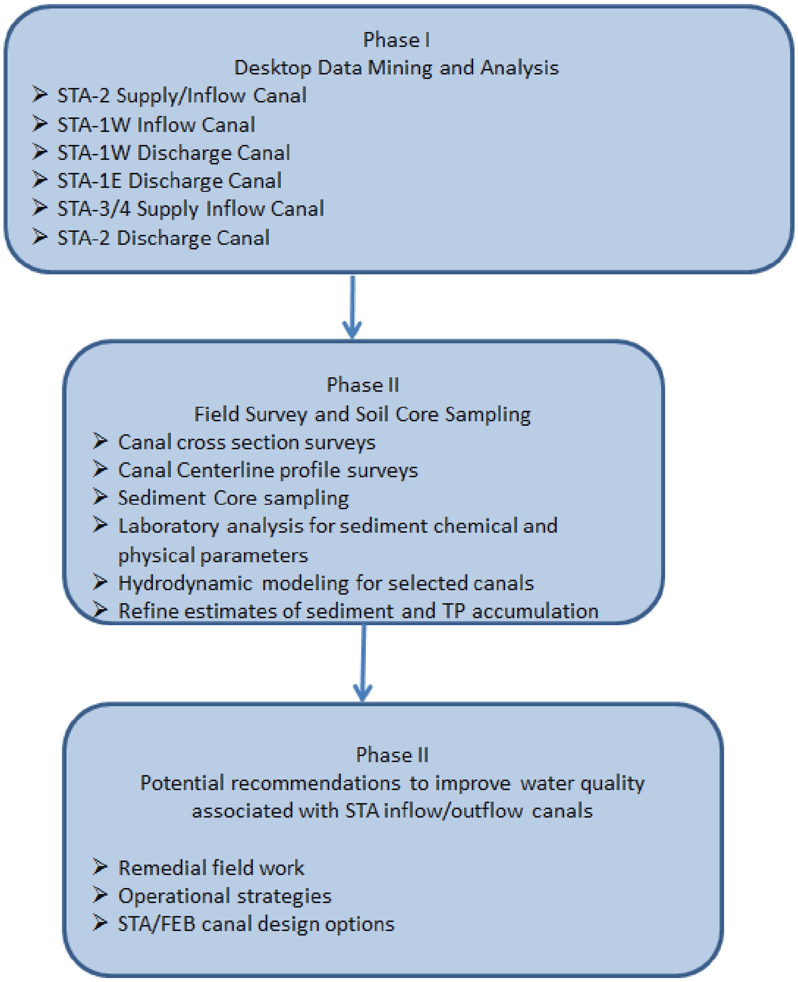

Figure 5-2. Canal study flow chart.

5.5 DETAILED STUDY PLAN

5.5.1 Study Plan Description

An overall location map of the Everglades STAs is presented in **Figure 5-3**. In this study, canals identified for investigation during Phase I are listed below. The canals to be studied under Phase II will be based on the recommendations from Phase I. The discharge canals for STA-3/4 and STA-5/6 are not included in this study because the canal compliance sites and the flow-way outflow structures are identical.

- STA-1E outflow canal (**Figure 5-4**)
- STA-1W outflow canal (**Figure 5-5**)
- STA-1W Inflow Canal between S-5A to G-302 (**Figure 5-6**)
- STA-2 supply/inflow canal (**Figure 5-7**)
- STA-2 outflow canal (**Figure 5-8**)
- STA-3/4 inflow canal (**Figure 5-9**)

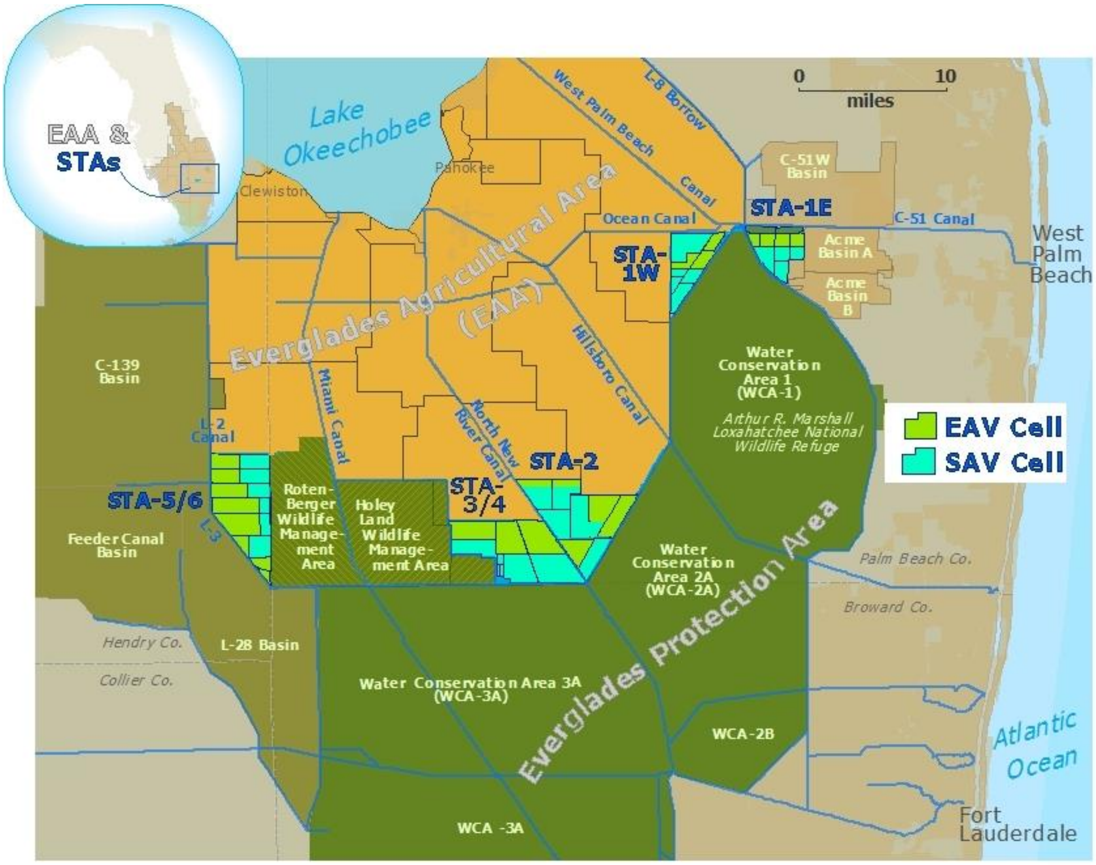

Figure 5-3. Location of the Everglades STAs.

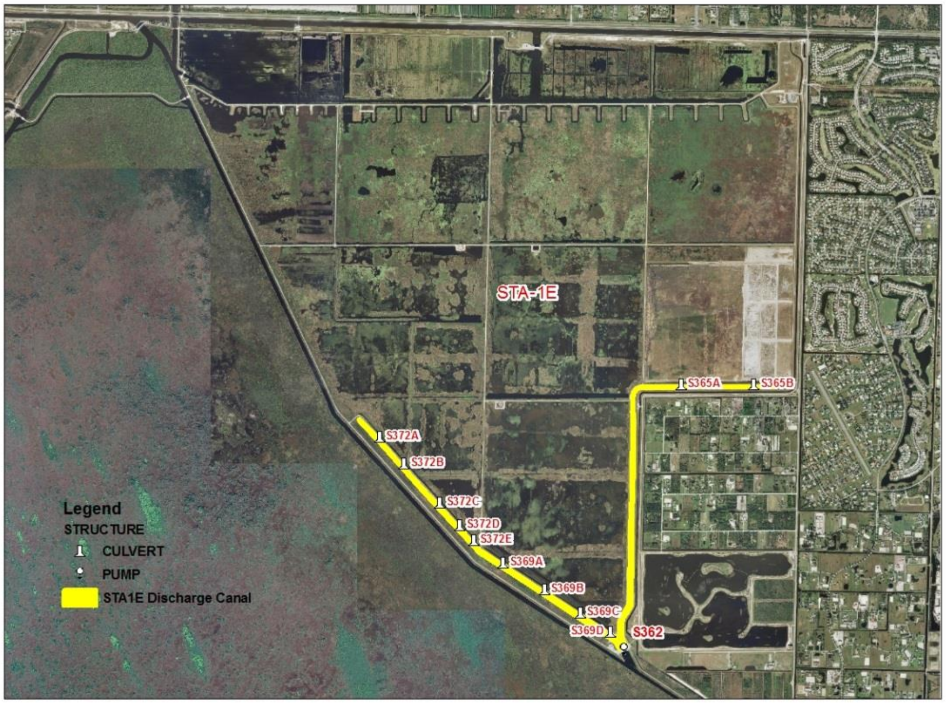

Figure 5-4. STA-1E discharge canal.

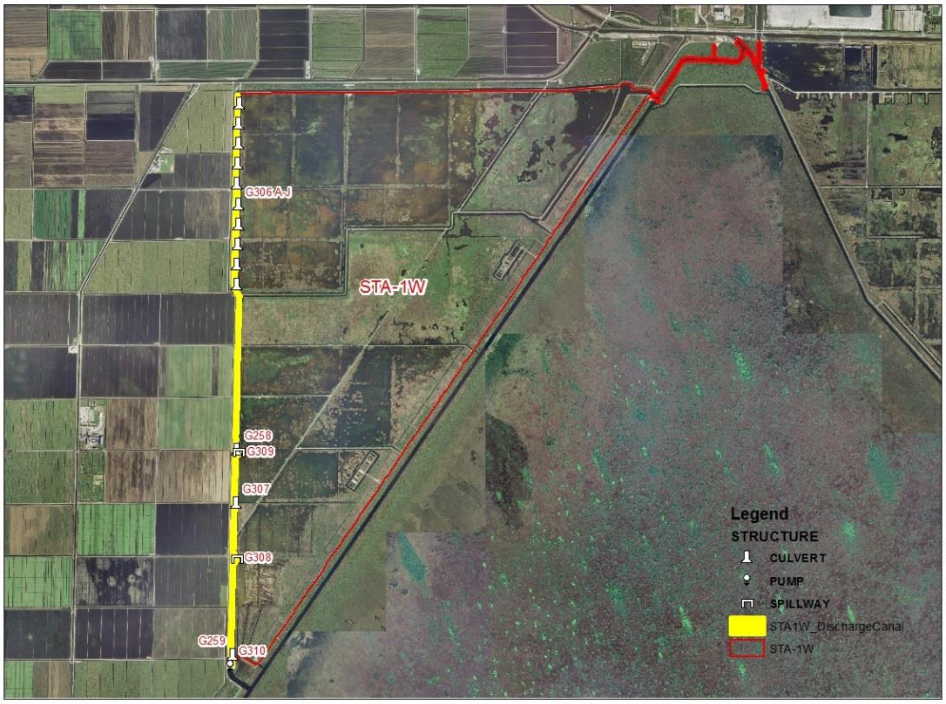

Figure 5-5. STA-1W discharge canal.

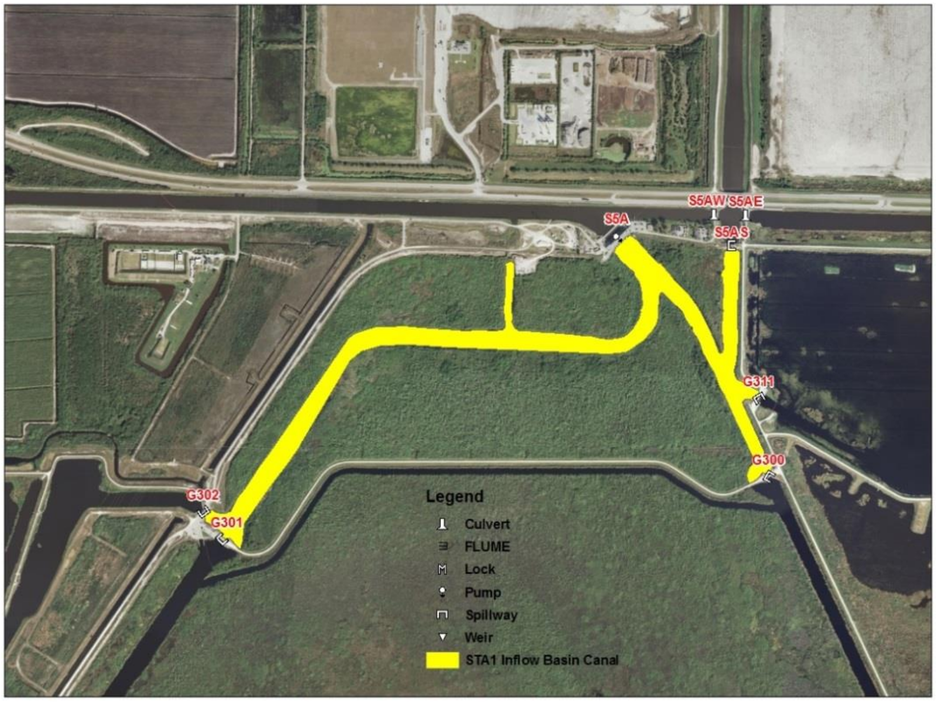

Figure 5-6. STA-1W canal segment between S-5A and G-302.

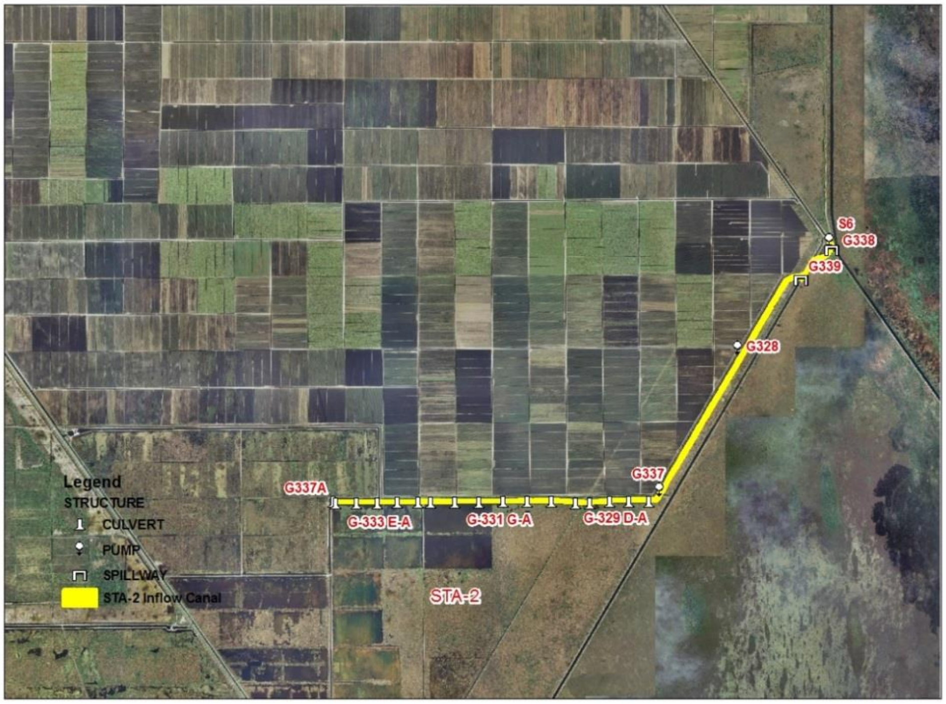

Figure 5-7. STA-2 supply/inflow canal.

Figure 5-8. STA-2 discharge canal.

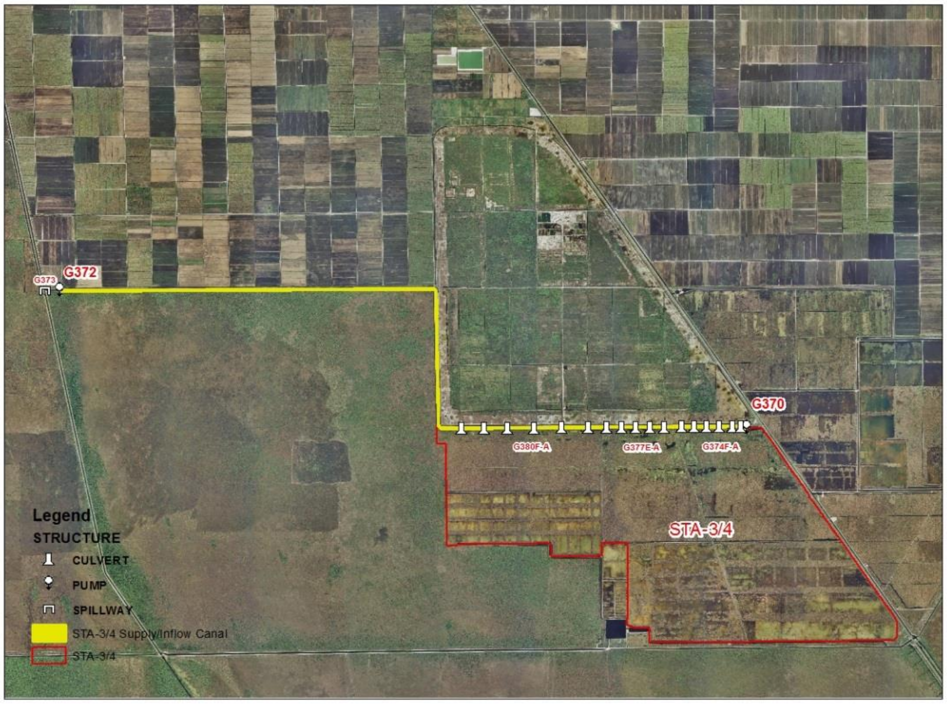

Figure 5-9. STA-3/4 supply/inflow canal.

5.5.2 Study Plan Components

Phase I

<u>Task 1: Literature Review, As-built Drawing Review and Data Query</u>

<u>Literature Review</u>

A literature review will be conducted to understand how previous canal sediment studies were conducted, what types of analytical approaches were used, and what types of results were obtained. The papers should be related to canal conveyance, suspended sediment transport, nutrient transport, nutrient sediment reflux, nutrient sediment sequestration, etc. Technical reports related to this study are also appropriate documents for review. A short summary shall be provided for each document reviewed. Information gained from the literature review will be used to inform the analytical methods (statistical analysis, modeling approaches) of the study tasks.

