Appendix 4-2

Proposed Everglades Marsh Dissolved Oxygen Site Specific Alternative Criterion Technical Support Document

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Section 1.0 Introduction

The Everglades Forever Act (EFA, Section 373.4592(1)(a), Florida Statutes) passed by the Legislature in 1994 finds "*that the Everglades ecological system not only contributes to South Florida's water supply, flood control, and recreation, but serves as the habitat for diverse species of wildlife and plant life. The system is unique in the world and one of Florida's great treasures. The Everglades ecological system is endangered as a result of adverse changes in water quality, and in the quantity, distribution, and timing of flows, and, therefore, must be restored and protected.*" The EFA (Section 373.4592(4)(e), F.S.) directs the Florida Department of Environmental Protection (Department) and the South Florida Water Management District (SFWMD) to evaluate existing water quality standards applicable to the Everglades Protection Area (EPA) and Everglades Agricultural Area (EAA) canals. Furthermore, the EFA specifies that "*the Department's evaluation of any other water quality standards must include the Department's antidegradation standards*".

The 1999 Interim Report (Bechtel *et al.*, 1999) and 2000 Consolidated Report (Bechtel *et al*., 2000) provide ongoing annual assessments of the water quality status in the Everglades based on the concentrations of a broad array of monitoring parameters at a network of 56 monitoring stations. Among a list of nine parameters identified in these reports as in need of further review based on excursions from current standards, dissolved oxygen stood out as the most pervasive and potentially problematic. Dissolved oxygen concentrations below the current Class III dissolved oxygen standard of 5.0 mg/L (Chapter 62-302, F.A.C.) commonly occur in all areas of the EPA, including sites minimally impacted by nutrient enrichment or cattail invasion. These frequent excursions below the dissolved oxygen standard are not unexpected since it is widely recognized that dissolved oxygen concentrations in macrophyte dominated wetlands are normally low due to natural marsh processes.

Oxygen is a necessity for most life on Earth and all aerobic life, including plants and animals. Fish and macroinvertebrates are largely dependent upon oxygen dissolved in the water, although many species possess adaptations to allow them to obtain oxygen from other sources. For instance, Rader (1994) found that many macroinvertebrates in the Everglades have the ability to utilize atmospheric oxygen. Nonetheless, the nature of the Everglades dissolved oxygen regime has been an important selection factor influencing species colonization and overall community structure. Most of the fauna of the Everglades are adapted to these periodically low oxygen concentrations and can survive short periods of anaerobiosis. However, successful colonization by species that are sensitive to low dissolved oxygen levels is restricted by the natural dissolved oxygen regime, with greater limitations occurring in impacted areas that exhibit further depressed oxygen levels. Due to the importance of dissolved oxygen to the flora and fauna, it is necessary to have an understanding of the complex array of factors influencing dissolved oxygen levels in the Everglades marshes.

In any aquatic system, water column dissolved oxygen concentrations are regulated by a variety of sinks and sources. In a healthy environment, these controlling factors are balanced. **Figure 4-2-1** summarizes the major oxygen fluxes operating in a typical marsh environment. The primary oxygen sinks include chemical oxidation and aerobic respiration by vegetation, periphyton, and other organisms in both the water column and sediments. Under natural conditions respiration by periphyton and submerged aquatic vegetation (P/SAV) is the largest sink, followed by sediment oxygen demand (SOD) (Belanger and Platko, 1986; McCormick and Laing, in Review). Advective transport between oxygen rich sloughs and dissolved oxygen poor emergent macrophyte stands is a largely unquantified, yet important component of the marsh oxygen budget. (McCormick and Laing, in Review).

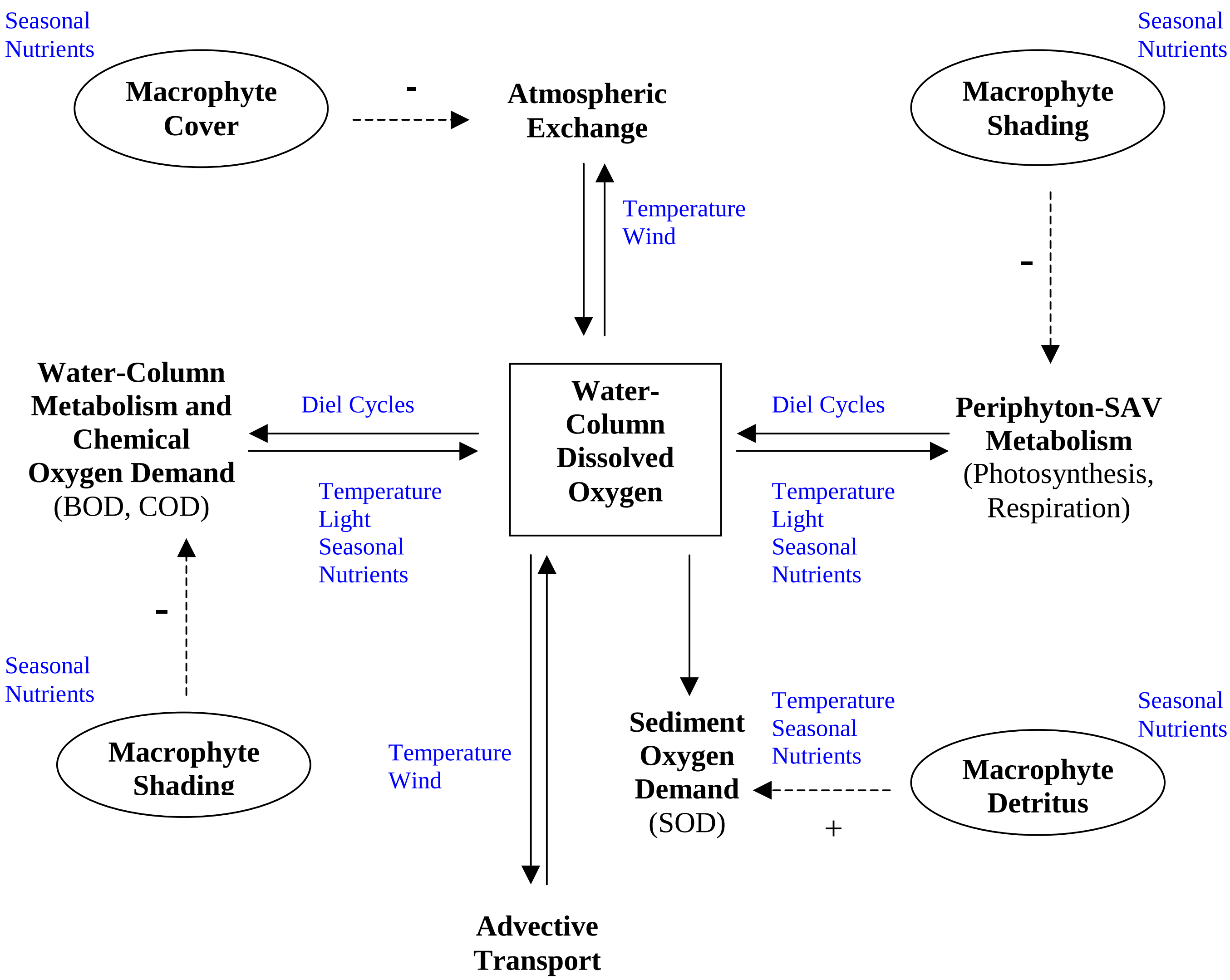

Figure 4-2-1. Conceptual model of major oxygen fluxes affecting water-column dissolved oxygen concentrations in wetlands. Solid arrows show the direction of the fluxes. Dashed arrows and associated symbols show the effects of emergent and floating macrophyte cover. The direction and/or magnitude of each flux is influenced by environmental factors such as season, temperature, wind speed, light (intensity and photoperiod), time of day (diel cycles) and nutrient enrichment. The blue text indicates major environmental factors acting on the given flux. Model was modified from McCormick and Laing (in Review) with permission from the authors.