<u>As-built Drawings, Design Drawing Review and Field Observations</u>

For all the canals evaluated this study, the as-built drawings, related design document, and the related Operation Plan shall be reviewed. Field observations will also be made to obtain general visual information such as apparent turbidity, vegetation material in canal segments to be studied. A technical summary shall be prepared for each canal. The technical summary shall include but not limited to:

- General description and design intend
- Conveyance capacity
- Dimensions of canal cross sections at different locations (include side slope, bottom width, levee top elevation, etc.)
- Canal centerline profiles
- Other critical physical features and related configurations

This information will be used in Tasks 3 and 6 in estimation of accumulated sediment and velocities. The cross-sections data will also be used in development of the seepage modeling and the hydrodynamic modeling to be included in Phase II of the study.

<u>Data Query by the Nutrient Load Program and Preliminary Review</u>

For each canal to be investigated in this study, subject to the data availability, period of record (POR) data beginning with the first complete water year and ending at WY2013 will be analyzed. For all the structures related to the canals included in this study (**Appendix A**), flow data and the water quality data related to the parameters as summarized in **Table 5-1** will be retrieved from the District's databases by the Nutrient Load Program—a tool developed by the District. This program provides nutrient load values according to a user-configurable interface. Preliminary data review will be conducted and outliers will be eliminated from further analysis based on the best professional judgment.

Table 5-1. Parameters to be queried by the Nutrient Load Program.

Parameter	Unit	Test ID
dissolved chloride (CLD)	µg/L	32
turbidity (TURB)	NTU	12
phosphate, dissolved as P (TDP)	µg/L	26
phosphate, ORTHO as P (SRP)	µg/L	23
phosphate, total as P (TP)	µg/L	25
total suspended solids (TSS)	mg/L	16
flow	cfs	---
conductivity	µs/cm	9

Rainfall Data Query

Both rain gauge-based and Nexrad (radar)-based rainfall data for POR beginning with the first complete water year and ending at WY2013 will be queried and reviewed. The rainfall data will provide the basis for identifying different storm events for further investigation. Current active rain gauges are summarized in **Table 5-2**.

Table 5-2. Active rain gauges within the study area.

STA-1W and STA-1E			
Station	**DBKEY**	**Remark**	**Preferred DBKEY***
ENR101	15851		no
ENR203	15874		no
ENR301	15877		no
ENR308	15888		no
ENR401	15862		no
Average	KN809	Thiessen weighted average of available gauges	yes
STA-2			
Station	**DBKEY**	**Remark**	
G331D	PT420		no
STA-3/4			
Station	**DBKEY**	**Remark**	
S7	15204		yes
EAA5	15184	PREF DBKEY JW233 discontinued in 2010	no

Task 2: Water Quality Concentration Data Variability and Trend Analyses

The purpose of this task is to evaluate if the concentration data measured at different locations along a canal vary and if the concentration difference contains any trend throughout the POR (see **Figure 5-10** as an example). The results of this task will provide preliminary results regarding the source/sink condition in the canal. Due to the different sampling frequencies and methods, only well-matched (e.g., collected within the same approximate 2-3 hour period) grab samples will be used in this analysis. This task will focus on water quality concentration data of TP, TDP, DOP, SRP, TSS, and CLD.

The proposed approach for this task is testing for statistically significant differences between pairs of locations. This type of analysis will be applied to all the appropriate parameters.

Figure 5-10. STA-2 inflow/supply canal.

The time series of concentration difference will also be evaluated for trend throughout the POR. The purpose of the trend analysis is to examine if the sediment and TP settling and resuspension status changes over the time.

Task 3: Seepage Flow Rate Modeling

To quantitatively estimate daily seepage flow into and out of STA-2 Inflow/Supply Canal for the POR, a three-dimensional transient groundwater flow model will be developed using the U.S. Geological Survey's MODFLOW (McDonald and Harbaugh, 2000). The groundwater model will cover all STA-2 and major canals and drains around STA-2. Flow exchanges between different hydraulic elements (i.e., STA-2, canals) in the model domain are modeled since they are hydraulically connected. Simulated daily flow rates in and out of the STA-2 Inflow/Supply Canal will be used for mass balance analysis. A modeling report that documents data, assumptions and results will be submitted for review. The review comments will be carefully incorporated into the model and the report accordingly. The daily seepage values predicted by this model will be used in Tasks 4 and 5 for the mass balance computations.

Task 4: Canal Sediment and TP Accumulation Estimate Based on Annual/Monthly/ Wet Season/Dry Season Mass Balance

This task will provide an annual, monthly, wet/dry season mass balance for the parameters listed in **Table 5-3**. Mass balance will be conducted using different concentration interpolation modes within the Nutrient Load Program as appropriate. The load and sediment estimates based on the different calculation modes by the Nutrient Load Program will be evaluated for appropriateness and reasonability. At a preliminary level, a canal will be evaluated from the following perspectives:

- Is the canal a TP source or sink?
- What is the sediment accrual status in a canal (i.e., qualitative assessment)?
- How much sediment (in mt) has been accumulated in a canal during the POR based upon mass balance and direct measurement at the inflow and outflow structures?
- How much TP (in kg or mt) has been accumulated in a canal during the POR?

Table 5-3. Parameters and scenarios for mass balance analysis.

Annual	Monthly	High Flow Event	Medium Flow Event	Low Flow Event
flow mass balance				
TP mass balance				
TSS mass balance				
CLD mass balance				
DOP mass balance				
TDP mass balance				
SRP mass balance				

Conducting TP and flow mass balance is an essential step for this study. It provides a direct estimate of the TP removed from or accumulated in a canal. Dissolved chloride (CLD) acts like a conservative tracer. CLD mass balance allows checking on the accuracy of the water budget. Total suspended solid (TSS) mass balance will give a direct estimate of sediments that have accumulated in a canal. Mass balance for various P fractions will help us understand the P transformation and physical movement features**.** For example, increased particle P (PP) concentration from upstream to downstream under high velocity condition may indicate canal scouring or increased resuspension. Reduction in soluble reactive phosphorus (SRP) may indicate it is being taken up by plants and converted into tissue phosphorus or may become absorbed by wetland soil and sediments. A mass balance of the total dissolved phosphorus (TDP) in the system will provide an indication of any sorption of soluble P by suspended organic material in the canal water column or interaction of this P fraction with soil and vegetation in a canal. Mass balance of dissolved organic phosphorus (DOP) in STA canals will provide important information if this organic P fraction undergoes any microbial or enzymatic transformation in these canals.

Task 5: Flow Event-Based Mass Balance for Different Parameters

This task will start with review of rainfall data and corresponding STA inflow events. Inflow events of varying sizes will be identified. Water quality and flow data related with the identified events will be isolated for further analysis. An example event is provided by the chart below (**Figure 5-11**).

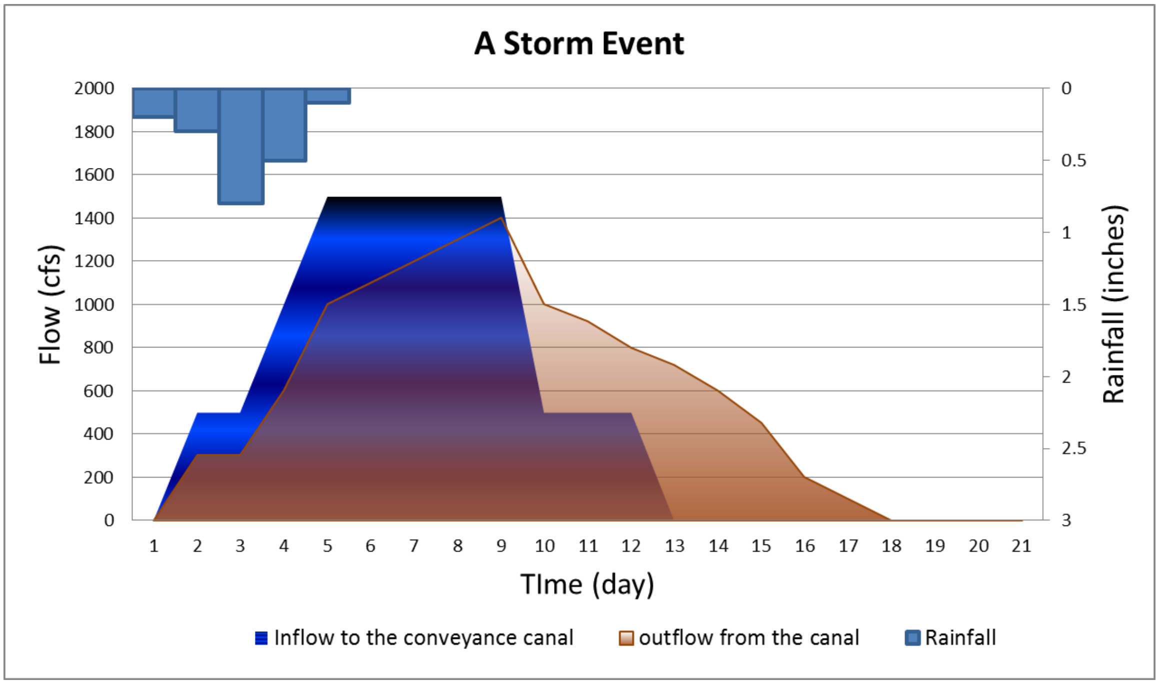

Figure 5-11. An example of a flow event.

The event analyses will provide additional information to evaluate the settling and resuspension status of sediment, TP, SRP and other p speciation s and the source/sink status of a canal. Periods with no inflow and no outflow will also be analyzed. Settling is a physical process where solids sink in water due to the density difference between the particular and water; Resuspension is a movement of particles from the sediments to overlying water (Kadlec and Wallace, 2009).

Events from three inflow scenarios will be selected for analyses. These three scenarios are when the canal inflow structures are operated at full capacity, medium capacity, and low capacity. This study may also evaluate the water quality data at different locations when the structures are closed and the water quality data are available. Different inflow scenarios (i.e., flow-rate based) can be from different flow events or different phases of a flow event. For each inflow scenario, multiple events will be analyzed. Breakpoint flow data may be needed for this analysis. The major named storm events, like Tropical Storm Isaac, should be analyzed. The parameters to be included in the flow event based mass balance analyses are summarized in **Table 5-3**.

For major pump stations (S6, S362, G310, G370 and G372, etc.), historical flow data will be analyzed to summarize the frequency distribution of different pumping capacities.

Each canal will be evaluated as follows:

1. Evaluate settling and resuspension status under different scenarios.

2. Compare event based mass balance results against the results from annual/monthly mass balance for the same period. If needed, refine the estimate of the TP (in kg or mt) accumulated in a canal during the POR.
3. Compare event based mass balance results against the results from annual/monthly mass balance for the same period. If needed, refine the estimate of the sediment (in mt) accumulated in a canal during the POR.

Task 6: Analyses of the Potential Influencing Factors Related to P Concentration Changes

This task involves an evaluation of the potential influencing factors related to changes in phosphorus concentrations (both TP and speciation) at different sample locations. Some example factors are velocity and canal stage. Different approaches such as dimensional analysis and statistical correlation analyses will be explored.

The flow data, water quality data, and results obtained in Task 5 will be used for this task when appropriate. Further data mining will be conducted to increase the number of samples. Data used in the analyses for this task include but not limit to breakpoint flow data, canal cross sections, stage, and grab sample water quality data, etc. The flow/velocity data will be paired with water quality data carefully. Flow data at major pump stations located in the studied canals will be analyzed to quantify the relationships between pumping capacities and frequencies.

Task 7: Recommendations for Phase II Work

The findings from Phase I will be compiled in a summary report that will include recommendations for Phase II. A flow chart is provided in Figure 2. If the Phase I results suggest a canal is behaving as a TP sink, the Phase II investigations will be focused on confirming the Phase I results and determining the factors that cause the canal to behave as a TP sink. If the Phase I results suggest a canal is behaving as a TP source, the Phase II investigations will be focused on confirming the Phase I results, and determining the factors that cause the canal to behave as a TP source. Phase II activities will generally include a cross-section and centerline field survey, sediment core sampling, and laboratory analysis for sediment sample chemical and physical characterizations. Sediment chemical and physical characterizations will provide basis for evaluating the biological factors and activities affecting canal water quality and quantifying the mobile solids in a canal. If additional data collection is needed, it can be defined after Phase I is complete for inclusion in Phase II. For example, sediment traps or turbidity probes could be installed, etc. Hydrodynamic modeling to simulate sediment and nutrient transport is also planned for Phase II. The data needed for the modeling work will be evaluated as work proceeds.

Phase II will also include, but not be limited to, the following items:

- Sediment Core Sampling: Sampling spatial interval, sample locations, samples need to be collected at each location.
- Parameters for Laboratory Analysis: The parameters need to be analyzed for chemical characterization; the parameters need to be analyzed for physical characterization.
- Survey: Cross-section survey spatial frequency and approximate locations, survey limits, and continuous canal centerline profile.

Task 8: Phase I Final Report

A draft report will be developed to summarize all the work conducted for Phase I of the study. The report will include study background information; literature reviewed; summary of the data, analyses, and modeling work performed in this study; conclusions; and recommendations for Phase II.

Phase II

The results of Phase I will be used to define the details of Phase II. The preliminary proposed tasks for Phase II of the study are as follows.

- Task 1: Canal cross section and profile survey
- Task 2: Sediment soil core sampling
- Task 3: Laboratory analyses of sediment chemical and physical characterization
- Task 4: Refine the estimate of sediment and TP accumulated in the selected canal based on the field and laboratory data
- Task 5: Hydrodynamic modeling to simulate sediment transport and identify the optimized operational factors
- Task 6: Recommendations of remedial field work
- Task 7: Final report and presentation

5.6 STUDY SCHEDULE

• Phase I	Initiate FY2013	Complete FY2016
• Phase II	Initiate FY2015	Complete FY2017

5.7 LITERATURE CITED

Harbaugh A.W., M.C. Hill and M.G. McDonald, 2000. MODFLOW-2000, the U.S. Geological Survey modular ground-water model — User guide to modularization concepts and the Ground-Water Flow Process. Open-File Report 00-92. U.S. Geological Survey.

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Palm Beach County. 2010. C-51 Canal Muck Dredging, Palm Beach County Sediment Management Project. Fact Sheet, February 2010.

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APPENDIX A: STUDY STRUCTURES, DBKEYS AND WATER QUALITY SITES

Structures		Preferred DBKEYS	Source DBKEYS	Water Quality Sites
STA-1E				
Outflow	S362	TP369	T0897	S362
STA-1E, Eastern Flow-way				
Outflow	S365A	W3904	SG561	S365A
	S365B	W3905	SG563	S365B
STA-1E, Central Flow-way				
Outflow	S369A	W3911	TA355	S369B
	S369B	W3912	TA356	S369B
	S369C	W3913	TA318	S369C
	S369D	W3914	TA357	S369C
STA-1E, Western Flow-way				
Outflow	S372A	W3918	TN560	S372B
	S372B	W3919	TY236	S372B
	S372C	W3920	TA330	S372B
	S372D	W3921	TN561	S372D
	S372E	W3922	TY238	S372D

Structures		Preferred DBKEYS	Source DBKEYS	Water Quality Sites
STA-1W				
Inflow	G302	JW221	JJ806	G302
Outflow	G251	JW222	15848	ENR012
	G310	M2901	PK919	G310
STA-1W, Northern Flow-way				
Inflow	G304A	W3860	V2485	G302, G303
	zG304B	W3861	V2486	G302, G303
	G304C	W3862	V2487	G302, G303
	G304D	W3863	V2488	G302, G303
	G304E	W3864	VW951	G302, G303
	G304F	W3865	VW802	G302, G303
	G304G	W3866	VW952	G302, G303
	G304H	W3867	VW876	G302, G303
	G304I	W3868	VW872	G302, G303
	G304J	W3869	VW953	G302, G303
Outflow	G306A	W3870	L9866	G306C
	G306B	W3871	L9867	G306C
	G306C	W3872	L9868	G306C
	G306D	W3873	L9869	G306C
	G306E	W3874	L9870	G306C
	G306F	W3875	L9871	G306G
	G306G	W3876	L9872	G306G
	G306H	W3877	L9873	G306G
	G306I	W3878	L9874	G306G
	G306J	W3879	L9875	G306G
STA-1W, Western Flow-way				
Inflow	G255	WF797	VM838	G255
Outflow	G258	---	SG916	G309
	G309	W3882	L9849	G309
	G307	---	VM853	G307
STAA-1W, Eastern Flow-way				
Inflow	G303	W3880	L9830	G303
	G255	WF797	VM838	G255
Outflow	G308	W3881	L9846	G308
	G259	W3884	SG917	G259
	G251	JW222	15848	ENR012

Structures		Preferred DBKEYS	Source DBKEYS	Water Quality Sites
STA-2				
Inflow	S6	15034	06741	S6
	G328	J0718	MQ903	G328
	G328I_P	---	TA605	G328R
	G328I_C	---	TA607	G328R
	G338 from	---	MC705	S10D
	G338 to WCA-	---	MC705	S6
	G339 from	---	MC706	G335
	G339 to WCA-	---	MC706	S6
	G434	---	AI368	G434
	G435	---	AI386	G435
Outflow	G335	N0659	LG726	G335
	G436	---	AI400	G436
STA-2, Flow-way 1				
Inflow	G329A	W3926	N0748	G329B
	G329B	W3927	LG703	G329B
	G329C	W3928	LG704	G329B
	G329D	W3929	LG705	G329B
Outflow	G330A	W3930	LG706	G330D
	G330B	W3931	LG707	G330D
	G330C	W3932	LG708	G330D
	G330D	W3933	LG709	G330D
	G330E	W3934	LG710	G330D
STA-2, Flow-way 2				
Inflow	G331A	W3935	LG711	G331D
	G331B	W3936	LG712	G331D
	G331C	W3937	LG713	G331D
	G331D	W3938	LG714	G331D
	G331E	W3939	LG715	G331D
	G331F	W3940	LG716	G331D
	G331G	W3941	LG718	G331D
Outflow	G332	W3942	LG719	G332

	Structures	Preferred DBKEYS	Source DBKEYS	Water Quality Sites
STA-2, Flow-way 3				
Inflow	G333A	W3943	LG720	G333C
	G333B	W3944	LG721	G333C
	G333C	W3945	LG722	G333C
	G333D	W3946	LG723	G333C
	G333E	W3947	LG724	G333C
Outflow	G334	W3948	LG725	G334
STA-2, Flow-way 4				
Inflow	G337A	---	W1982	G337A
	G434	---	AI368	G434
Outflow	G368	---	VN385	G368
STA-2, Flow-way 5				
Inflow	G435	---	AI386	G435
Outflow	G441	---	AI621	G441

Structures		Preferred DBKEYS	Source DBKEYS	Water Quality Sites
STA-3/4				
Inflow	G370	TA438	T0973	G370
	G372	TA437	T0975	G372
STA-3/4, Eastern Flow-way				
Inflow	G374A	W3964	T8434	G374B
	G374B	W3965	T8435	G374B
	G374C	W3966	T8436	G374B
	G374D	W3967	T8437	G374E
	G374E	W3968	T8438	G374E
	G374F	W3969	T8439	G374E
STA-3/4, Central Flow-way				
Inflow	G377A	W3970	T9945	G377B
	G377B	W3971	T9946	G377B
	G377C	W3972	T9947	G377B
	G377D	W3973	T9948	G377D
	G377E	W3974	T9949	G377D
STA-3/4, Western Flow-way				
Inflow	G380A	W3975	T9955	G380B
	G380B	W3976	T9956	G380B
	G380C	W3977	T9957	G380B
	G380D	W3978	T9958	G380E
	G380E	W3979	T9959	G380E
	G380F	W3980	T9960	G380E

SECTION 6:
EVALUATION OF INUNDATION DEPTH AND DURATION THRESHOLD FOR CATTAIL SUSTAINABILITY

6.1 OVERALL STUDY PLAN SUMMARY

The purpose of this study is to identify the inundation depth and duration threshold for cattail (*Typha domingensis*) sustainability in the Everglades STAs. Field observations and studies have indicated that prolonged moderate and deep inundation is perhaps a cause for cattail decline in the STA treatment cells, e.g., STA-1E Cell 7 and STA-1W Cell 5A. This study will include three components: (1) the analysis of period-of-record (POR) hydrologic data, (2) an in-situ study, and (3) Test Cell study. The analysis of POR hydrologic data will help define the hydrologic characteristics (including inundation depth, duration, and frequency) that occur in EAV cells.

The in-situ study will be conducted in STA-1W Cell 2A and STA-3/4 Cell 2A and help identify environmental factors influencing cattail sustainability in the STAs. The results of the POR data analysis and the in-situ study will provide useful information to finalize the experimental design of the Test Cell study. In the Test Cell study, healthy cattail stands will be established in the STA-1W North Test Cells and will be allowed to mature. Subsequently, the ecophysiology of cattail will be assessed at variable inundation depths for certain inundation durations. Cattail survival, growth, biomass, reproduction, mortality, photosynthesis, leaf area index (LAI), and tissue nutrient concentration will be examined through field measurements and laboratory analyses. These data will then be used to identify the duration threshold at an inundation depth for cattail sustainability. The ability of the cattail stands to reduce phosphorus concentrations in the water column will be also evaluated. The results of this study are intended to provide data that will facilitate the development of effective hydrologic strategies for sustainable vegetation management in the STAs and FEBs.

6.2 BASIS FOR THE PROJECT

Key Science Plan Question Study Addresses

- Key Question 3: What measures can be taken to enhance vegetation-based treatment in the STAs and FEBs? [Note: Vegetation-based treatment in the STAs and FEBs refers to the use and management of EAV, SAV, and periphyton to provide treatment.]

Science Plan Sub-Question Study Addresses

- How does water depth affect sustainability of dominant vegetation?

6.3 BACKGROUND/LITERATURE REVIEW

The STAs are operated by the SFWMD to treat P enriched runoff from agriculture and other sources as part of the Everglades restoration effort (see **Figure 6-1**), and are influenced by fluctuating hydrologic conditions (Ivanoff et al., 2012). High flows during storm events can result in excessive inundation for cattail which provides important functions in P retention processes in the STAs. Uneven topography within treatment cells also leads to deep water areas even when target stages are maintained. Understanding the influence of hydrologic conditions on cattail is essential for the development of management strategies to improve the water quality of STA discharges.

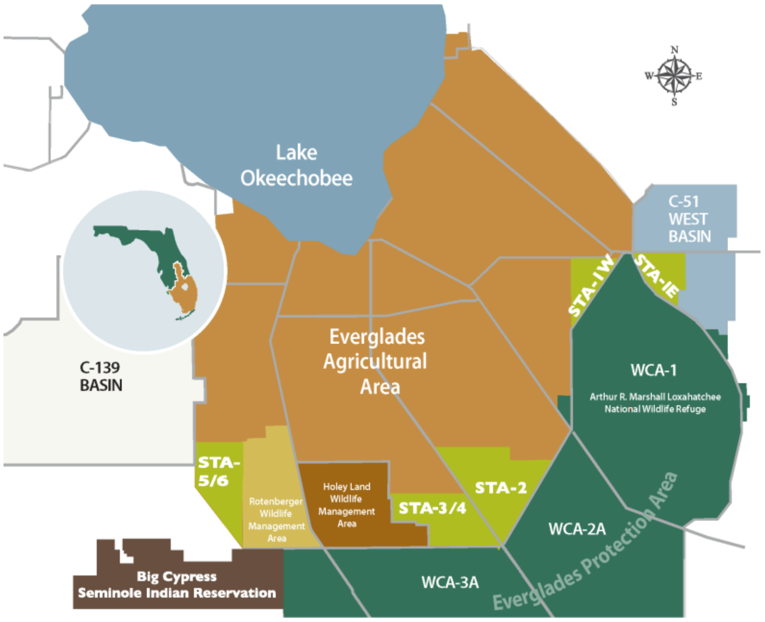

Figure 6-1. Location of the Everglades STAs (STA-1E, STA-1W, STA-2, STA-3/4, and STA-5/6) in relation to the Everglades Protection Area and major basins south of Lake Okeechobee.

Mesocosm and field-based studies on impacts of inundation to cattail and P removal have been conducted by the SFWMD since 2006. The mesocosm-based study indicated that increasing inundation depth from 1.3 to 3.0 or 4.5 ft during a six-week inundation period decreased biomass, growth, and photosynthesis of cattail. Following a four-week recovery period, adverse impacts to cattail caused by the 3.0 ft inundation were reversed, except for belowground biomass loss and decreased total non-structural carbohydrate (TNC) storage, while the impact of the 4.5 ft inundation to cattail was not reversed in the four-week recovery period (Chen et al., 2010; 2013). The field-based study in Cell 7 of STA-1E demonstrated that prolonged inundation at higher than target depths decreased shoot elongation, density and photosynthesis of cattail and has the potential to adversely impact the capacity of the system to treat P (Chen and Vaughan, 2013). It is important to maintain vegetation within an optimal zone of inundation to maintain a sustainable treatment system. This proposed investigation is a follow-up to previous studies and will examine combined effects of depth and duration of inundation on cattail sustainability.

Cattail grows in a wide range (i.e., -0.2−4.0 ft) of inundation depths (Grace, 1989) and survives up to 5.0 ft of inundation (Fraga and Kvet, 1993) because it supplies oxygen to the rhizosphere through highly pressurized gas flow (Sorrell and Hawes, 2010). However, cattail has high shoot density and flowering incidence only under shallow inundation conditions (Chen and Vaughan, 2013; Grace, 1989). Studies have demonstrated that increasing inundation depth from 1.3 to 3.0 ft for a period of six weeks decreased growth, biomass, photosynthesis, and belowground non-structural carbohydrate storage of cattail, while increasing inundation from 1.3 to 4.5 ft for six weeks caused 6 percent cattail mortality (Chen et al., 2010). After cattail was returned to a depth of 1.3 ft during a four-week recovery period, damage to roots, belowground biomass loss, and decreased belowground TNC storage experienced by those plants in excess of 3.0 ft inundation was not reversed (Chen et al., 2013). Increasing inundation also reduces the anchorage capacity of cattail, sometimes resulting in floating cattail mats, as the plants respond to deep inundation by decreasing biomass allocation to rhizomes and roots and increasing allocation to shoots (Grace 1989; Chen et al. 2010; Miao and Zou 2012).

The duration of inundation is critical to wetland plants (Keddy, 2010). Prolonged deep water conditions result in irreversible oxygen depletion to plant cells and an increase in the buildup of harmful chemicals, such as hydrogen sulfide (Li et al., 2009). Other deleterious effects of extended inundation stress on cattail include inhibited root and rhizome growth and shoot photosynthesis and may result in the development of floating cattail mats (Chen and Vaughan 2014). Beule (1979) reported that once *T.* × *glauca* was established, this hybrid withstood continuous 2.5 ft inundation for a two-year period without apparent loss or thinning of plants. In the 3rd year, 50 percent of plants died or did not produce living sprouts and shoot density was 50 percent lower than the previous year and marshes became open-water habitats in the fourth year. However, limited information is available on the effect of inundation duration on cattail survival and sustainability in the STAs.

Hydrologic regimes are a major driving factor in constructed wetlands (CWs) and affect the physical, chemical, and biological processes of P cycling (Kadlec and Wallace, 2009; Chen, 2011). Prolonged deep inundation results in stress to plant roots, limiting cattail growth and propagation and subsequently decreases P removal in the STAs (Chen and Vaughan, 2013). The volumetric P removal rate consistently decreased with increasing inundation depths in two CWs in Listowel, Ontario and Hillsboro, Oregon (see Kadlec and Wallace, 2009). A study on several shallow Florida lakes indicates that at low hydraulic loading rates, the lakes exhibit similar P removal rates to emergent dominated CWs, but at high hydraulic loading rates P removal performance declines (Knight et al., 2003). In contrast, Coveney et al. (2002) demonstrated that TP removal rate increased with increasing HRL from 2 to 12 cm/day in a treatment wetland.

Water stages in cattail cells are generally managed with a target depth of about 1.2 ft between flow events. However, peak flows in the wet season, particularly during and/or following storm events, can cause continual inundation of >3.0 ft in treatment cells for up to two months. Knowledge of how cattail growth and propagation relate to variable inundation depths and durations is vital to understanding and predicting the responses of cattail stands to management of hydrologic regimes. Little research is available on the depth and duration threshold for cattail sustainability in STAs.

The study is designed to evaluate how the survival, growth, propagation, and mortality of cattail are influenced by depth and duration of inundation. Specifically, the growth, photosynthesis, LAI, density, mortality, and tissue nitrogen and P concentrations of cattail in response to variable depths and durations of inundation will be evaluated. The ability of the cattail stands to reduce P concentrations in the water column will also be evaluated.

6.4 CONCEPTUAL MODEL

To accomplish the objective, a controlled experiment designed to evaluate the impacts of inundation depths and durations to growth and ecophysiology of cattail will be conducted in the Test Cells of STA-1W. **Figure 6-2** presents the conceptual model to guide this research. The basic premise of this model is that changes in inundation depth and duration, along with the resulting changes in soil Eh and oxygen and nutrient availability, control photosynthesis, survival, aboveground and belowground growth, and mortality of cattail, further influencing nutrient and carbon dynamics, litter decomposition, and subsequently P removal in the STAs.

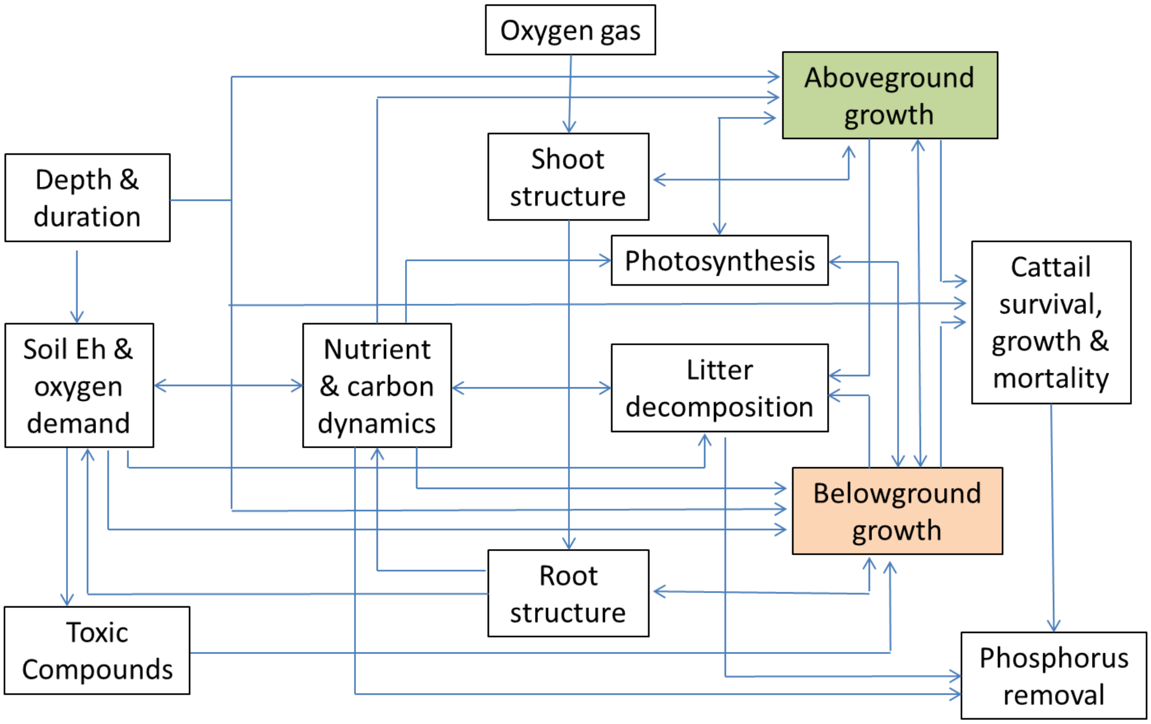

Figure 6-2. Conceptual model showing the effect of inundation depth and duration on the survival and growth of cattail in constructed wetlands.

6.5 STUDY PLAN OBJECTIVES

The depth of inundation can be categorized into optimal, subtle stress, and lethal stress zones in cattail dominated treatment cells of the STAs (Chen and Vaughan 2013). It is assumed that both short-period deep inundation (lethal stress) and prolonged moderate inundation (subtle stress) are harmful to cattail and P removal in the STAs. Too much or too little inundation (extremely prolonged deep inundation or severe drought) does not frequently occur in the STAs although cattail survives a wide range of inundation depths. Based on field observation and the analysis of period-of-record hydrologic data, frequently occurring inundation varies from 1.0 ft to 3.0 ft in the STAs. With FEBs constructed in the near future, STAs may experience relatively shallower inundation depth and less flooding pulses.

In this study, it is hypothesized that (1) there is an inundation duration threshold for cattail sustainability at a specific inundation depth, in terms of survival, growth, and propagation of cattail; (2) the inundation duration threshold is longer at relatively shallow inundation depth than at deep inundation depth; and (3) longer inundation durations than the threshold result in a decrease in cattail sustainability, thus impacting P removal efficiency in the STAs. The objective of this study is to quantify the survival, growth, photosynthesis, LAI, density, mortality, and nutrient (N and P) concentration of cattail growing in variable inundation depth and duration conditions. This study includes three components: the analysis of period-of-record hydrologic data, an in-situ study, and Test Cell study. The results of those research studies will help identify the depth and duration threshold for cattail sustainability and will facilitate the development of water level management strategies in the STAs and FEBs.

6.6 DETAILED STUDY PLAN AND EXPERIMENTAL DESIGN

6.6.1 Study Plan Description and Experimental Design

Healthy uniform cattail stands in the North Test Cells of STA-1W will be established through natural recruitment, allowed to mature, and then assessed for the survival, growth, density, photosynthesis, LAI, mortality, and tissue nutrient concentration responses to proposed inundation depths varying from 1.0 to 3.0 ft for certain inundation durations varying from few weeks to one year or two that will be determined and finalized when the results of the historical data analysis and the in-situ study are available. These data will then be used to identify the duration threshold at an inundation depth for cattail sustainability. The ability of cattail stands to reduce P concentration in the water column will be determined by evaluating changes in TP, SRP, and TDP concentrations from inflow to outflow in the Test Cells. The study results will provide data to develop effective hydrologic strategies for vegetation management in the STAs and FEBs.

6.6.2 Historical Hydrologic Data Analysis

Historical stage data will be examined to determine the range of frequency, timing, depth, and duration of inundation in the following EAV cells of the STAs (**Table 6-1**). The analysis of historical hydrologic data will help define the hydrologic characteristics (including inundation depth and duration) that occur in EAV cells and provide information for the experimental design of the Test Cell study.

Table 6-1. Selected EAV treatment cells in the STAs.

STA	Treatment Cells
STA-1E	Cells 3, 5, and 7
STA-1W	Cells 1A, 2A, and 5A
STA-2	Cells 1 and 2
STA-3/4	Cells 1A, 2A, and 3A
STA-5	Cells 1A and 2A

Static images will be created showing the estimated spatial distribution of water depths through the EAV treatment cells under a range of stages or water elevations. The static images will be generated using a digital elevation model (DEM) based on surveyed topographic data. ArcGIS will be used to differentiate water levels from the DEM under various operating water depths. A map showing the estimated water depths throughout the treatment cell will be generated using color-coded gradients. From these series of water depth maps, the distribution (acreage and percentage) of water depths within each treatment cell will be calculated.