The primary sources of water column oxygen are photosynthesis and atmospheric exchange. In a marsh habitat the primary photosynthetic contributors are P/SAV in open water sloughs with some minor contribution from phytoplankton. In contrast to the slough community, emergent and floating leaf vegetation contribute little oxygen to the water column and also affect dissolved oxygen levels negatively by shading underlying vegetation and retarding atmospheric exchange (Rose and Crumpton, 1996; Brandenburg, 1996; McCormick and Laing, in Review). Depending on several factors, atmospheric exchange can either be a source or sink. The rate and direction of atmospheric exchange is dependent upon dissolved oxygen levels in the water column (*i.e.*, percent of saturation based on solubility), temperature, and water agitation (*e.g.*, wave action). The maximum quantity of dissolved oxygen that can be held in solution (*i.e.,* saturation concentration) is controlled by the solubility of oxygen in water. Dissolved oxygen saturation concentration and solubility are inversely related to temperature and chlorinity or salinity of the water (APHA 1992). That is, less dissolved oxygen can be maintained under conditions of higher temperature and salinity than under cooler lower salinity conditions. Under certain conditions, dissolved oxygen concentrations in the water column can exceed the saturation concentration. Within Everglades sloughs P/SAV photosynthesis occasionally exceeds atmospheric exchange and demands (sinks), causing supersaturation of the water column and oxygen bubble formation for short periods.

The rate of photosynthetic production is largely dependent upon light intensity and occurs only during the photoperiod (daylight). In open water slough communities, where light penetration is adequate, photosynthesis by P/SAV typically results in increasing oxygen concentration during daylight hours (Belanger and Platko 1986; McCormick *et al.*, 1997). At night, after photosynthesis has slowed, respiration and sediment oxygen demand (SOD) reduce oxygen concentrations until the next morning when the cycle starts again. The balance between photosynthesis and respiration during the day produces a characteristic diel dissolved oxygen curve (**Figure 4-2-2**). Beyond the factors already discussed, the magnitude of the natural diel curve can also be altered by

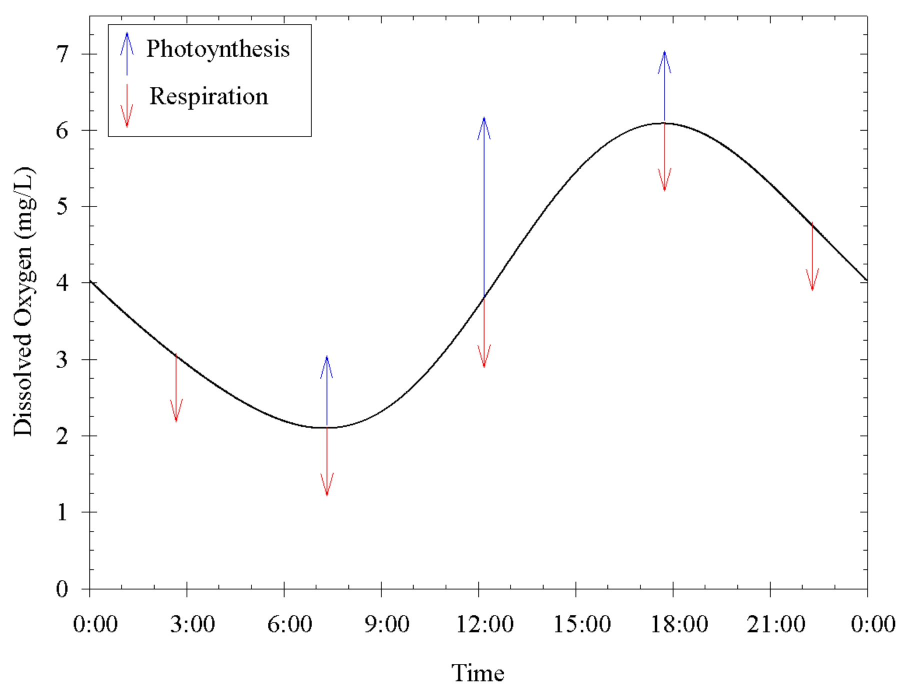

Figure 4-2-2. Typical diel dissolved oxygen curve. The blue (up) and red (down) arrows show the generalized influences of photosynthesis and respiration, respectively. Arrows are not scaled, but are meant to indicate the relative influences of photosynthesis and respiration as

human-induced impacts to the system such as nutrient enrichment.

Nutrient enrichment typically produces a variety of system changes that ultimately result in a depressed water column oxygen regime. The Everglades is a highly phosphorus limited system and as phosphorus levels are increased productivity increases, as does organic matter accumulation in the water and sediments (*i.e.*, increased BOD and SOD). This increased demand for oxygen results in greater oxygen consumption in both the water column and sediments. In addition to an altered community metabolism, changes in vegetation occur in response to eutrophication. Initially and/or under a moderate level of enrichment, there is typically a replacement of the native peripyton-*Utricularia*-*Eleocharis* community by dense water lily (*Nymphaea*) marshes (McCormick and Laing, in Review). As the degree of eutrophication (intensity or duration) increases, the open water slough communities are invaded by cattail (*Typha*) and other emergent vegetation including sawgrass (*Cladium jamaicense*) (Rutchey and Vilchek, 1994; McCormick *et al.*, 1998). The loss of the P/SAV complex and shading (reduced light penetration) resulting from increased growth of broadleaf floating and emergent vegetation reduces the photosynthetic production of oxygen in the water column (Brandenburg 1996; McCormick and Laing, in Review). Ultimately, the phosphorus enrichment results in an imbalance in the oxygen sources and sinks by increasing the demand for oxygen and reducing the photosynthetic oxygen production. Therefore, the diel dissolved oxygen curves for phosphorus enriched areas are characterized by overall lower dissolved oxygen levels and dampened diel fluctuations as depicted in **Figure 4-2-3**.

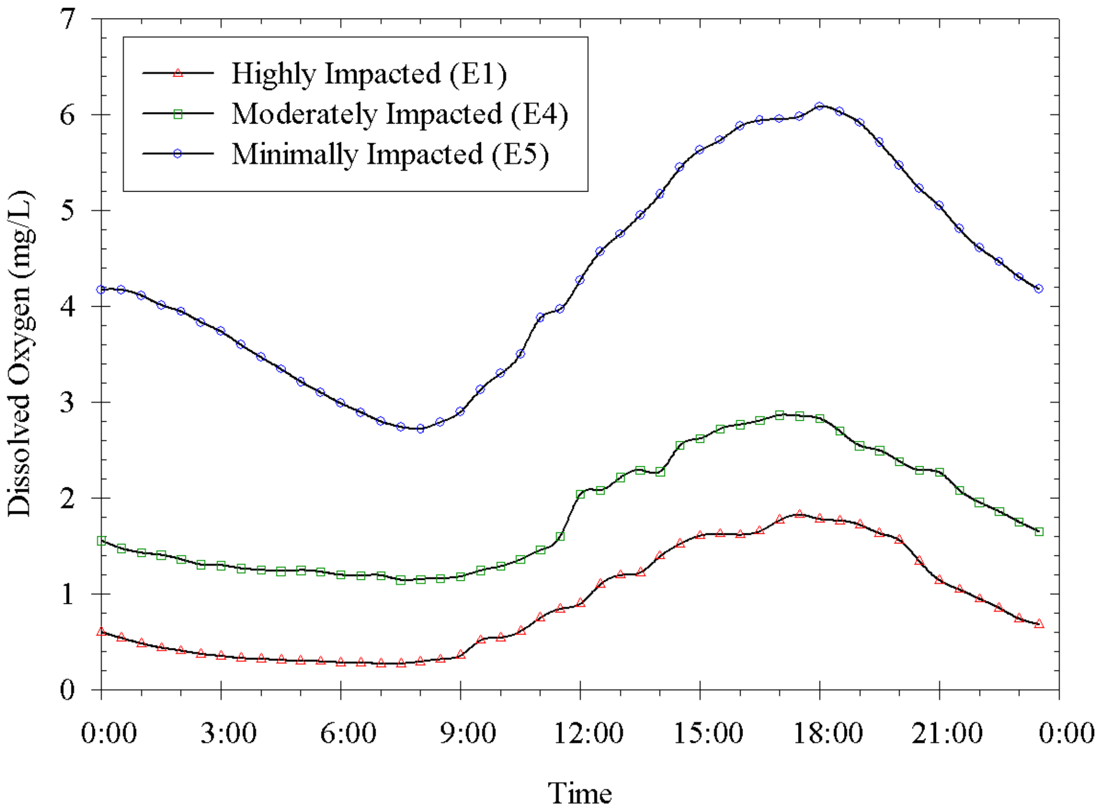

Figure 4-2-3. Average diel dissolved oxygen curves at three levels of nutrient impact. Impact in this case is defined as the degree of phosphorus enrichment above the background condition (approximately 10 µg P/L).

Dissolved oxygen concentrations below the current 5.0 mg/L Class III dissolved oxygen standard (Chapter 62-302, F.A.C.) occur commonly throughout the EPA, including interior marsh sites minimally impacted by nutrient enrichment or cattail invasion. Frequent dissolved oxygen levels below 5.0 mg/L are typical in macrophyte dominated wetlands where natural marsh processes of photosynthesis and respiration result in wide diel swings in dissolved oxygen levels. Since the low dissolved oxygen concentrations often measured in the Everglades represent the natural variability in this type of ecosystem, the

Department does not consider these excursions violations of the Class III dissolved oxygen standard. Therefore, the Class III standard of 5.0 mg/L is not believed to be applicable to the Everglades. This stance is supported by Paragraph 62-302.500(1)(f), F.A.C., which states that "*dissolved oxygen levels that are attributable to natural background conditions or man-induced conditions which cannot be controlled or abated may be established as alternative dissolved oxygen criteria for a water body or portion of a water body*." Additionally, any alternative criteria must "*not result in a lowering of dissolved oxygen levels in the water body, water body segment or any adjacent waters, and shall not violate the minimum criteria specified in Subsection 62-302.500(1), F.A.C.*" and must maintain the normal daily and seasonal fluctuations (Paragraph 62-302.500(1)(f), F.A.C.).

Pursuant to Section 62-302.800, F.A.C., the Department has undertaken the development of a Site Specific Alternative Criterion (SSAC) for dissolved oxygen in the Everglades that formally recognizes the natural background dissolved oxygen regime. The purpose of this report is to document the Department's efforts in establishing a SSAC for dissolved oxygen in the Everglades. Additionally, the report provides a procedure by which compliance with the SSAC can be evaluated.

Section 2.0 Procedures

Over the past three decades, dissolved oxygen concentrations have been measured in the surface water at various sites across the Everglades Protection Area (EPA) using a combination of grab and diel sampling methods. Grab sampling generally involves the collection of a single dissolved oxygen measurement using a Hydrolab or similar field probe at mid-depth at a specific geographic location and time. During 1994, the SFWMD initiated dissolved oxygen grab sampling at the current suite of 56 marsh stations in the three Water Conservation Areas and Everglades National Park (**Figure 4-2-4**). Results of the dissolved oxygen grab measurements collected by the SFWMD at the 56 station monitoring network were retrieved from the SFWMD DBHYDRO database for use during the development of the SSAC.

To provide a better characterization of the behavior of dissolved oxygen concentrations in the marsh during the day (*i.e.*, the timing and amplitude of the daily diel fluctuations), numerous research studies have also collected diel measurements. Diel sampling typically involves the periodic (every 15-60 minutes) measurement of the dissolved oxygen concentration at a specific site throughout a day or series of days using a suitable field probe suspended at mid-depth in the water column (McCormick and Laing, in review). Typically, other parameters such as water temperature, pH, and specific conductance are measured in conjunction with both grab and diel dissolved oxygen measurements. Diel measurements taken by the probe are automatically recorded by a data logger attached to the field instrument for later download to a PC.

The SFWMD has collected diel oxygen measurements at a series of 13 transect stations located within WCA-2A during two dry season (April 24 - 28, 1995 and February 24 – 27, 1998) and three wet season (August 28 - 31, 1995; September 1 – 6, 1996; and October 27 – 31, 1997) monitoring periods. (McCormick and Laing, in review; **Figure 4-2-5**). Monitoring periods were restricted to three to five days due to the increased potential for erroneous measurements due to biofouling of the probe when deployed for longer periods. Diel dissolved oxygen monitoring sites were located in the transition zones between stands of emergent macrophytes and open-water at all stations. Monitoring of a similar habitat at all stations allowed the data to be more comparable among stations.

Diel studies, in similar open water habitats, have also been conducted recently in WCA-1 and historically (early 1980's) in WCA-1, WCA-2A and WCA-3A. Diel data collected historically were not widely used due to the lack of adequate corresponding biologic data and period of record issues. Additionally, the use of recent diel dissolved oxygen data collected along a nutrient gradient in WCA-1 over three periods between June 1997 and February 1998 was limited since they encompass less than one year, and thus do not adequately reflect seasonal or annual variability. Diel data utilized during development of the SSAC was downloaded from the SFWMD ESRD database.

2.1 Data Handling

Once the grab and diel measurements were retrieved from the SFWMD databases, the data were screened based on data qualifiers and unreasonable values to assure that the highest quality data was utilized. No data were excluded due to qualifier codes, however 13 data points were excluded from analyses involving temperature (*e.g.*, percent saturation, saturation deficit) due to unreasonable temperature values. For example, samples collected in ENP on November 7, 1995 had temperatures reported between 75.8 and 81.6° C. Diel data were not qualified and no unreasonable values were observed and therefore were utilized as provided by the researchers.