6.6.3 In-situ Study: Environmental Factors Controlling Cattail Sustainability in the STAs

Primary environmental factors controlling cattail sustainability in wetlands are thought to be hydrologic regime and soil physic-chemical conditions. The hydroperiod (depth, duration, and frequency of inundation) is the hydrologic signature of wetlands. The hydroperiod is not only an important determinant of the establishment and maintenance of cattail community but it also modifies and determines the abiotic physic-chemical environment, e.g., soil fertility and redox potential (Kadlec and Wallace 2009; Chen 2011). In this study, it is assumed that floc and soil characteristics do not dramatically change in cattail-dominated cells of the STAs in one year or two. The hydroperiod is considered a primary factor affecting cattail sustainability in the STAs, in terms of cattail growth and density. The objective of the proposed in-situ study is to identify environmental factors influencing cattail sustainability in the STAs. The results will be used to finalize the Test Cell experimental design. This in-situ study will be conducted in 2014 and 2015.

STA-1W Cell 2A and STA-3/4 Cell 2A (**Figures 6-3** and **6-4**, respectively) have been selected to examine cattail sustainability. STA-1W Cell 2A has cattail decline, deep soil layer, and a hydraulic short circuiting along the flow path, while STA-3/4 Cell 2A has relatively healthy cattail stands, shallow soil layer, and a typical north-to-south flow path. Vegetation plots (5 ft × 10 ft) with four PVC pipes at the four corners will be set up in both unhealthy and healthy cattail stands (in terms of cattail growth and density), with different inundation conditions, in part based on cell topography. The number and location of the vegetation plots will be further determined following initial field survey. The GPS coordinates for the four corners of each plot will be recorded with a GeoXT6 device.

Cattail health indicators and environmental variables will be determined in the vegetation plots. Environmental variables include inundation, floc, and soil parameters. The depth, duration, and frequency of inundation will be monitored daily with a water level sensor (Levelogger Edge Model 3001), at least, one unit at each of three locations (inflow, interior, and outflow areas) in each of the two treatment cells. Floc and soil samples will be collected in the vegetation plots for baseline data, once after the vegetation plots are set up during the study period. Floc and soil bulk density, ash-free dry weight (AFDW), TC, TN, and TP as well as soil thickness, texture, labile N, and labile P will be determined. Soil redox potential will be measured bimonthly along with the measurement of cattail health index described below.

Cattail health indicators include maximum shoot height, coverage, juvenile and adult cattail density, growth vigor (shoot elongation), photosynthesis, LAI, and leaf nutrient concentration. Cattail health index will be determined in the vegetation plots bimonthly in June through January each water year based on site accessibility by air boat. Aboveground shoots will be harvested for the determination of leaf TN and TP concentrations. In order not to disturb subsequent vegetation survey and measurement in the vegetation plot, initial harvest will be conducted in cattail stands nearby the vegetation plot, with 0.25 m^2 quadrat at the beginning of the study period. A final harvest will be conducted in the vegetation plot with 0.25 m^2 quadrat by the end of the study period. Other species cover will be estimated if they occur in plots.

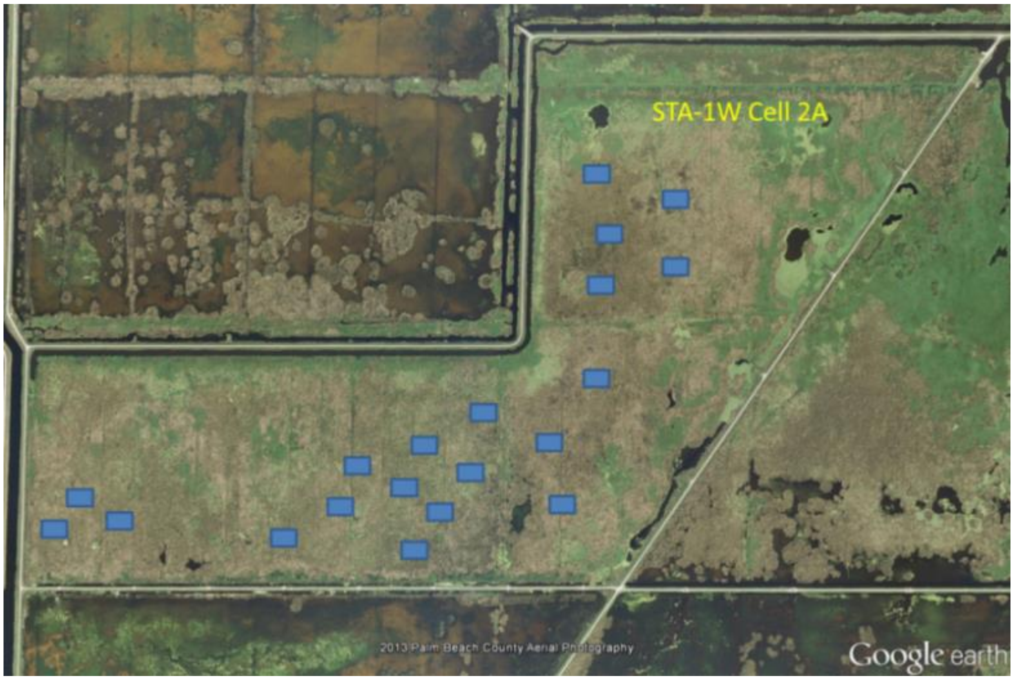

Figure 6-3. Layout of sampling plots in cattail stands in STA-1W Cell 2A. Blue rectangles represent (5 ft × 10 ft) vegetation plots (imagery from 2013 Palm Beach County aerial photography).

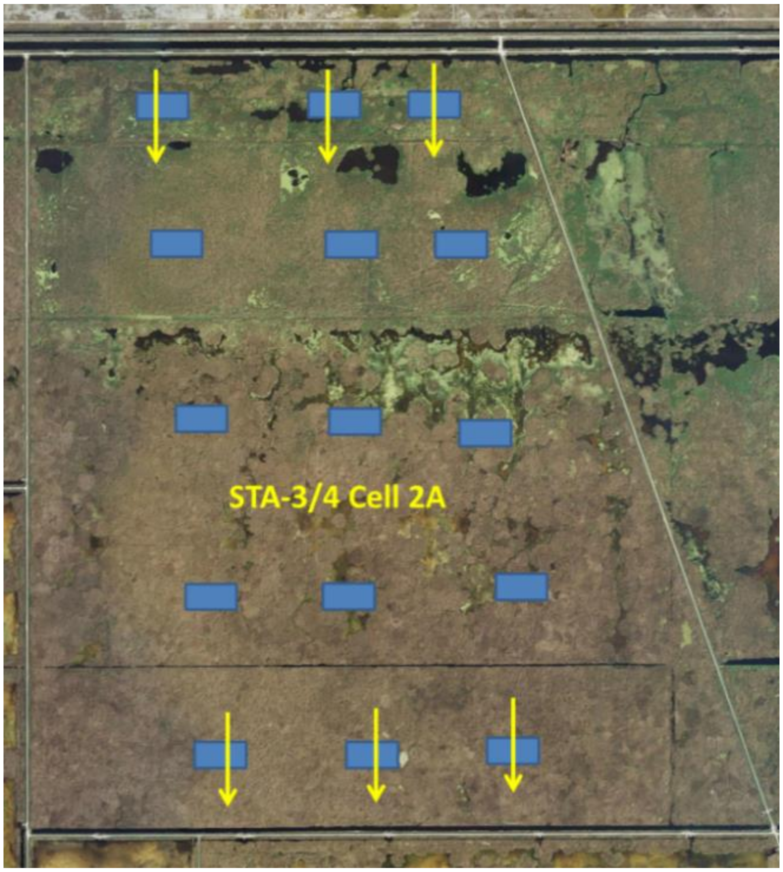

Figure 6-4. Layout of sampling plots in cattail stands in STA-3/4 Cell 2A. Blue rectangles represent (5 ft × 10 ft) vegetation plots and yellow arrows depict the flow path and direction (imagery from 2013 Palm Beach County aerial photography).

6.6.4 Experimental Design for Test Cell Study

The experiment will have two factors: inundation depth and duration. The inundation depth treatment has five levels varying from 1.0-3.0 ft for certain inundation durations varying from few weeks to one year or two. The results of the historical data analysis and the in-situ study will help refine these treatment levels. The maximum inundation duration will be achieved to push cattail into a lethal stress stage in terms of mortality. The experiment will have three replicates and 15 experimental units (Test Cells) in total.

This study will be conducted in the Test Cells of STA-1W (**Figure 6-5**) after modification to these cells is finished to obtain uniform soil fertility for cattail establishment. The Test Cells are 0.2 ha, shallow, fully lined CWs arranged into two groups of 15 cells each, located at the northern and southern ends of STA-1W. The northern Test Cells will be used in this study. Inflow to the Test Cells will come from the inflow to STA-1W. Each Test Cell is hydrologically isolated from adjacent Test Cells and the surrounding STA, which allows for independent control of inflow as well as the depth and duration of inundation. Water from the inflow to STA-1W will be pumped at the discharge point of structure G-303 (approximating untreated stormwater) into a water storage pond, which will be maintained at a stage of 1−2 ft above the maximum water levels in the Test Cells. Water from the storage pond will be gravity fed to the adjacent Test Cells and regulated by separate hand-controlled inflow valves. Outflow from each test cell will be controlled by an adjustable 90° v-notch weir.

During the modification, uniform new organic soil will be added to the 15 north Test Cells. The organic soil in 30-cm top layer will have approximately 400−500 mg P /kg. The refurbished Test Cells will receive the same inflow with a TP concentration of 100−150 µg P/L. The refurbishment of the Test Cells will also include the construction of boardwalks that will allow access to the interior portions of each cell without disturbing the vegetation or soil substrates (**Figure 6-6**). Soil samples from each of the 15 Test Cells will be collected to obtain baseline data and analyzed for pH, bulk density, AFDW, TC, TN, TP, and available N and P, following cell hydration.

The Test Cell facilities contain inherent limitations in comparison to STA treatment cells, i.e., cattail communities within the Test Cells will not replicate all the conditions found within the STA treatment cells (Chimney and Newman, 2006). The hydrology and hydraulics of the Test Cell differ from those of the treatment cells in terms of wave action, flow, and flow velocity. Therefore, the effects of these variables will not be evaluated in this study. In addition, this study may be impacted by significant weather events, such as extreme rainfall, large storm events, or severe regional droughts.

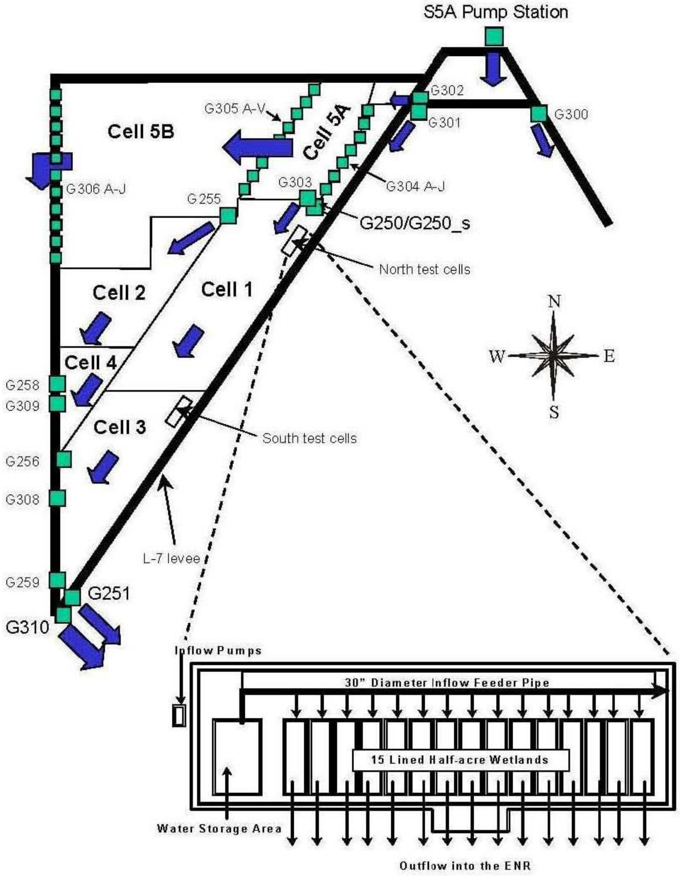

Figure 6-5. Northern and southern Test Cells of STA-1W.

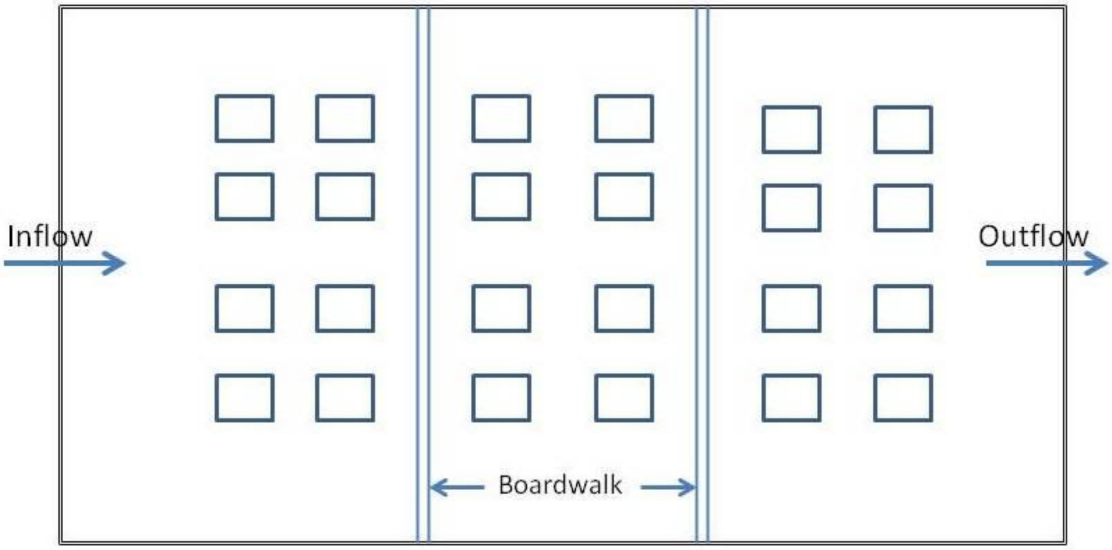

Figure 6-6. Setup of boardwalks and sampling plots in a Test Cell. Each of the squares represents a 5 ft × 5 ft sampling plot.

6.6.5 Methods

Cattail Recruitment

Natural recruitment will be used to establish the cattail stands in the Test Cells. The water levels in the Test Cells will be saturated to inundation of 0.5−1 inch above the soil surface to encourage cattail seed germination. Cattail seeds will be broadcast by hydro-seeding and/or cattail plants may be manually planted as needed.

Establishment of Cattail Community and Sampling Plots

Uniform, mature, and healthy cattail stands will be established via natural recruitment and hydro-seeding as needed during a one-year or longer period. Cattail will be allowed to grow and fully mature, prior to the initiation of experimental testing, to better emulate cattail stands within the STAs. In the period of cattail establishment, the inundation in the Test Cells will be maintained consistently at 1.3 ft or lower depths. Soil samples will be collected at depths of 0−4 inch and analyzed to determine baseline conditions after a uniform healthy cattail stands are established, but prior to the initiation of the experimental testing.

Permanent vegetation plots (5 ft × 5 ft) will be constructed using PVC piping and placed along the inflow, interior, and outflow regions of the Test Cells to be used for vegetation sampling and other necessary measurements, such as photosynthesis, shoot density, and shoot elongation (**Figure 6-6**). Cattail harvesting will be conducted randomly in each of the 15 Test Cells using a 0.25 m^2 quadrat, prior to the initiation of experimental testing, which will be used for additional baseline data.

Implementation of the Experimental Plan

The study will be implemented in the Test Cells as described above. During the study period (~2 to 3 years), surface water, soil/sediment, and vegetation samples will be collected (**Table 6-2**) and analyzed. Specific sample frequencies and analyses are described below.

- Surface Water: Total P, SRP, TDP, total N, ammonia, nitrate, calcium, organic carbon, pH, and DO will be measured bi-weekly at the inflow, interior, and outflow of the Test Cells .
- Soil: Soil (0–5 cm) samples will be collected annually and will be analyzed for pH, bulk density, AFDW, total C, bioavailable N, total N, bioavailable P, and TP. Soil cores will be collected annually at each of sampling stations (inflow, interior, and outflow) in each of the 15 Test Cells. Soil Eh will be measured in three labeled, permanent sampling stations (inflow, interior, and outflow) on a bimonthly basis.
- Vegetation survey and measurement: In the cattail establishment period, photosynthesis, LAI, shoot density, and shoot elongation will be measured bi-monthly in each of the Test Cells and used as baseline data. Cattail harvesting will be conducted randomly using a 0.25 m^2 quadrat, prior to the initiation of experimental testing. Cattail harvest samples will be separated into live leaf, rhizome, shoot base, root, and dead plant material.

  In the implementation period of the study, the frequency of measuring for photosynthesis, LAI, shoot density, and shoot elongation in each of the Test Cells will decrease from bi-weekly, to monthly to bi-monthly, in order to capture the responses of cattail to each of the inundation durations varying from few weeks to one year or two. Cattail harvesting will be conducted randomly with a 0.25 m^2

quadrat in the 5 ft × 5 ft permanent vegetation plots in each of the Test Cells at the end of the inundation durations, with three replicates.

- Hydrology: The flow and water stage of inflow and outflow structures of the Test Cells will be monitored. Water depths will be estimated using state and ground elevations within the Test Cell. Water depth in plots will also be measured while each vegetation survey is conducted.

Table 6-2. Measured variables and their sampling frequency.