The measurement frequency for the diel data collected along the WCA-2A transect, varied among the sampling periods. During the first three periods (April 1995, August 1995, September 1996), measurements were taken at a 15 minute frequency. On subsequent samplings measurements were recorded every 30 minutes. In order to provide a nearly consistent number of sample points per time interval, the dataset was reduced to only measurements taken on the hour and half hour (*e.g.*, 12:00, 12:30, 1:00, 1:30, 2:00, 2:30). Collection times for all data evaluated in this report were adjusted to Eastern Standard Time. That is, one hour was subtracted from the reported sample time for datasets collected during Daylight Savings Time. This adjustment standardizes the daily time period and avoids abrupt phase shifts in the diel curve that would occur at the beginning and end of Daylight Savings Time.

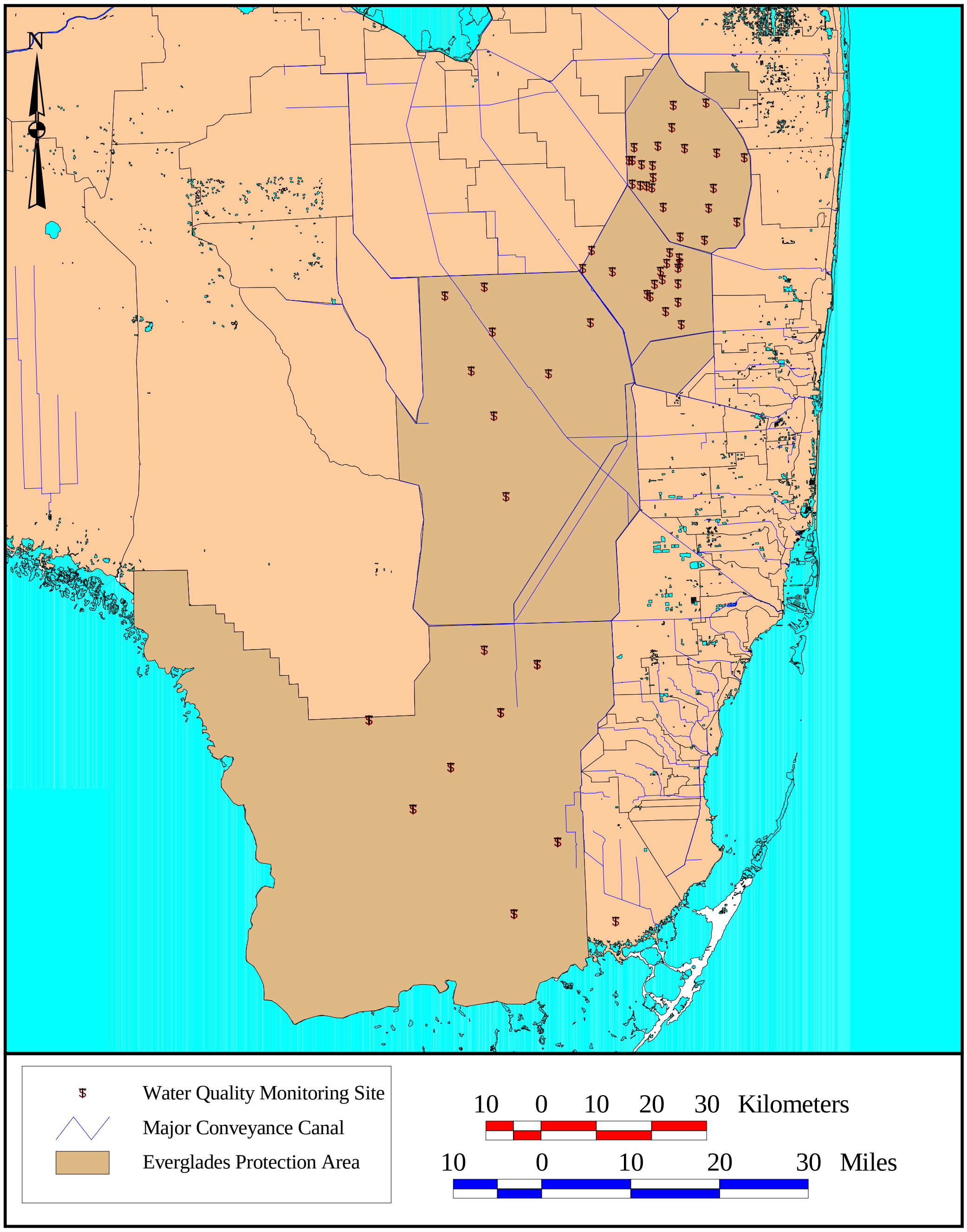

Figure 4-2-4. Location of interior marsh monitoring sites in the Everglades Protection Area.

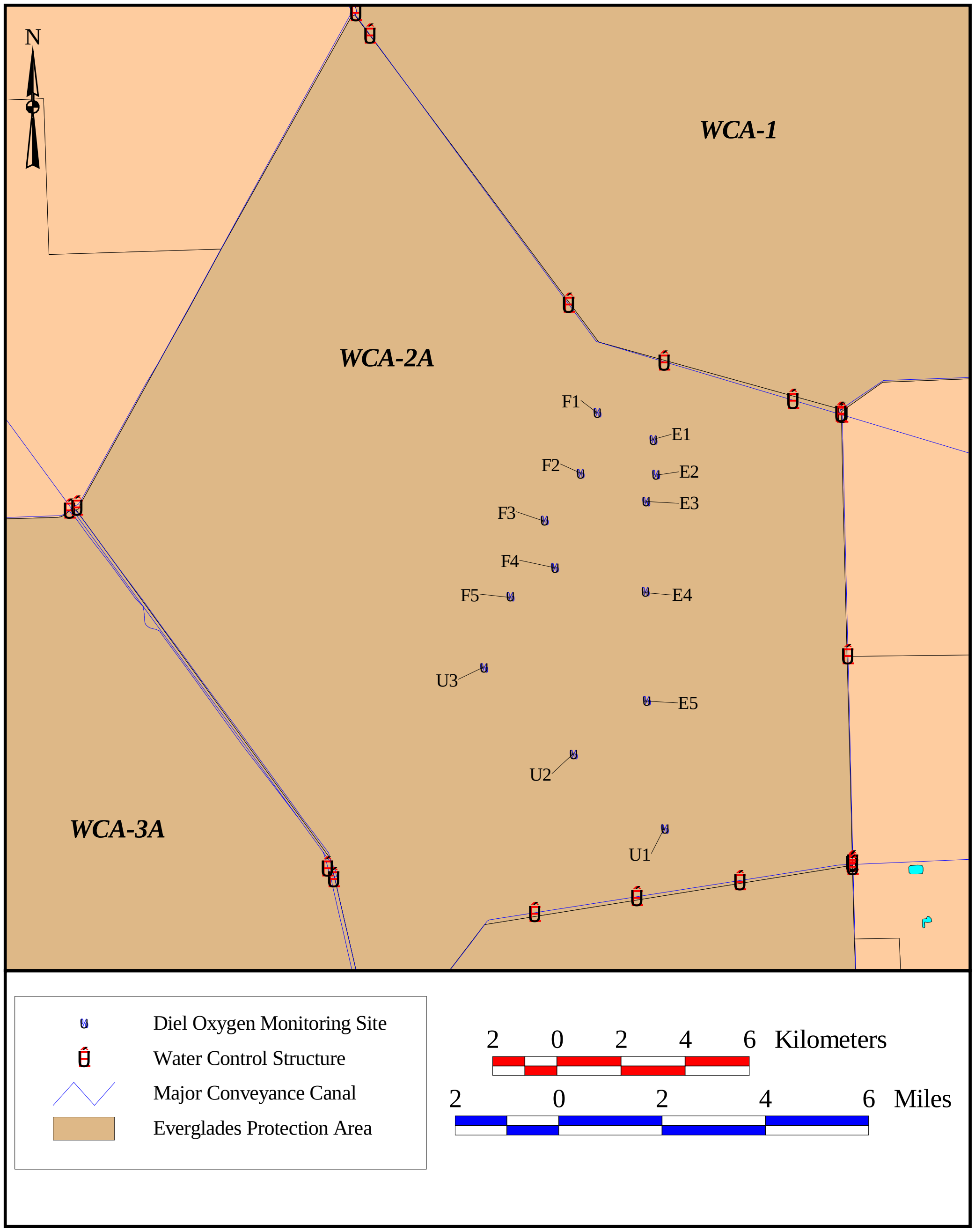

Figure 4-2-5. Location of diel dissolved oxygen monitoring sites in WCA-2A.

2.2 Selection of Reference Sites

Florida Administrative Code Subsection 62-302.800 (2) requires that during the establishment of a SSAC, an affirmative demonstration be made that the "*proposed alternative criteria would exist due to natural background conditions or man-induced conditions which cannot be controlled or abated*". Therefore, one of the initial steps in the development of a SSAC is to define the "natural background conditions" with respect to the parameter of concern. One method commonly used to define the natural background is to characterize the conditions at a group of reference sites that are representative of conditions within the ecoregion and are minimally impacted by point and nonpoint sources of pollution and other human activities that influence the parameter of concern (Hughes and Larsen, 1988; Hughes *et al*., 1994). In order to establish the natural background dissolved oxygen regime within the Everglades, both the diel and grab sample DO monitoring stations were classified as either "impacted" or "reference" sites based on an evaluation of the biogeochemical characteristics reported for each station.

Ideally, the definition of the background dissolved oxygen conditions in the Everglades should be based on a thorough understanding of the biology (*e.g.*, vegetative community, macroinvertebrates, fish), water chemistry, sediment chemistry, and hydrology of the region and how they affect and are affected by dissolved oxygen levels in the water column. The primary anthropogenic factors causing impairment of the natural dissolved oxygen regime in Everglades marshes are increased ground water inputs and phosphorus enrichment. Increased inputs of ground water, which is naturally low or devoid of dissolved oxygen, act to lower dissolved oxygen levels by diluting the more oxygen rich surface waters. Intrusion of ground water into surface water tends to be a larger problem near canals dug below the ground water level than within interior marsh areas. The influence of ground water can also be observed near water control structures or canals which discharge into the marsh. Therefore, due to the potential for adverse influence from ground water, any station located within one km of a canal or discharge structure was classified as “impacted”.

As previously discussed, increased phosphorus concentrations have been associated with depressed dissolved oxygen levels in the Everglades marshes. Phosphorus enrichment works to lower dissolved oxygen concentrations indirectly through increases in the rate of community metabolism (*i.e.*, increased production and subsequent oxygen demand), reductions in the periphyton and submerged aquatic vegetation community, and increases in dense emergent macrophyte stands (Swift, 1981; Flora *et al.*, 1986; McCormick *et al.*, 1997; McCormick and Laing, in Review). Comparison of marsh grab sample data to the geometric mean total phosphorus concentrations for the preceding 12-month period, indicates an inverse relationship between dissolved oxygen and total phosphorus (TP) at phosphorus concentrations ranging from background concentrations (2-10 µg/L) to 30-40 µg/L (**Figure 4-2-6).** Dissolved oxygen concentrations collected at background phosphorus levels were greater than all higher phosphorus ranges including the 10-15 µg/L range (t-test, $p<0.0001$). Total phosphorus concentrations of 10 µg/L or less are widely believed to represent natural background levels in the Everglades. Additionally, a TP concentration of 10 µg/L is specified as the default phosphorus criterion by the EFA. Therefore, for the purpose of defining background conditions for this SSAC, a TP concentration of 10 µg/L was used to differentiate impacted from reference or minimally impacted sites. A site was classified as reference if the average annual geometric mean TP concentration for the years 1994 to 1999 was less than or equal to 10 µg/L. Using a one-tailed t-test (α=0.05), 42 out of the 56 sites were initially classified as reference.

Sediment phosphorus acts to integrate nutrient conditions over time and has been utilized by several researchers to demonstrate areas where phosphorus enrichment have occurred (Craft and Richardson, 1993a and 1993b; DeBusk, *et. al*., 1994, Reddy, *et. al*., 1991; McCormick *et al.*, 2000). Therefore, sediment phosphorus concentrations were also used to help classify sites with respect to phosphorus enrichment. Total sediment phosphorus concentrations between 500 and 600 mg/kg have frequently been used by researchers to indicate were significant enrichment has occurred (McCormick *et al.,* 2000). The 500-600 mg/kg zone can be used as a surrogate measure for other parameters, including macrophytes and SOD, which influence dissolved oxygen. In WCA-2A, sediment TP concentrations exceeding the 500 to 600 mg/kg range are particularly indicative of macrophyte changes including increases in the aerial extent of emergent vegetation (FDEP, 1999; McCormick *et al.,* 2000**). Figures 4-2-7** through **4-2-9** provide sediment TP contour maps of WCA1, WCA-2, and WCA-3 generated using the best available data. A more detailed description of how the maps were generated can be found in the Department's report concerning P criterion development (FDEP, 1999). Dissolved oxygen monitoring sites located outside of the 600 mg/kg sediment P contour were classified as impacted.

In addition to being chemically unimpacted, reference sites should also possess a balanced community of aquatic flora and fauna. Here, the balanced community of flora may be defined as one which provides the natural background photosynthetic regime; that is a mosaic of sawgrass, slough (containing periphyton and submergent aquatic vegetation), tree island and other natural vegetative communities. Faunal data were also reviewed to provide an assurance that reference site dissolved oxygen concentrations are sufficient to prevent impairment in these biological communities.

Where available, flora and fauna monitoring data were utilized to confirm (support) reference site designations. All diel data used during this analysis were collected along the nutrient gradient in WCA-2A, which is the most intensively studied region of the Everglades. Therefore substantial biological data exists for the diel and grab sites along this gradient. However, biological data for the other grab sampling sites is generally unavailable or highly limited.