	Weekly	**Biweekly**	**Monthly**	**Bimonthly**	**Yearly**
Inflow & outflow water stage	×				
Surface water quality parameters		×			
Soil chemistry					×
Soil Eh				×	
Shoot density		×	×	×	
Shoot elongation		×	×	×	
Photosynthesis & LAI		×	×	×	
Biomass harvesting and tissue TC, TN, TP	Initial harvesting and sampling at the end of each of the 8 inundation durations				

Methods for Determining Variables

Surface water samples will be analyzed according to the standard methods of the Chemistry Laboratory of the SFWMD (American Public Health Association 1998). Soil/sediment samples will be analyzed according to the standard methods of the Chemistry Laboratory of the SFWMD (Standard analytical methods of Soils, Soil Science Society of America). Phytomass samples (including live cattail leaf, rhizome, shoot base, root, and dead plant material samples) will be analyzed for TC, TN, and TP concentrations according to the standard methods of the Chemistry Laboratory of the SFWMD. The number of cattail shoots will be counted in each of the 2m×2m permanent vegetation plots in each of the Test Cells. Shoot density will be estimated as the number of plant shoots per square meter. Cattail mortality will also be recorded in each of the vegetation plots in each of the test cells. Shoot elongation rate of cattail will be determined by tagging and recording the height of the smallest leaf for each selected ramet. The elongation rate will be recorded and calculated, and will be represented as the change in leaf growth expressed in cm day $^{-1}$.

$$Elongation = \frac{(h_2 - h_1)}{T},$$

Where h_1 = initial leaf length (cm), h_2 = final leaf length (cm), T = time interval between two measurements. Photosynthesis of shoots will be measured for 3−5 young but fully expanded leaves in each of selected three plots in each of the Test Cells, one leaf per ramet between 10:00am – 2:00pm, with a LI–6400 portable gas exchange system (LI-COR, Lincoln, Nebraska, USA). The LAI will be measured with LAI-2200 plant canopy analyzer (LI-COR, Lincoln, Nebraska, USA). Plant relative growth rate (RGR) will be calculated as the difference in the natural logarithm of initial and final biomass (dry weight, g) divided by the inundation duration (days):

$$RGR = \frac{(Ln(Biomass2) - Ln(Biomass1))}{(Time2 - Time1)}$$

Where Biomass1 = cattail biomass (dry weight) on Time1 (days), Biomass2 = cattail biomass on Time2 (days), (Time2 – Time1) = time interval (days) between two measurements.

6.6.6 Detailed Study Plan Tasks

Literature Review

Relevant published information will be collected and reviewed, including peer-reviewed journal articles and technical reports.

Data Query and Historical Data Analysis

The POR (ending at WY2013) data on STA stage level, elevation, and other related variables will be retrieved from the SFWMD databases. The depth, duration, frequency, and timing of inundation will be calculated, analyzed, and summarized.

Cattail Recruitment and Establishment in Test Cells

This task will be conducted under contract, supervised by the SFWMD.

Monitoring in Test Cells, STA-1W and STA-3/4

Monitoring tasks include surface water monitoring and measurement, soil sampling and measurement, vegetation survey including measurement, and harvesting in Test Cells and STA-1W Cell 2A and STA-3/4 Cell2A.

Data Management

All data and field notes will be maintained by the District. All data products and associated metadata will undergo quality assurance/quality control screening. All sampling staff will certify that all field notes, chains of custody, and other information comply with SFWMD standards. Accuracy and precision in laboratory analysis will be validated and reflected in the data sets and reports. Data on surface water quality will be stored in SFWMD’s DBHYDRO database. Data on soil and vegetation will be stored in SFWMD’s ERDP database.

Reporting

A quarterly progress report will be prepared and shared with the Science Plan Core Team. Annual reports will be prepared as part of the South Florida Environmental Report, along with a final report with key findings and recommendations.

6.6.7 Study Schedule

• In-situ study	Initiate FY2014	Complete FY2016
• Test cell refurbishment	Initiate FY2015	Complete FY2015
• Test cell grow-in	Initiate FY2016	Complete FY2016
• Test cell implementation	Initiate FY2017	Complete FY2019

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SECTION 7:
STA WATER AND PHOSPHORUS BUDGET IMPROVEMENTS

7.1 OVERALL STUDY PLAN SUMMARY

The purpose of this study is to produce improved annual STA water and phosphorus budgets for Stormwater Treatment Area (STA) treatment cells in order to meet the needs of the Restoration Strategies Science Plan. Water budget analysis is an important tool used to understand the treatment performance of STAs, and accurate water budgets are critical to developing accurate phosphorus budgets. Recently reported STA treatment cell annual water and phosphorus budgets contained high error terms (Pietro, 2013), thereby bringing into question the usability of the data to characterize and understand treatment performance.

The *STA Water and Phosphorus Budget Improvements Study* will be implemented in a phased approach. Phase I included a test case for improving water budgets for individual treatment cells using simplified desktop approaches. Phase II will implement the methodologies investigated during Phase I on an expanded list of treatment cells and will also include developing improved phosphorus budgets for these treatment cells. Phase III, only if determined to be necessary, will include more extensive approaches to further reduce errors in water and phosphorus budgets.

During Phase I, staff evaluated the sources of error associated with annual water budgets, and conducted a desktop evaluation of STA-3/4 Cells 3A/3B (**Figure 7-1**) as a test case for improving STA water budgets. Phase I results were promising, producing greatly reduced annual water budget residuals for the test case as well as simplified flow and seepage estimation methods that can be applied to other treatment cells. It is important to note that the sources of error and improvements in water budgets for Cells 3A/3B may not apply to all treatment cells. It is also noted that STA phosphorus budget evaluation was not included in Phase I, but is proposed to be included in Phase II.

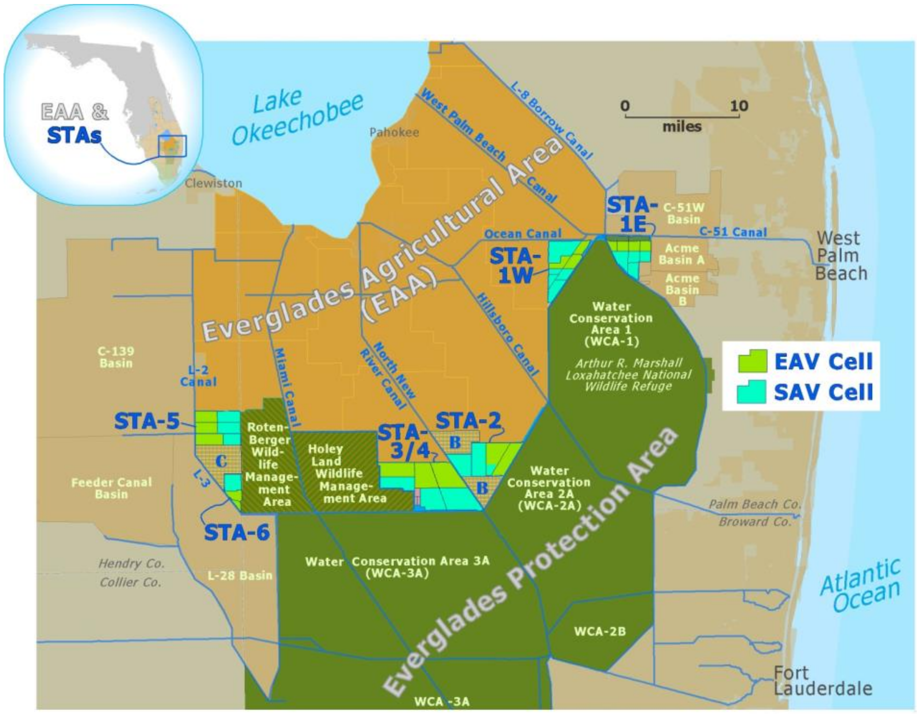

Figure 7-1. Location of the Everglades STAs.

7.2 BASIS FOR THE PROJECT

Science Plan Question Study Addresses

- Study Question: Can improvements be made to STA water and phosphorus budgets published in recent editions of the SFER?

Science Plan Sub-Question Study Addresses

- What are the sources of errors in STA water budgets?
- How can errors in water budget components be reduced?

7.3 CONCEPTUAL MODEL

STA water budgets (**Figure 7-2**) are comprised of structure flows (inflows and outflows), rainfall, ET, seepage, and change in storage. Structure flow is calculated using a hydraulic equation developed for each water control structure. Rainfall is estimated using data collected at rain gauges located within or near each STA. Evapotranspiration (ET) is estimated from first-order models with coefficients specific to different wetland vegetation communities. Seepage is estimated as flow through perimeter levees, and is based on head differences between the treatment cell and outside area water levels, levee length, and a first-order seepage coefficient [cubic feet per second per mile per feet (cfs/mi/ft)]. The Water Budget Application Tool is used to develop estimates of seepage (if calculated), rainfall volume, ET, and change in storage volumes. The water budget residual, the mathematical difference between all outflow and inflow sources, is used as a measure of overall accuracy of the estimates of these components. Developing a closed water budget is not a simple task due to the physical characteristics of wetland systems and errors associated with the measurement and estimation of each of the water budget components. Attempts to reduce the errors and uncertainties with each of the water budget terms should be made where feasible in order to improve the usability of the data to characterize treatment performance in individual cells.

STA phosphorus budgets are intrinsically tied to the water budgets and are comprised of structure flows (inflows and outflows), rainfall, and seepage (see **Figure 7-2**). Phosphorus concentration data collected at STA treatment cell structures is combined with flow data (using the web-based Nutrient Load Program) to develop the structural flow component of the phosphorus budgets. The phosphorus load in precipitation is based on annual rainfall volume multiplied by the median rainfall phosphorus concentration [4 parts per billion (ppb)] monitored at STA-1W (Site ENR308) from May 1, 1999–April 30, 2012. STA seepage phosphorus concentration data is limited but estimates can be developed using available data and best professional judgment. Phase II of this study is intended to reduce the overall errors associated with individual treatment cell annual water budgets for select treatment cells using a desktop approach, similar to the approach used during Phase I of this study and will include improvements to phosphorus budgets for the selected treatment cells. Improved historical annual water and phosphorus budgets will be developed for STA-2 (Cells 1-3) and STA-3/4 (All Cells) for each STA's entire period of record (POR) and for other STA treatment cells for WY2014 through WY2020. STA-2 and STA-3/4 were selected for historical (POR) water and phosphorus budget improvements as a result of a resource prioritization effort to focus on treatment cells most relevant to the successful completion of the Science Plan studies.

If the Phase II desktop approach is not successful in adequately reducing the error term(s) associated with a particular treatment cell's annual water budget(s), other more resource intensive efforts to reduce the error(s) may be considered on a case by case basis. Such work, if needed to meet the needs of the Science Plan, would be initiated under Phase III of this study. The *Study Design* section of this plan includes a list of potential Phase III activities that may be considered.

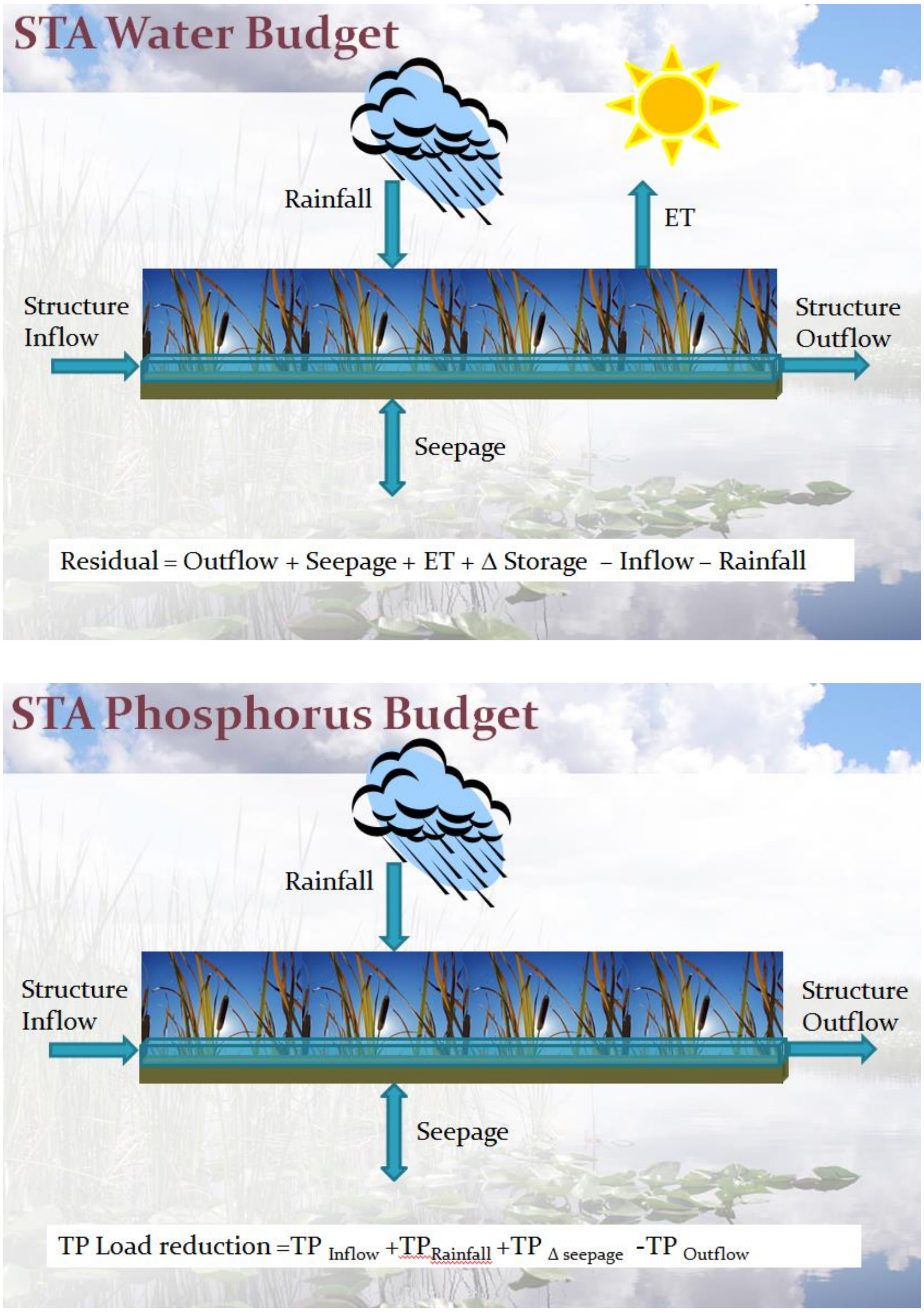

Figure 7-2. STA water budget (top) and phosphorus budget (bottom) conceptual models.

7.4 STUDY PLAN OBJECTIVES

The purpose of this study is to produce improved annual STA water and phosphorus budgets for STA cells in order to meet the needs of the Restoration Strategies Science Plan. Within the current scope, STAs for which historical POR water and phosphorus budgets will be created are STA-2 (Cells 1-3) and STA-3/4 (All Cells). Furthermore, in approximately FY2015, improved water and phosphorus budgets are proposed to be developed for other STA treatment cells starting with the WY2014 data. Strategies developed during Phase I will be utilized where possible in Phase II to develop improved water budgets. For example, simplified methods used to develop improved flow estimates for mid-levee culverts will be evaluated for use in improving flow estimates for culverts in other treatment cells. Similarly, simplified methods used to estimate seepage for the test case will be utilized for other treatment cells where possible. Based on discussions at the August 7, 2013 workshop with the Science Plan Technical Representatives, a reduction of the annual water budget residual to 10 percent or less will be considered an acceptable level of improvement. Once complete, the improved water budgets will be used to develop the phosphorus budgets for each of the treatment cells.

7.5 DETAILED STUDY PLAN

7.5.1 Study Plan Description

Historical Data Analyses

Phase I of this study consisted of two key data analysis components. The first component involved an evaluation of errors associated with annual STA water budgets. The second component of Phase I consisted of developing an improved annual water budget for the test case of STA-3/4 Cells 3A/3B. The following sub-sections provide summaries of the two Phase I data analysis components, which have been completed to date.

Phase I – Evaluation of Errors in Water Budget Components

Structure flows are generally the largest component of annual water budgets (80-90 percent). Inflows to and outflows from STA cells are generally controlled by culverts that must be large enough to pass peak storm flows without exceeding design stages. Much of the time these large culverts operate under very small head differences resulting in flow estimates that are subject to large errors. The impact of these errors on the annual water budget residual can be significant.


Figure 7-3. A typical rain gauge.

Rainfall is relatively a smaller component of the annual water budget (e.g., STA-1W, 11 percent of inflows, 1999-2012, Abtew et al., 2013). The rainfall component of STA water budgets is currently estimated from rain gauges (**Figure 7-3**) in the STA or the nearest gauges in the surrounding area. NEXRAD (Radar) rainfall estimates are also available and have been used to fill in gaps in the rain gauge data. And while the NEXRAD rainfall estimates are adjusted using available rain gauges, evaluation of the NEXRAD and gauge rainfall shows the two data sets can have notable differences. It is currently not possible to determine which data set is superior; therefore, it is recommended that STA water budget analyses continue using rain gauges when available with NEXRAD data used to fill gaps.

A list of the current rainfall gauges associated with each of the STAs follows, with details shown in **Table 7-1**.

- STA-1E rainfall estimates are from gauges in STA-W which are stored in a preferred DBKEYS as the average of five rain gauges inside STA-1W. It is noted that when there is a rain storm that is localized to STA-1E, there is estimation error.
- The STA-1W rainfall network of five gauges inside the STA provides good rainfall measurements on a daily basis.
- STA-2 rainfall is estimated from one gauge (G-331D) at the STA northern edge.
- STA-3/4 rainfall is estimated from two rain gauges, one in the southeast corner (S7) and one north of the STA (EAA5).
- STA-5 and STA-6 rainfall is estimated from a rain gauge at weather station STA5WX at the far northeast corner of STA-5.

Table 7-1. Rainfall stations currently used for STA water budget calculations.