Review of the biological data and extensive data evaluations conducted by the Department (FDEP, 1999) and SFWMD (McCormick *et al*., 2000) along the WCA-2A transects indicate that there is a substantial change in community composition and function between enriched and unenriched areas. Analyses indicate that P enrichment has adversely influenced the biological (periphyton, macroinvertebrate, and macrophyte) communities at sites located less than 7 to 8 km from the S-10 discharge structures (FDEP, 1999; McCormick *et al.*, 2000). Some of the most notable changes in enriched areas include loss of sensitive and increased tolerant periphyton species, increased community productivity, loss of SAV and increased emergent macrophyte (especially cattail) stands, and taxonomic shifts in macroinvertebrate communities. Based on the biological data evaluations, diel dissolve oxygen monitoring sites E5, F5, U1, U2, and U3 in WCA-2 are functioning as natural background sites and can be classified as reference or minimally impacted sites.

In addition to the review of biological data, site reconnaissance trips to the Water Conservation Areas and the ENP were conducted for diel and grab sampling sites on February 28-March 2, 2000, May 3-4, 2000, and May 23, 2000. All DO monitoring sites classified as reference were observed to conform to background vegetation patterns. Vegetation at sites designated impacted varied, but all diverged from that observed at reference sites and showed a loss of natural periphyton and SAV communities found at minimally impacted sites. Site visits corroborated site designations based on chemical and biological data.

The geometric mean phosphorus concentrations for the diel and grab sampling sites are provided in **Table 4-2-1** along with the site classifications based on sediment P levels, proximity to canals or discharge structures, and the review of biological information. In the final site classification, only sites judged to be unimpacted in all respects were retained as reference sites with sites where contradictory information was present being classified as unknowns. Based on water column P concentration, sediment P, and potential ground water or canal influences, 14 sites were classified as impacted and 39 were classified as reference sites. Due to high sediment P concentrations or potential human induced influences sites LOX10, CA27 and C35 were classified as unknowns. Site CA27 is within several 100 m of the L-6 canal on the northwest edge of WCA-2A and was thus classified as an unknown due to an increased possibility of being influenced directly or indirectly by the canal. Review of sediment contours tended to be in agreement with site classifications based on water-column P, with two exceptions (**Figures 4-2-7 to 4-2-9**). Sites LOX10 (WCA-1) and CA35 (WCA-2), which had geometric mean water column concentrations less than or equal to 10 µg/L, lie outside the 600 mg/kg contours. Due to uncertainty in potential eutrophication impacts LOX10 and CA35 could not be considered as reference and were therefore classified as unknowns.

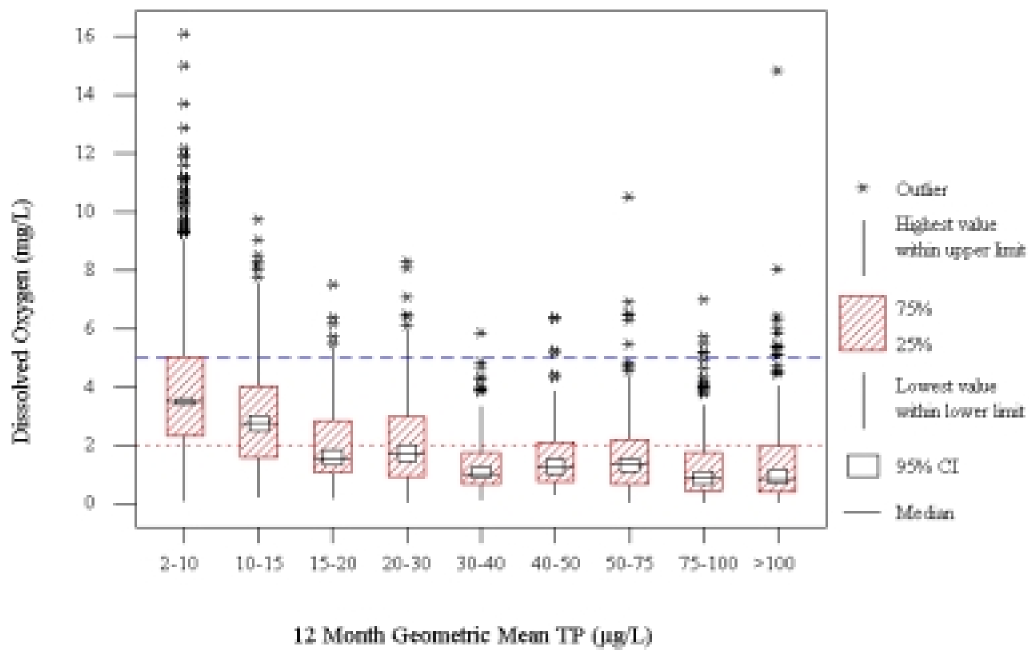

Figure 4-2-6. Boxplot of dissolved oxygen concentrations at 56 monitoring stations in the Everglades Protection Area versus 12 month prior geometric mean total phosphorus concentration. Dissolved oxygen data were collected as grabs. Total phosphorus ranges were selected as increments above background (2-10 µg/L), with the ranges 10-15, 15-20 and 20-30 being representative of various phosphorus threshold recommendations. Phosphorus values reported as less than the MDL were replaced with ½ the MDL. The dashed lines are reference lines showing 2.0 mg O_2/L and the state standard of 5.0 mg O_2/L.

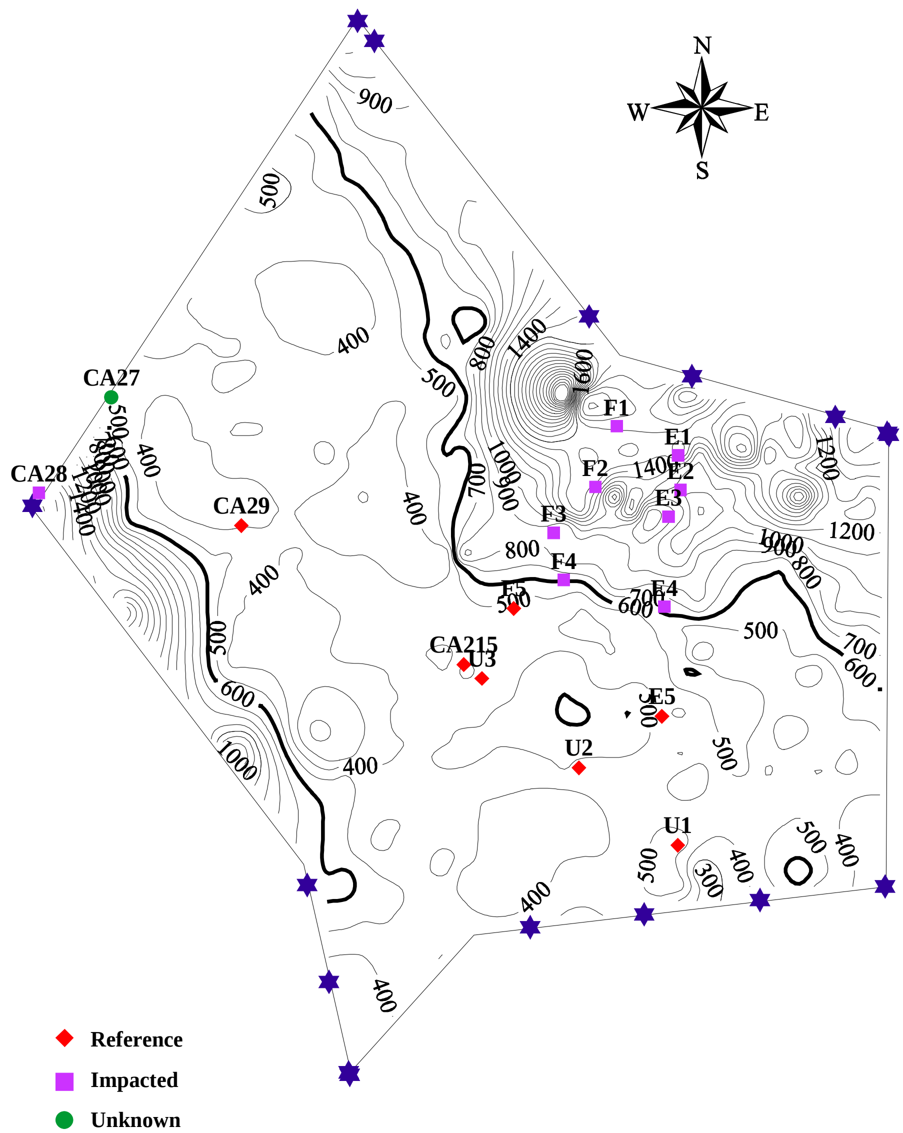

Figure 4-2-7. Map of WCA-2A with sediment total phosphorus contours determined from a combination of 1991 Reddy and 1995-96 EMAP, 1997 Duke studies, SFWMD transect, Duke transect and 404 Permit monitoring data (0-10 cm depth).

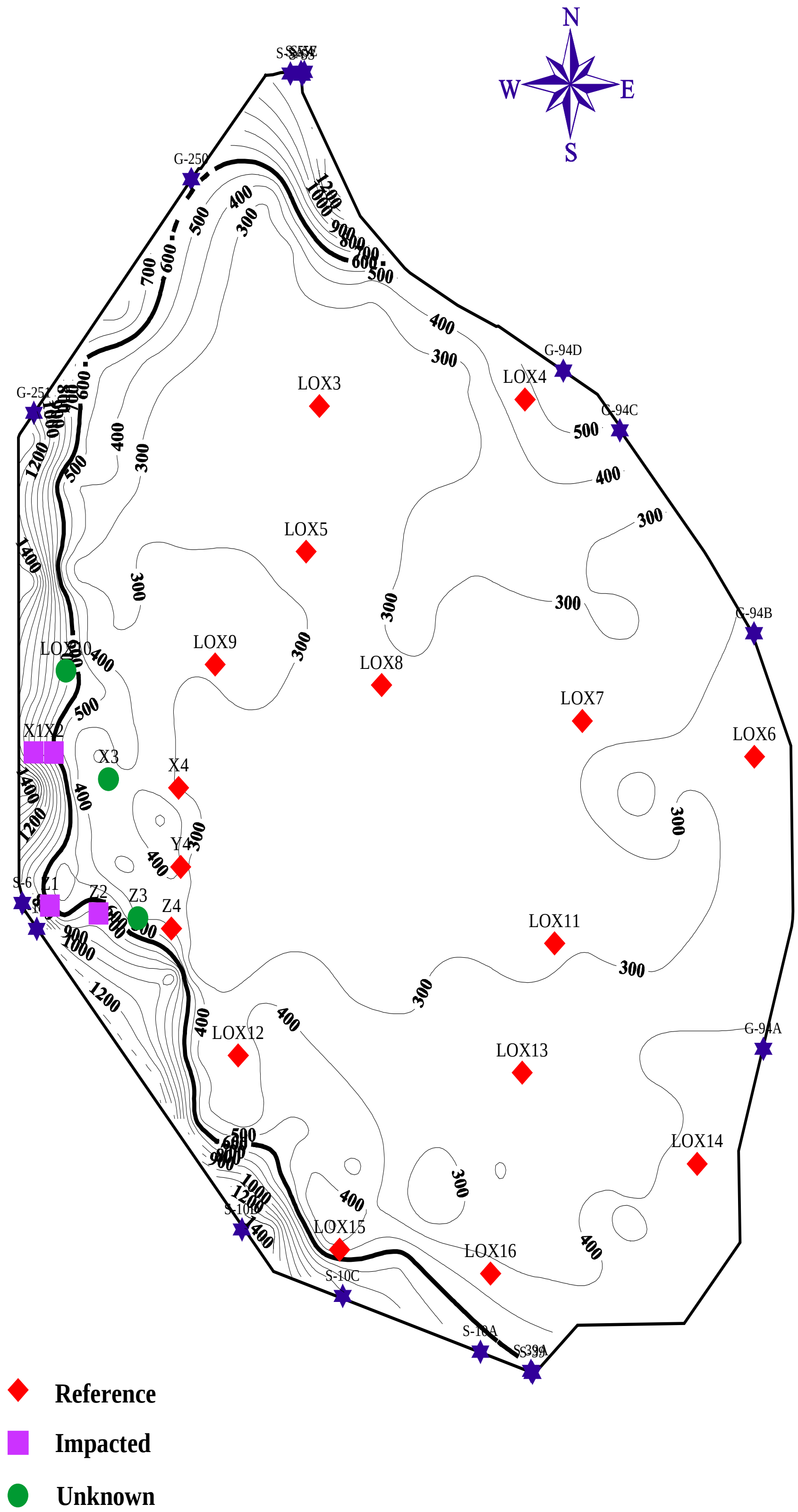

Figure 4-II-8. Map of WCA-1 with sediment total phosphorus contours determined from a combination of 1991 Reddy and 1995-96 EMAP data (0-10 cm depth).