STA 1-W & STA-1E			
Station	**DBKEY**	**Remark**	**Preferred DBKEY***
ENR101	15851	---	no
ENR203	15874	---	no
ENR301	15877	---	no
ENR308	15888	---	no
ENR401	15862	---	no
Average	KN809	Thiessen weighted average of available gauges	yes
STA-2			
Station	**DBKEY**	**Remark**	**Preferred DBKEY***
G331D	PT420	---	no
STA-3/4			
Station	**DBKEY**	**Remark**	**Preferred DBKEY***
S7	15204	---	yes
EAA5	15184	Preferred DBKEY JW233 discontinued in 2010	no
STA-5			
Station	**DBKEY**	**Remark**	**Preferred DBKEY***
STA5WX	UK533	---	yes
STA-6			
Station	**DBKEY**	**Remark**	**Preferred DBKEY***
STA5WX	UK533	---	yes

* Preferred DBKEYs undergo a QA/QC process of data improvement and estimation of data gaps

.*ET* is a relatively smaller contributor to the annual water budget (e.g. STA-1W, 12 percent of outflows from 1999–2012, Abtew et al., 2013). Several different ET estimation methods including existing models and satellite-based estimation were evaluated as part of this study. The results of the evaluation showed they had no significant differences; seasonal variations were canceled out in the annual analysis. ET for the STAs is currently considered to be one of the better quantified components in the STA water budgets. DBHYDRO has PREF (Preferred) data for Potential ET which is evaporation/evapotranspiration from wetlands and water bodies. The model for ET computation was developed from lysimeter experiments in the Everglades Nutrient Removal Project (ENR). This model has been published in many peer-reviewed journal articles and books and is applied in several countries. Any additional effort to estimate ET for the STA water budgets might not make a significant change to the accuracy of the water budgets. The STA ET data is derived from a model that uses input data from the closest weather station as follows:

- STA-1E ET estimation with input from ENR308 weather station
- STA-1W ET estimation with input from ENR308 weather station
- STA-2 ET estimation with input from ROTNWX weather station
- STA-3/4 ET estimation with input from ROTNWX weather station
- STA-5 ET estimation with input from STA5WX weather station
- STA-6 ET estimation with input from ROTNWX weather station

Seepage can be difficult to quantify and it may be highly variable from STA to STA, from cell to cell and from season to season. As a result, seepage estimates can have varying levels of impact on water budget analyses. In some case, seepage may be in both directions making the net effect on the water budget small, however, the impact can be significant in cells where head differences with the surrounding areas are high and the geology is favorable for seepage. Seepage estimates were added to the STA-3/4 Cells 3A/3B water budget, thereby improving previously reported water budgets for these cells. The estimated contribution of seepage to the annual water budget for Cells 3A/3B water budget was relatively small (test case range of approximately 2 to 4 percent).

Change in Storage is typically the smallest component of the overall annual water budget (less than 1 percent). It should be noted that for daily or weekly water budget analyses, relative magnitude of this component may be larger and the daily estimates of change of storage should rely on end of the day stage values, as opposed to daily mean stages.

Phase I – Water Budget Improvements Test Case (STA-3/4 Cells 3A/3B)

1. Low head differentials across large culverts in mid-levees were found to be the main source of error in Cells 3A & 3B flow estimates and annual water budgets.
2. The test case annual water budgets greatly improved by revising mid-levee structure flow data using several simplified methods; overall annual water budget error terms reduced from as high as 100 percent to as low as 10 percent or less.
3. *Rainfall, ET* and *Change in Storage* terms were minor contributors to the test case annual water budgets. It is noted however that Rainfall and ET can be larger contributors to water budgets during extreme drought years. Regardless, the current estimation methods for these water budget components were found to be acceptable, and no changes were made for the Cells 3A & 3B annual water budgets for these components.
4. *Seepage* estimates were developed for Cells 3A and 3B based on seepage coefficients obtained from the analysis of historical dry periods (no rainfall, inflow, or outflow) and were added to annual water budgets as an improvement to previous SFER reporting. The estimated

contribution of seepage to the annual water budget for Cells 3A/3B water budget was relatively small (test case range of about 2 to 4 percent). The current method for estimating seepage for STA treatment cell annual water budgets is considered acceptable for Phase II. No seepage studies are planned for Phase II of this study; they may be part of Phase III on a case by case basis or generated by other ongoing or planned Science Plan Studies. It is further noted that for Phase II, the Water Budget Tool needs to be updated to include seepage estimates for all STA cells (similar to Phase I work for Cells 3A & 3B).

5. Overall annual water budget results in Phase I for STA treatment cells are considered acceptable for use in characterizing TP performance, and for preliminary hydraulic and TP modeling efforts.
6. One or more Technical Publications are expected as result of Phase I effort. These technical publications will provide the details of the analyses conducted in support of the results and recommendations that resulted from the Phase I effort.
7. Note that the test case results may not translate to other STA cells. Different methods and potentially more intensive and costly efforts may be required to produce acceptable improvements to the water and phosphorus budgets for some cells. Such Phase III work would be investigated on an as needed basis.
8. Ongoing streamgauging, structure ratings, re-surveys, and flow data improvement efforts for the STA structures are assumed to continue by the District.

7.5.2 Study Plan Components

Phase I – Complete

Phase I included an evaluation of errors associated with annual STA water budgets and development of improvements to annual water budgets for STA-3/4 Cells 3A/3B as a test case.

Phase II – Proposed Activities

As a result of the Phase I activities and results, as well as input from the Science Plan Study Leads, the following recommended Phase II activities are proposed to meet the needs of the Restoration Strategies Science Plan.

1. Develop improved historical flow estimates for the entire POR for STA-2 (Cells 1-3) and STA-3/4 (Cells 1-3) structures using one or more of the simplified methods investigated during Phase I to develop more accurate water budgets for all STA cells. The improved historical flow data will be entered in new MOD DBKEYS in DBHYDRO.
    - One-year effort (FY2014) to produce the improved historical flow data for STA-2 and STA-3/4 structures and enter the improved historical flow data in new MOD DBKEYS in DBHYDRO.
    - Once modelers and other technical staff start using the improved historical flow data, its suitability can be determined and further refinements to the flow data may be made. In cases where significant data inadequacies still exist, further improvements can be considered on a case by case basis as needed to meet the needs of the Science Plan (see *Phase III - Potential Items* below).
    - Once the new MOD DBKEYS are populated with the improved historical flow data, these MOD DBKEYS will continue to be populated with improved flow data for all structures for the life of the modeling project through FY2020 at which time the current study and associated flow data refinement would end. Continued development of water and phosphorus budgets based on the techniques and methods

developed by this study may continue based on the outcome of a needs assessment at that time but is outside the scope of this study.

2. Develop improved water budgets using the improved historical flow data in the new MOD DBKEYS and the Water Budget Tool. Seepage estimates will be added to treatment cells that previously did not include them. The target of this task is to reduce the overall treatment cell water budget error to 10 percent or less.
   - Initial effort is estimated to occur over the first half of FY2015 and then annually thereafter.
   - Will require agency-wide support from to help update Water Budget Tool to include seepage estimates for all STA cells. Work is expected occur in FY2014 in parallel with creation of improved flow data in MOD DBKEYs.
   - Future water and phosphorus budget reporting to include appropriate data usability caveats and documentation of assumptions.
3. Develop improved phosphorus budgets using the improved flow estimates and water budgets. In order to complete this task, output files from the Water Budget Tool and the Nutrient Load Program will be combined in an Excel spreadsheet that will then be used to develop phosphorus budgets (using the same method that has been previously used in the annual SFER reporting). Phosphorus concentrations for rainfall and seepage will be selected based on previous studies and best available data. The final product for this task will be a summary table that includes all the water and phosphorus budget components for the subject treatment cells on an annual (water year) basis.
   - Initial effort is estimated to occur over second half of FY2015; then annually thereafter at a reduced level of effort.
     - Seepage estimates will rely upon best available information and will be clearly documented.
     - Results replace cell by cell historical phosphorus budget tables previously reported in the annual SFER.

Phase III – Potential Activities

If the results of Phase II indicate the need for further improvements to flow estimates or other components of STA water or phosphorus budgets, following are potential activities that may be considered. This list provides a general idea of the types of items that could be completed; additional items and details such as costs can be provided in the future if any of these items are determined to be necessary. It is also noted that some of the items below may be utilized during Phase II on a case by case basis.

Structure Flows

- Install small temporary pumps during periods of low flow
- Perform structural retrofits such as installing removable v-notch weirs at culvert inlets or permanent v-notch weirs next to culverts to pass low flows during dry periods
- Resurvey reference elevations at stilling wells at flow structures in a closed loop
- Install additional pairs of stilling wells/stage sensors for structures that use surrogate stage data

- Install temporary flow meters at select structures to collect accurate field flow measurements intended to improve rating calibration and verification at these structures
- Perform periodic inspections and calibrations to correct for sensor drift, sensor malfunctions, or reference elevation
- Collect more field data at low head conditions
- Investigate alternative analytical methods for estimating flow through culverts
- Automate the gate operations at structures as a function of head differential
- Change the timing of discharges by operating the structures for shorter periods with higher discharge rates, or operate fewer structures, if possible

Seepage

- Develop a physical seepage model for each STA cell that is identified for in-depth seepage evaluation (Note: This could take significant effort to produce meaningful results)
- Conduct field assessment to locate any existing groundwater wells and analyze existing well data for each STA cell where applicable
- If no additional groundwater wells are located in the field assessment, then develop a detailed groundwater/seepage investigation program to provide the water and conservative constituent budget components
    - Installation of monitoring wells on transects that incorporate the levee, marsh, and levees on the other side of transect
    - Collection of transient water level and temperature data from groundwater wells
    - Collect quarterly ionic water quality samples for existing and future wells
    - Collection of in-situ seepage measurements taken during various operational and climatic events within each STA cell
    - Develop a conservative constituent budget for the STA that can be used to verify/calibrate the existing water budget; the conservative constituent budget would use chloride data from surface, groundwater and rainfall
    - This would be an iterative process to reduce uncertainty in the flow component
    - Apply conservative constituent budget to seasonal water budget to determine the largest losses to the system and to determine where the greatest uncertainties lie
    - Validate the flow component of the water budget with the conservative constituent budget
    - Collect isotope water quality samples for source water and age dating

7.5.3 Data Management

- Improved structure flow data will be stored in DBHYDRO (e.g., “MOD” DBKEYs).
- Water budgets and phosphorus budgets will be summarized using Excel spreadsheets which will be stored on the District’s server.

7.5.4 Reporting

- Quarterly Progress Report: FY2014–FY2020
- Annual Water and Phosphorus Budget Summary: FY2015–FY2020

7.5.5 Study Schedule

Phase I - Initiate	February 2013	Complete	September 2013
Phase II -Initiate	October 2013		
• STA-2 and STA-3/4 flow data Improvements		Complete	September 2014
• POR STA-2 & STA-3/4 Water & Phosphorus Budgets		Complete	September 2015
Phase III			TBD, if needed

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SECTION 8:

EVALUATION OF SAMPLING METHODS FOR TP

8.1 OVERALL STUDY PLAN SUMMARY

The objective of the Restoration Strategies Science Plan is to gather scientific information to reduce phosphorus discharge concentrations to meet WQBELs established in the NPDES permits for the Everglades STAs. Integral and fundamental to this objective is the proper collection of representative water quality samples from compliance stations and research programs. However, the unique use of autosamplers in South Florida as the primary method for collecting data for flow-weighted concentrations has been shown in general to produce slightly higher total phosphorus ($TPO_{4)}$ results than grab samples and, on occasion, can produce values that are significantly higher. These unexplainable differences are often not large, but they can be a substantial contribution to TP measurements at low concentrations, adding extraneous and unrepresentative noise to analytical values.

In order to meet the low TP concentration WQBEL discharge levels, it is essential that the Science Plan identify factors that may improperly bias results and begin the process of improving sampling regimes in the STAs and at other water quality monitoring sites to lessen the frequency and magnitude of extraneous noise. The Remote Environmental Sampling Test (REST) Project will collect significant amounts of water quality data using a variety of methods under tightly specified conditions to develop a robust database for the study sites, with supplemental information from deployed probes and cameras. Using these methods in concert, issues with each individual method and data stream can be isolated and offending factors identified. The ultimate goal of this project is to make recommendations on how to prevent, mitigate, or minimize for various factors that interfere with representative sampling such that the samples collected for environmental monitoring, compliance and research present the best possible estimate of water column TPO_4 in a cost-effective manner. REST does not have any preconceived goal of supporting one particular sampling regime over another. Rather, it seeks to produce information to optimize TP sampling across a range of environmental conditions and monitoring locations.

8.2 BASIS FOR THE PROJECT

Science Plan Questions/Sun-Questions Study Addresses

- What sampling regime provides the most accurate representation of TPO_4 in the target water column?
- What factors influence sampling results such that autosampler and grab samples are routinely produce different estimates of ambient conditions?
- If biasing factors are identified, can they or should they be addressed or mitigated?
- If biasing factors cannot be eliminated, can the impacted data be identified as unrepresentative?
- Are there alternative sampling regimes that can provide representative results in a cost effective manner that eliminate or minimize the impact of biasing factors?

8.3 BACKGROUND/LITERATURE REVIEW

In general, the District uses three primary methods to collect surface water samples: grabs (single point measurements), ACF (flow-proportional samples collected using a robotic pump and composited over approximately one week), and ACTs (time-proportional samples collected using a robotic pump and composited over approximately one week). Additionally, variations on these methods exist in which the composite sample can be done at different time steps, including hourly samples collected into a daily composite (ADTs). Variations in grab samples also exist and can include direct collection into the analytical bottle, use of a van Dorn, use of a peristaltic pump, and, in some cases, use of the robotic peristaltic pump for the collection of ACF samples.

Currently, the District uses the ACF method to create a single TPO_4 value for the entire week of flow, which is then used to generate an annual flow-weighted mean (FWM) concentration. However, this is not the only manner to generate a FWM. Other options include the use of grabs, ACTs, ADTs, and even flow-proportional samples collected over shorter timeframes and analyzed discretely. It is possible that one of these methods with or without modifications, or another yet to be implemented or explored would provide a more accurate representation of surface water TPO_4.

Over the past two decades, increasing numbers of autosamplers have been used in South Florida to estimate discharge concentrations, with grab samples often collected as backups for autosampler failure. Over the long term (e.g., months to years), the methods have been shown to produce very similar data or trends, although autosampler results tend to be slightly higher than grab values; the reverse situation is also known to occur. However, significant deviations in data from grabs and autosamplers have been documented for shorter periods at some stations. These short-term or ephemeral events, in which TPO_4 results from grabs and autosamplers vary widely, are of particular concern as they have the potential to impact long-term estimates of loads and concentrations and increase uncertainty in the process especially at low ambient concentrations.

It would not be possible or practical to review the extensive literature on sampling methods for water quality or list all the prior investigations conducted by the District on the differences between autosamplers and grabs. Yet, it is important to note the study findings by Millian, Swain and Associates, Inc. (2007), in which 10 time-proportional discrete autosamplers (ADTs) were deployed and samples were analyzed for TPO_4. Of the 10 stations, two did not show a statistical difference in concentration between the grab and the last bottle of the week or the weekly average. In contrast, the other eight stations showed either a difference between the grab and the last bottle or the weekly average. A load analysis for eight of the stations found that only four were significantly different statistically and, in all cases, grab calculated loads were lower than those calculated from autosamplers. This pattern was also reflected in the concentration data. However, statistical significance can be found with no real impact on the use of data for environmental management, and there are notable differences in the costs of sampling using different methods. Overall, the report acknowledged that grab and autosampler results were different, but provided no information about the key factors causing such deviations. Nonetheless, the generally held explanation is provided in the literature on urban stormwater monitoring:

> Data obtained from too few grab samples are highly variable, particularly for industrial monitoring programs, and subject to greater uncertainly because of experimenter error and poor data-collection practices. In order to use stormwater data that is scientifically defensible for decision-making purposes, it is more appropriate to replace grab sampling with more accurate and frequent, continuous sampling methods that are flow-weighted. Flow-weighted composite monitoring should continue for the duration of the rain event (NRC, 2009).

Similar conclusions were found by Ackerman et al. (2011). Volume-paced micro-sampling and targeted volume-paced sampling with analysis of discrete samples provide alternatives that improve accuracy without costing as much as pollutograph sampling. The common features of both these approaches are (1) use of volume pacing, not time pacings; (2) their ability to capture a range of different storm types (i.e., sizes and timing); and (3) their inclusion of multiple discrete samples. Other recent work has previously documented that volume-based sampling is more accurate than time-based because it provides a better representation of the overall storm (Leecaster et al., 2002; King et al., 2005; Ma et al., 2009).

Therefore, it is often concluded that flow-proportional monitoring provides the most accurate (meaning close to actual value) estimation of loading during stormwater events based on the fundamental notion that grab sampling programs have an insufficient number of samples at an inappropriate frequency to properly capture the action (e.g., Richards, 1998). With this in mind, when the District observes discrepancies in TPO_4 values between grabs and ACFs, the tendency has been to conclude that the ACF has appropriately characterized the TPO_4 for the week, although the in-situ conditions changed since the grabs were taken.

Ackerman et al. (2011) provide specific suggestions about how such sampling should be conducted. By targeting the volumetric pacings based on anticipated storm size, sampling is better able to capture a representative portion of the storm. Given the error inherent in weather predictions, it is preferable to overestimate (i.e., storm is smaller than expected) than to underestimate when setting the sample pacings. Although more costly, analyzing discrete samples as opposed to compositing into a single sample allows for better representation over the course of a storm, results in more accurate event mean concentrations (EMCs), and provides greater flexibility if the storm does not materialize as predicted.

Costs can be partially reduced by shortening the duration of sampling or reducing the number of discrete samples analyzed from ten to four. Because concentrations are typically higher during the early portion of storms, ending sampling when flow is 50 percent of peak flow can reduce costs with little overall effect on the accuracy of the EMCs because at that point the majority of the pollutant mass and storm volume have flowed pass the station. However, this requires accurate assessment of timing of peak flow in the field; inaccurate determination of peak flow may introduce additional bias.

This design of dynamic storm-based sampling is similar to that proposed by Abtew et al. (1997), which proposed autosamplers with variable trigger volumes based on the flow rates of structures during the current and preceding week. Ackerman et al. (2011) warned that the "volume-paced composite sampling resulted in overestimates of EMC, particularly for large storms." Indeed, that study suggests that the sampling strategy may have to be tailored for various types of runoff, particularly those with lower and longer amplitudes. This also raises a question concerning confounding factors not through inclusion, but rather by absence. Most of the studies on stormwater sampling focus on urban, riverine, or industrial systems, but the Everglades STAs are shallow wetlands with significant amounts of emergent, submergent, and floating macrophytes, periphyton, and algae (i.e., conditions not common in the studied systems). Also, wildlife—including fish, birds, amphibians, reptiles (turtles, snakes and alligators), and large mammals, all of which are common in STAs and could impact sampling equipment and water quality conditions—were likely absent from the referenced studies, particularly in urban and industrial settings. The impact of water body type, vegetative detritus, and wildlife on sampling equipment deployed over the long-term (months to years) is essentially unknown.

The literature suggests some important issues that must be addressed to assure that the flow-proportional monitoring is accurate. The assumption of studies advocating for flow-proportional monitoring is that the sampling is being done in response to short-lived storm events (hours to days) that have defined beginnings and ends, and that the flows and loads mimic that same cycle. South Florida's cycle of wet and dry seasons seem opposing to this design and, while storm events do occur in this subtropical region, the resulting flows tend to last for extended periods.

This condition may be exacerbated by how those flows are managed, essentially artificially metered, into and out of the STAs, which tend to flatten peak flow and extend the duration of the event.

Indeed, the volume-based program developed by the District for use in its ACF monitoring program is essentially insensitive to storm size and makes no attempt to define individual storm events. The triggers are set at volumes that assure that during high-flow events the samplers will not overflow but, at the same time, collect a sufficient number of samples to process and analyze. However, this regime means that no samples are collected for extended periods of low flow. Additionally, for extended periods (greater than a week) of high flow the storm event may be split between multiple samples. As such, the District's flow-based monitoring is neither dynamic (adjusted for storm size) or event-based (focused on definable storms). Unlike the systems described in the literature, which are dominated by punctuated storm events with definable beginnings and ends, the South Florida environment and the STAs can be viewed as a wet season of regular rainfall punctuated with higher flow events from localized and regional storms, followed by a dry season. This difference also results in a system that attracts and sustains biological communities that are not present in more controlled settings. Consequently, while it is tempting to assume that the differences in ACF and grab samples are solely attributable to differences in sampling frequency, there are other factors that must be examined.

8.4 DATA ANALYSIS SUPPORTING THE STUDY

An overarching flaw in the literature on water quality sampling studies is that they usually compare grabs with some form of automated, flow-proportional sampler and assume that more flow-paced data is a better representation of ambient concentrations. In most cases, dynamic pollutographs will be better estimated with more frequent flow-paced data. However, South Florida's unique canal based system does produce dynamic hydrographs. In this system, it is more likely that the higher values often generated by autosampling are the result of extraneous, unrepresentative sample collections. The District now has an advanced system that can be expected to produce highly representative data for use in this sampling study to better deal with the problem of extraneous values.

Over the last several years, the District has been testing a new method for sampling and analyzing TPO_4 using a Remote Phosphorus Analyzer (RPA), which allows TP concentration data to be generated continuously in the field. Analysis of the method, which allows the deployment of automated chemistry laboratories into the field, has found the results for TPO_4 to be comparable to the results from traditional sampling and analysis (Struve et al., 2008). Using the RPAs has provided TPO_4 data at several stations at a three-hour time step. The sheer volume of such data allows for the detailed analysis of weekly grabs and ACFs in a new light and highlights some interesting and relevant events.

Several examples of TP sampling help to illustrate problems with each sampling approach. However, it is important to note that environmental monitoring is for long-term trends and annual compliance values and there a large differences in costs among methods. **Figure 8-1** shows the daily flow and TPO_4 results from grabs, ACF, and RPA at G310, one of the discharges from STA-1W. At the beginning of the two-week period, the RPA, ACF, and grab had results ranging from 40 to 50 µg/L. As the week progresses, the flow reaches zero and then picks up slightly by the second sampling date. At this time, the TPO_4 from the RPA rose to a range of 50-60 µg/L, the grab sample was 69 µg/L, and the ACF reported 41 µg/L. Subsequently, there was a slight rise in flow while the RPA data dropped, with two aberrant sampling events—one at 68 µg/L and the other at 47 µg/L. The period ends with RPA data between 25-35 µg/L, the grab sample at 37 µg/L, and the ACF at 47 µg/L.

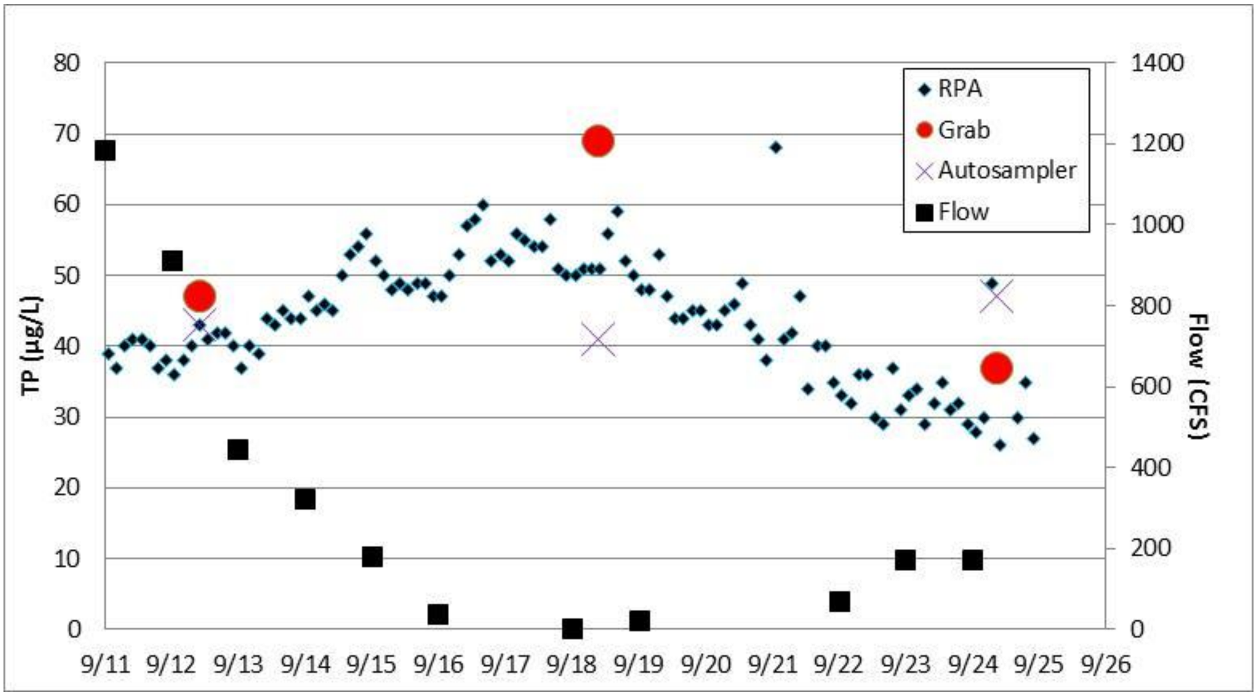

Figure 8-1. A snapshot of daily flow and TP results at G310.

The three sampling methods appear to be in agreement during the first sampling event, but that agreement doesn't hold during the second and third weeks. However, a careful review of the flow data, the sampling triggers and the RPA data can help us understand what is going on in these apparently inconsistent results. The first issue to understand is the difference between the RPA and the grab data. It must be understood that the grab and the RPA samples are not taken at the same time and they are processed and analyzed using different methods and reagents. Consequently a slight difference between the results, and one that in this case suggests a slightly positive bias in the grab is to be expected and the differences as presented here are likely not significant.

The ACF data is more troubling but equally explainable. Through the course of the week prior to the second sampling, the TPO_4 (according to the RPA) increased but the flow decreased. As a result, the samples that make up the composite in the ACF are weighted toward the beginning of the sampling period during higher flow when the RPA was reporting around 40 µg/L. Calculation of the flow-weighted mean from the RPA data for the same period produced a value of 46 µg/L. Therefore, while it appears that the values contradict each other, they in fact support each other. Taken on its own, this week of data supports the thought that weekly grab samples may be too infrequent to document a flow event. Indeed, as an artificial storm event this data set suggests that as a rule of thumb if flow has decreased during the course of the week, the ACF should be more related to the sample from the week before, while if flow has increased during the sampling period the ACF and grab from the same week should be more related, as the ACF samples are biased in either temporal direction by the weight of flow through time.

The last set of results may be just as easy to understand, but more troubling. It is tempting to look at the RPA data for the week and then explain the third ACF event of 47 µg/L as a result of the water quality values from September 18 to 21, which according to the RPA ranged from

60-40 µg/L, with one value at 68 µg/L. However, there was low flow during this period and the ACF did not trigger until the morning of September 24 and then only collected three samples. Whatever those three values were, it seems apparent they were highly influenced by the same event that showed up in the RPA and produced a result of 49 µg/L. Whatever occurred that impacted the RPA and the autosampler severely skewed the weekly results, calculation of the flow weighted mean for the week using RPA data produced a result of 31 µg/L. Additionally, it should be noted that during this period, 75 percent of the flow had to pass through the structure before the ACF activated, highlighting the concern that ACFs designed for storm-event monitoring may not be properly programmed to sample low and long amplitude events that have poorly defined beginnings and ends. This failure to collect combined with an ephemeral event that lasts for less than six hours severely biases the ACF data for the entire week.

Another ephemeral event can be observed in data from S362 in STA-1E during October 2011 (**Figure 8-2**). In this case, the RPA TPO_4 data is relatively stable between 15 to 25 µg/L, with one value dropping to almost 10 µg/L and another at 41 µg/L. The factors that influenced the RPA to reach 41 µg/L for less than six hours also appear to have caused the ACF to report a value of 35 µg/L, while the grab and FWM concentrations calculated from the RPA data were both below 20 µg/L.

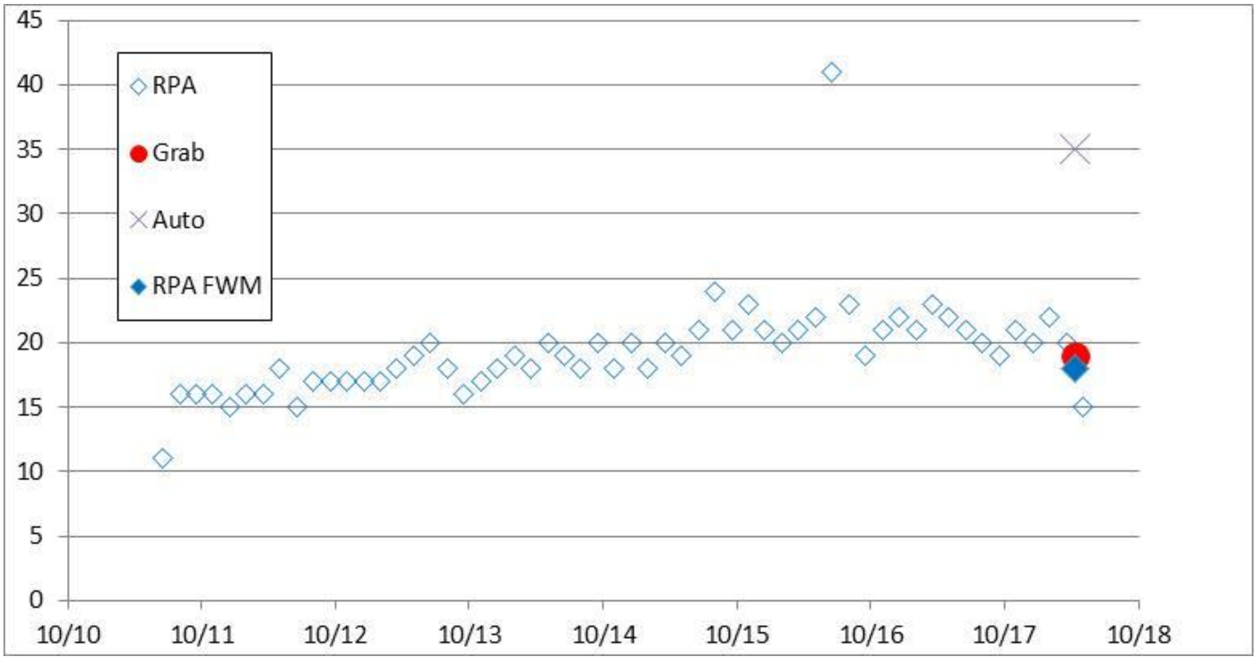

Figure 8-2. A snapshot of TP results at S362.

A third example is illustrated by data from G302, the inflow to STA-1W (**Figure 8-3**). In this case, rapid sudden flows near the end of the sampling period trigger the ACF quickly, but the sudden decrease in TPO_4 values are not captured. Consequently, the ACF reports a value of 250 µg/L, but the calculated FWM concentration from the RPA is only 162 µg/L, similar to the value reported from the grab. In this example, there is no indication in the RPA data that the data reached up to 250 µg/L or higher during the flow event, as the highest value reported is 233 µg/L. It is possible that the difference is simply a result of analytical and sampling variation, or it may be that the events that resulted in higher TPO_4 concentrations occurred between the three-hour RPA sampling events and therefore were not captured by the RPA.

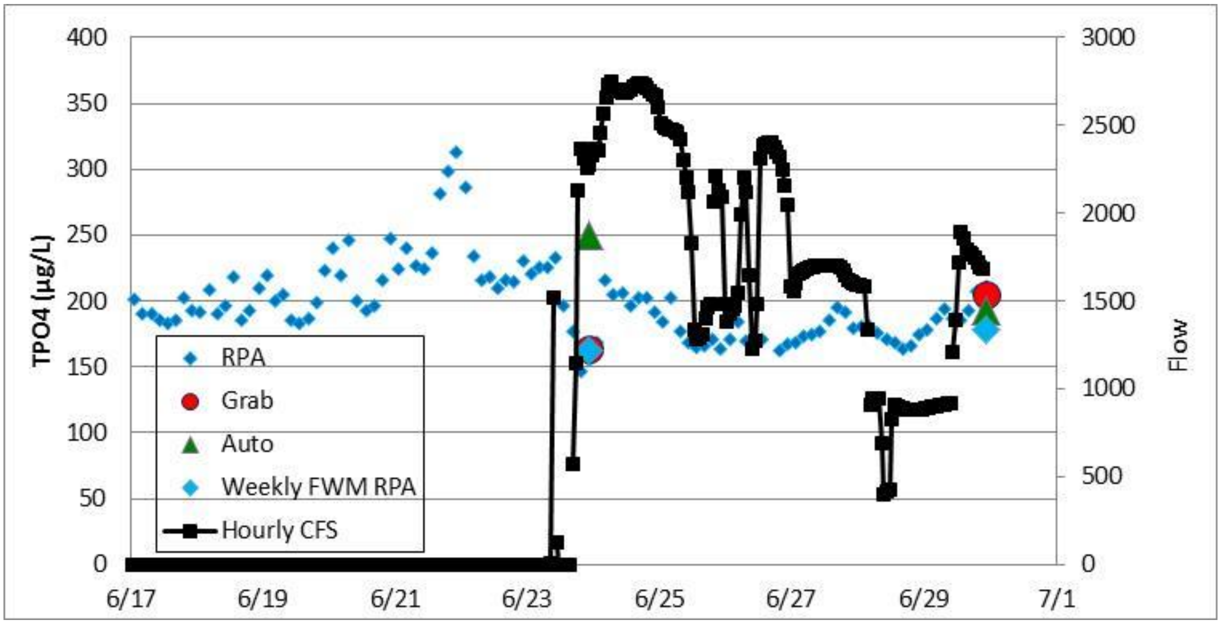

Figure 8-3. A snapshot of TP and flow results at G302.

Overall, these examples suggest that during some sampling events factors other than sampling frequency are creating differences among the resulting values from various sampling methods.

8.5 STUDY PLAN OBJECTIVES

8.5.1. Specific Hypotheses to be Tested

- **Ephemeral events are driven by detritus:** It is hypothesized that the ephemeral events occasionally captured by the RPA are responsible for the differences between ACF and Grab sample results, and that these ephemeral events are caused by floating detritus that generates patchiness with impacts the sample estimates.
- **Ephemeral events are driven by biota:** It is hypothesized that the ephemeral events occasionally captured by the RPA are responsible for the differences between ACF and Grab sample results, and that these ephemeral events are caused by biota (fish, reptiles, birds, mammals) that interact directly or indirectly with the sampling equipment, possibly through waste products that generate unrepresentative patches in the water column.
- **Ephemeral events are driven by physiochemical processes:** It is hypothesized that the ephemeral events occasionally captured by the RPA are responsible for the differences between ACF and Grab sample results, and that these ephemeral events are caused by plumes or conglomerations of sediment that impact the sampling environment for very brief periods of time.
- **Ephemeral events are driven by anthropogenic activities:** It is hypothesized that the ephemeral events occasionally captured by the RPA are responsible for the differences between ACF and Grab sample results, and that these ephemeral events

are caused by anthropogenic activities including site maintenance such as mowing and application of herbicides. Many herbicides are phosphorus based and can therefore influence the sampling environment directly, but the death and decay of plant material can also have indirect impacts.

- **Ephemeral events are driven by hydrologic variations:** It is hypothesized that the frequency and magnitude of ephemeral events are correlated with flow regime. Concentration spikes at high flows are expected to result from P transport thru the upstream STA marsh, although sampling problems may also be a factor. Unexplained concentration spikes at low flows are more likely to be related to artifacts or local phenomena. Loads created by small disturbances, local biota, contamination, etc. have less impact on concentrations at high flows because loads are dominated by P transport thru the marsh and the ephemeral loads are diluted by the high flows.

8.5.2 Alternative Hypotheses

- **Flow-based monitoring is biased by improper design:** It is hypothesized that the ACF data is biased by the improper setting of flow triggers, the failure for these triggers to be dynamic, and the temporal parsing of storm events across multiple sampling events.
- **Flow-based monitoring is biased by biofouling organisms:** It is hypothesized that the ACF data is biased by the growth of biofouling organisms such as periphyton on the tubing of the sampler itself and that periodic elevations in ACF data can in part be attributed to the periodic sloughing off of these communities.

8.5.3 Potential Management Implications

The results of the study could have significant implications for site management and maintenance, or policy. For example, if the results suggest that the cause is a controllable factor, it may be necessary to develop or install new devices to exclude detritus, birds or other wildlife, or change/improve maintenance. Alternatively, if changes in turbidity or specific conductivity are found to act as indicators of temporary changes in the data quality, it may be possible to exclude the impacted data from use. Sampling locations may need to be modified, as might trigger volumes. In extreme cases outflow structures might need to be redesigned. Finally, it might be possible to develop a new sampling strategy that still provides a flow proportional TPO_4 result, but lessens the impact of individual ACF events on the total value for the week. Many of these actions would have to be discussed with regulatory authorities and stakeholders.

8.6 DETAILED STUDY PLAN AND EXPERIMENTAL DESIGN

8.6.1 Study Plan Description

The purpose of this study is to determine the monitoring strategy that provides the most accurate results for water quality, and to ascertain what factors may improperly influence results, and if those factors need to be, or should be mitigated for. In order to accomplish this, routine monitoring of a site using ACF and Grab samples will be augmented with and ADT, an RPA, a set of in situ probes, and high frequency cameras.

The ADT will collect samples at set intervals and create an average TPO_4 value for each day, which can then be used in conjunction with daily flow data to develop a FWM to simulate and validate the value from the ACF.

The RPA data will provide instantaneous TPO_4 values at set intervals which can be used to simulate and validate the weekly grab, the weekly ACF value, the daily values from the ADT, and the simulated FWM calculated from the ADT.

In addition to the routine grab samples, grabs collected using the long-term deployed sampler tubing will be collected weekly. During quarterly replacement of the tubing, the fresh tubing will also be tested.

8.6.2 Basis for Design

Literature Influencing Design

The suggestion by Ackerman et al. (2011) that different types of flow might require different types or frequency of monitoring is fundamental to the design of this study. Additionally the concept of efficiency, a balance between data accuracy and cost, as discussed by Leecaster et al. (2002), must be acknowledge as an influence in the design of some of the analyses. With both of these issues in mind part of the evaluation of the data will include the use of RPA data to simulate alternative collection regimes at various time steps for compositing, and then an evaluation of the costs associated with each alternative.

Historical Data Influencing Design

As shown in previously presented RPA data, ephemeral events appear to be very short lived and may be manifest only as a single data point. As these data were collected at three hour intervals, they suggest that whatever caused these events may last less than six hours. Consequently, it has been suggested that the RPA period be set to a higher frequency. However, the cycle time for the RPA varies around a one hour interval and makes collecting hourly samples difficult. It is therefore suggested that a two hour interval be used and mimicked in the ADT to make comparisons easier. Additionally, the two hour window allows a sufficient amount of time for daily calibrations and equipment maintenance during weekly sampling events.

Locations and Rationale

This study is sampling intensive and has the potential to have to respond adaptively to field conditions. Consequently, it has been decided to leverage existing monitoring stations. Although this project has the potential to be applicable throughout the District, the highest priority stations are those at the discharges from the STAs where low TPO_4 values are prevalent. Toward this end it was decided to focus on sites at or close to STA discharges. The primary test site is G310 one of the outflows from STA-1W. The other station is G390B the inflow to the PSTA project in STA-3/4 which experiences relatively low TPO_4 values. While it would be possible to carry this out at other stations throughout the District, it is suggested that the lessons learned from these two sites be developed and transferred before repeating elsewhere.

8.6.3 Experimental Design

The design of the project hinges on the collection of samples using multiple methods, the comparison of those methods, and observations of the conditions during collection obtained using cameras and deployed probes. The use of an intensely monitored station to develop a massive database of TPO_4 values, physical measurements and images will allow for an unprecedented review of field conditions at a very frequent time step. These data will then be used to essential cross check individual values and understand the factors that may be impacting monitoring which have to date gone unobserved. Once critical issues are discovered and their impact evaluated options for remediation including changes in monitoring and monitoring regimes will be discussed.

8.6.4 Study Plan Components

The study consists of deploying multiple pieces of equipment at a given station. The deployed equipment can be summarized as follows:

- *G (Grab Sample).* This sampling method uses a minimum amount of equipment and a rather infrequent time step for sampling (weekly).
- *ACF (Autosampler Composite Flow).* This sampling method uses a robotic pump triggered on a set volume to fill a single container which is collected and processed every week. Under normal operations only the first trigger is recorded, for this study all triggers will be recorded.
- *ADT (Autosampler Discrete Time).* This sampling method uses a robotic pump triggered on time to fill a bottle at a set interval for an entire 24-hour period, and then shift to a new bottle at the start of the next day. For the study duration, the ADT will start sampling at 0100 each day and sample every 2 hours until 2300, creating 12 samples per day. On the day of sampling, two partial days will be generated. It would be preferable that the sampling occur after 1100 and before 1300.
- *GP-ACF (Grab Pumped via the ACF).* This sampling method uses the robotic pump from the ACF to manual collect a grab sample. This will be compared to the G and document the impact of long-term deployment of tubing on sampling results.
- *GP-ADT (Grab Pumped via the ADT).* This sampling method uses the robotic pump from the ADT to manual collect a grab sample. This will be compared to the G and document the impact of long-term deployment of tubing on sampling results.
- *NT-ACF (New Tubing for the ACF).* This sampling method uses a pump to manual collect a grab sample through the new tubing being deployed for the ACF. This will be compared to the GP-ACF and other samples to document the impact of long-term deployment of tubing on sampling results.
- *NT-ADT (New Tubing for the ADT).* This sampling method uses a pump to manual collect a grab sample through the new tubing being deployed for the ADT. This will be compared to the GP-ADT and other samples to document the impact of long-term deployment of tubing on sampling results.
- *RPA (Remote Phosphorus Analyzer).* This sampling method uses a pump and deployed laboratory to collect and analyze samples based on time. For the study duration, the RPA will start sampling every 2 hours on the odd hour, and a single

day will be considered from 0100 until 2300, creating 12 samples per day, the same as the ADT.

- *P (Probe).* This sampling method uses a deployed set of probes to collect data on Turbidity, Specific Conductance, pH and Temperature. For this study, the sampling frequency will be every 15 minutes.
- *C-UW (Camera Underwater).* This method aims an underwater camera at the intakes and documents the conditions with a still image every 15 seconds.
- *C-SW (Camera Surface Water).* This method aims a camera at the area above the sampling point and documents the conditions with a still image every 5 minutes, or in association with movement.
- *C-PF (Camera Platform).* This method aims a camera at the area around the sampling platform and documents the conditions with a still image every 5 minutes, or in association with movement.

8.6.5 Methods

Task 1A: Sampling Equipment Installation

- Install sampling equipment including autosamplers and RPAs

Task 1B: Additional Equipment Installation

- Install other monitoring equipment including cameras (3) and probes (1 set)

Task 2: Installation Evaluation

The basic assumptions of comparing method-specific results are that initially the samples are collecting the same water and the results are comparable. To test these assumptions an intensive sampling regime on the first day of deployment will be initiated. A summary of water quality monitoring for Task 2 is presented in **Table 8-1**.

- *Step 1. Blank Testing* – All equipment used will follow standard quality assurance (QA) protocols as detailed in the District's FSQM. Samples will be analyzed for TPO_4 only.
- *Step 2. Set up of Field Equipment* – The position of the sampling intake for the ACF autosampler will be used as the base for establishing position of the intakes for the time proportional autosampler (ADT) and the RPA, so that the intake points are as close as possible to each other.
- *Step 3. Field Test* – The RPA will be set to collect every 1.5 hours for 8 hours. Both the ADT and ACF will be used to collect discrete samples (60 ml) every 45 minutes for eight hours (This will require taking the ACF off line). Grab samples will be collected 45 minutes for 7.5 hours. Grab samples will be collected as close as possible to the sampling intakes. Using this design will generate 11 samples for the grabs and autosamplers each, collected simultaneously, and 6 samples for the RPA.
- *Step 4. Data Evaluation* – The equipment blank results for all three methods must be less than the method detection limit (MDL). The data will be grouped into sets of based on the time of collection and be analyzed for equivalency. A difference of <20 percent from the mean will be used as a target to indicate acceptance for each set of

results. For each set of comparisons 1 negative result is allowed. More than one negative result will indicate failure and will require repetition of Tasks 1A and 2.

Table 8-1. Summary of water quality monitoring for Task 2.

Equipment	Parameter	Frequency for 8 Hours	Analytical Method	Total Number of Samples
RPA	TPO_4	Hourly	In situ experimental	8
ACF offline	TPO_4	Semi-hourly	SM4500PF	15
ADT offline	TPO_4	Semi-hourly		15
Grab	TPO_4	Semi-hourly		15
QA (6)	TPO_4	Once		6

<u>Task 3: 13-Month Data Collection Effort</u>

A summary of water quality monitoring for Task 3 is presented in **Table 8-2**.

- *Step 1. ACF autosampler Operation* – The existing, unrefrigerated ACF autosampler will be run with no modifications. However, existing protocol has the District recording the time of the first trigger event. For the purposes of this study, all trigger times will be recorded. Weekly, the ACF autosampler tubing and pump also will be used manually to collect a grab sample for TPO_4 (GP-ACF).
- *Step 2: ADT autosampler Operations* – The unrefrigerated ADT autosampler will be set to collect every two hours on the hour, with 12 consecutive samples (0100 to 2300) composited into a single bottle. This should create 8 samples per week (six full samples, and two half samples for each day of collection). It is preferable that the samples be collected after 1100, but well before 1300 so that the ½ bottles can be averaged together to create a value for the day. Weekly, the ADT tubing and pump also will be used manually to collect a grab sample for TPO4 (GP-ADT).
- *Step 3: Grab Sample Operations* – Weekly grab samples will be collected using a Van Dorn. Collection must be timed to coincide with both the GP-ACF and GP-ADT so that temporal changes in the water column are minimized. Samples must be collected as close to the intakes as possible.
- *Step 4: RPA Operations* – The RPA will be set to collect data every two hours on the hour, with 12 consecutive samples (0100 to 2300).
- *Step 5: Quarterly Tubing Maintenance* – Quarterly, autosampler tubing must be changed. Prior to doing the change tasks 1b, 2b, and 3a will be carried out. Additionally, the two new lengths of tubing will be used to collect pumped grab samples (NT-ACF and NT-ADT) at the same time (or at least within 15 minutes of each other). After collection, the new tubing lengths will be installed.
- *Step 6: Digital Imagery* – The underwater camera will be set to collect images every 15 seconds; wildlife cameras will be set to collect images every 5 minutes or on

motion; and digital imagery will be downloaded weekly. Equipment will be cleaned and maintained weekly.

- *Step 7: Probes* –Data from probes will be downloaded weekly. Equipment will be cleaned and calibrated weekly.

Table 8-2. Summary of water quality monitoring for Task 3.

Equipment	Parameter	Frequency	Analytical Method	Total Number of samples
RPA	TPO_4	Every two hours		4,800
ACF	TPO_4	Flow-based composited weekly	See Field Sampling Quality Manual (SFWMD, 2011)	60
ADT	TPO_4	Every two hours composited into daily bottles		400
Grab	TPO_4	Weekly		60
GP-ACF	TPO_4	Weekly		60
GP-ADT	TPO_4	Weekly		60
NT-ACF	TPO_4	Quarterly		4
NT-ADT	TPO_4	Quarterly		4
Probes	pH, turbidity, specific conductivity, temperature	Every 15 minutes	See Field Sampling Quality Manual (SFWMD, 2011)	38,500
C-UW	Image	Every 15 seconds	NA	2, 304,000
C-SW	Image	Every 5 minutes or on motion		115,200+
C-PF	Image	Every 5 minutes or on motion		115,200+
QA				

8.6.6 Data Management

The vast majority of the data will be stored in DBHYDRO or DCVP, and these data will go through standard District data QA procedures. During data analysis, all data will be screened to ensure that aberrant values are detected and causes determined. Any problematic values will be dealt with as appropriate after investigation and reported in the database. Such values will not be used for data interpretation without justification. Data from the RPA will be handled and archived. Data transformations, calculations, interpretations, and presentations will be reviewed by the project team before presented to the District and stakeholders.

8.6.7 Reporting

Quarterly progress reports will be produced summarizing work to date, issues, data, and critical observations. In instances where deviations between sampling method results occur a preliminary analysis and interpretation may be carried out.

The final report will consist of sections on individual methods, comparison of methods, factors driving differences, other observations, suggestions for remediation, cost-benefit analysis, and alternative sampling plans.

- **Evaluation of sampling methods using unscreened data**. The primary evaluation of sampling methods will be in the ability of the RPA data to reproduce results produced by the ADT and the ACF. This will be accomplished by using the sampling times from the ACF and ADT to select data from the RPA which will then be used to mathematically reproduce the results seen in the other sampling methods. In this manner the use of the RPA data can be validated. For results that deviate by more than 10 percent a review of individual data points to identify suspect values will be undertaken. Development of cumulative load graphics may be especially helpful in this regard. The data analysis and interpretation process is outlined in **Figure 8-4**.
- **Supplemental monitoring**. Once suspect values are identified, the supplemental data will be reviewed to offer up potential causation, and develop a screening tool possibly based on changes in turbidity or conductance. This screening tool will be used to qualify data and eliminate it from analysis.
- **Evaluation of sampling methods screened data**. Data will be used to re-develop the mathematically simulations that were developed using the RPA data for comparison to the ADT and ACF results.
- **Development of alternative sampling regimes**. Using the RPA data, alternative strategies for sampling based on time, flow and combinations of the two will be developed and evaluated for benefits, costs, uncertainty and risks
- **Recommendations**. Will include estimates of costs, benefits, uncertainty, and failure rates for the most promising methods.

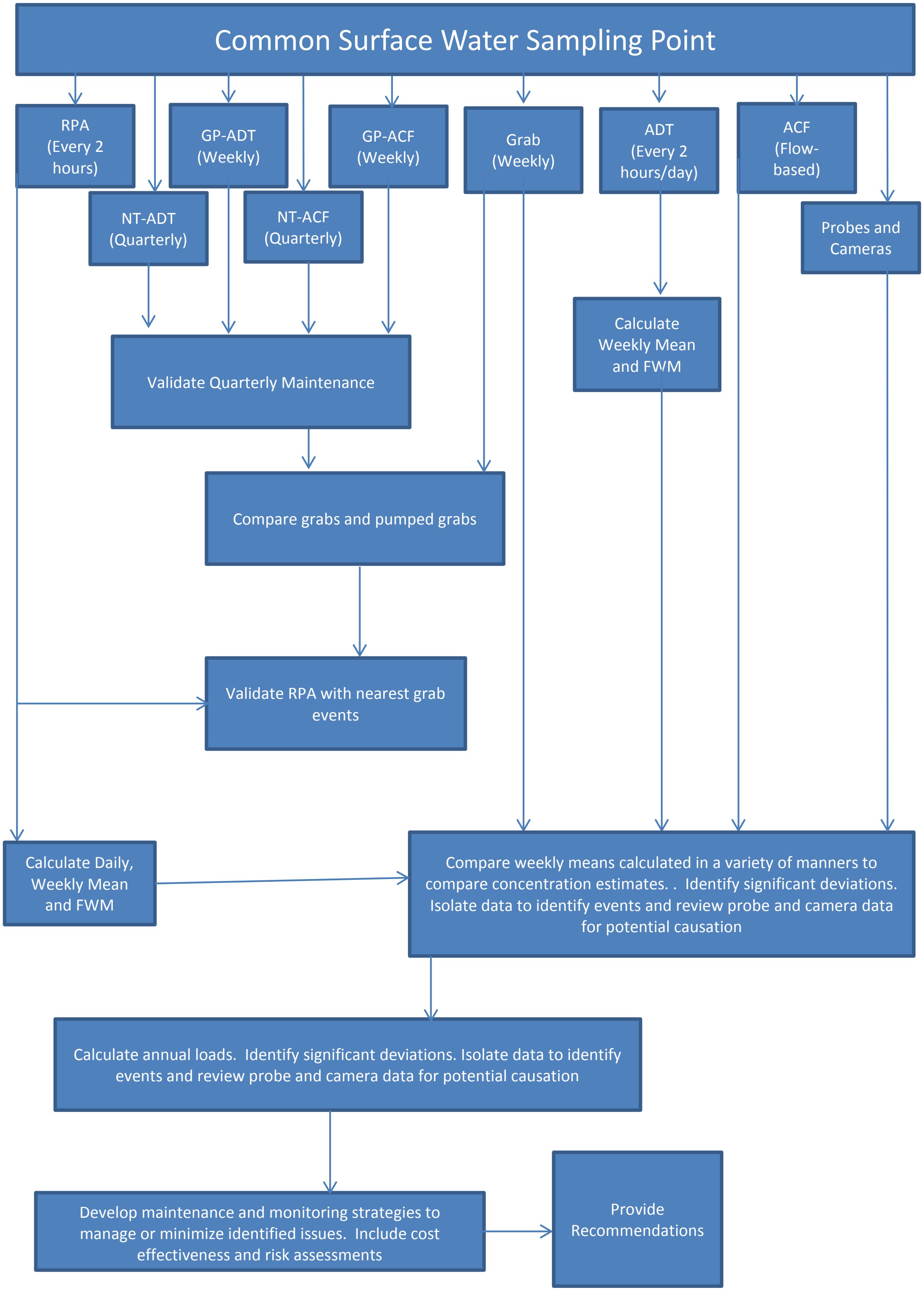

Figure 8-4. Overview of data analysis and interpretation process.

8.6.8 Study Schedule

Task 1 Sampling Equipment Installation	Initiate FY2013	Complete FY2014
Task 2 Installation Evaluation	Initiate FY2014	Complete FY2014
Task 3 Data Collection Effort	Initiate FY2014	Complete FY2015
Task 4 Analysis	Initiate FY2014	Complete FY2015

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