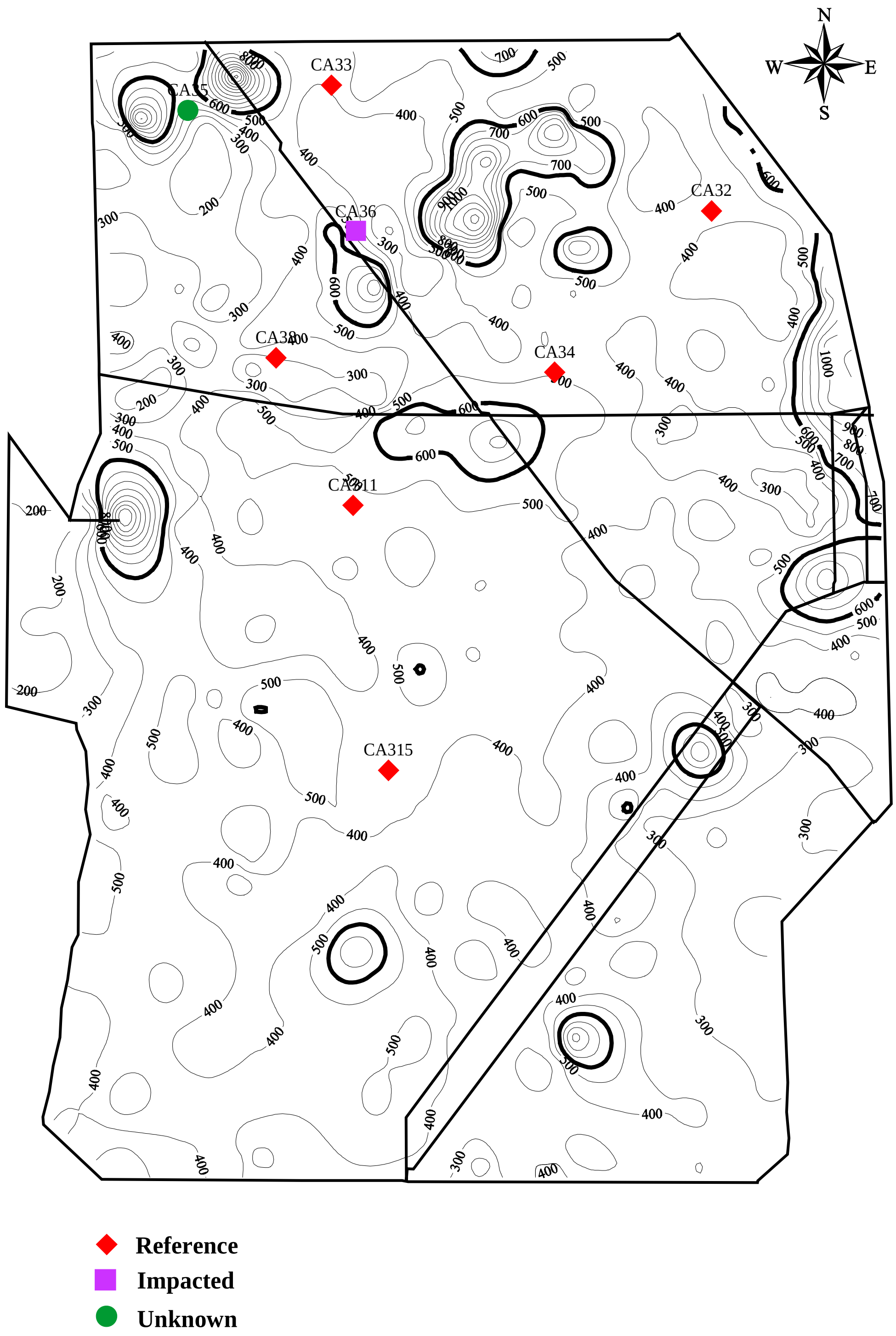

Figure 4-2-9. Map of WCA-3 with sediment total phosphorus contours determined from a combination of 1991 Reddy and 1995-96 EMAP data (0-10 cm depth).

Table 4-2-1. Results of site classifications based on water-column TP concentrations, sediment TP levels, proximity to canal influence, review of biological information, and site visits. Phosphorus concentrations were screened based on qualifier codes and measurements reported as less than the MDL were replaced with a value of ½ the reported MDL.

Site	Final Site Classification	Mean Annual Geometric Mean TP ($\leq$10 µg/L)	Outside 600 mg/kg Sediment P Contour	Proximity to Canal (<1 km)	Biological Impacted (Data Review/Site Visits)
CA28	Impacted	84.4	Yes	Yes	Yes
CA36	Impacted	26.7	Yes	Yes	Yes
E1	Impacted	55.2	Yes	No	Yes
E2	Impacted	43.4	Yes	No	Yes
E3	Impacted	30.7	Yes	No	Yes
E4	Impacted	15.2	Yes	No	Yes
F1	Impacted	111.9	Yes	No	Yes
F2	Impacted	61.3	Yes	No	Yes
F3	Impacted	24.9	Yes	No	Yes
F4	Impacted	16.6	Yes	No	Yes
X1	Impacted	28.3	Yes	Yes	Yes
X2	Impacted	12.7	Yes	No	Yes
Z1	Impacted	33.3	Yes	Yes	Yes
Z2	Impacted	13.6	Yes	No	Yes
CA215	Reference	5.9	No	No	No
CA27	Unknown	9.4	No	Yes	No
CA29	Reference	6.8	No	No	No
CA311	Reference	4.8	No	No	No
CA315	Reference	5.1	No	No	No
CA32	Reference	7.8	No	No	No
CA33	Reference	11.4	No	No	No
CA34	Reference	8.6	No	No	No
CA35	Unknown	10.1	Yes	No	No
CA38	Reference	6.4	No	No	No
E5	Reference	7.8	No	No	No
F5	Reference	9.7	No	No	No
LOX10	Unknown	9.1	Yes	No	No
LOX11	Reference	8.4	No	No	No
LOX12	Reference	6.9	No	No	No
LOX13	Reference	7.2	No	No	No
LOX14	Reference	7.1	No	No	No
LOX15	Reference	6.6	No	No	No
LOX16	Reference	8.0	No	No	No
LOX3	Reference	9.2	No	No	No
LOX4	Reference	10.0	No	No	No
LOX5	Reference	8.7	No	No	No
LOX6	Reference	7.2	No	No	No
LOX7	Reference	7.6	No	No	No
LOX8	Reference	7.5	No	No	No
LOX9	Reference	8.0	No	No	No
NE1	Reference	6.6	No	No	No
NP201	Reference	4.4	No	No	No
P33	Reference	4.7	No	No	No
P34	Reference	3.8	No	No	No
P35	Reference	7.5	No	No	No
P36	Reference	6.0	No	No	No
P37	Reference	3.1	No	No	No
TSB	Reference	4.1	No	No	No
U1	Reference	7.8	No	No	No
U2	Reference	8.0	No	No	No
U3	Reference	7.8	No	No	No
X3	Reference	7.2	No	No	No
X4	Reference	8.6	No	No	No
Y4	Reference	8.5	No	No	No
Z3	Reference	8.5	No	No	No
Z4	Reference	7.7	No	No	No

2.3 Approach to Defining Natural Background Conditions

Because the reference sites were selected based on parameters that influence or are influenced by dissolved oxygen levels, the data collected at these sites can be used to define the natural background dissolved oxygen conditions within the marsh. Since the SSAC results from the definition of the natural background conditions, it is important to utilize an approach that adequately captures the natural characteristics of dissolved oxygen within the reference areas. One possible approach is to utilize the average conditions based on the distribution of observations at the reference sites to develop a single point SSAC. For example, the average dissolved oxygen concentration at the references sites in the three Water Conservation Areas and Everglades National park is 3.81 ± 0.08 mg/L (mean ± 95% CI). Setting the criterion at the lower 95% confidence interval would yield a single point standard of 3.74 mg/L. However, the single point approach misses one of the most important characteristics of the natural background dissolved oxygen condition, the wide diel fluctuations. Because of the normal diel cycle, a single point criterion is under-protective for part of the day while being over-protective for another (**Figure 4-2-10**), that is, compliance determination would not be independent of sample collection time. Samples collected early in the morning would have a greater chance of being erroneously classified as out of compliance, even at background sites (**Figure 4-2-11**). Conversely, the probability of falsely classifying an observation as in compliance is elevated late in the afternoon when dissolved oxygen levels are at a maximum (**Figure 4-2-11**). Therefore, in order to more accurately characterize the natural diel cycle, a model, which varies the target concentration by time of day, was developed to define the natural background dissolved oxygen regime.

Using the diel data collected at the set of five reference stations (*i.e.*, E5, F5, U1, U2, and U3) located along the SFWMD nutrient gradient in WCA-2A, a predictive model was developed and refined that defines the average dissolved oxygen regime at the reference sites. The WCA-2 sites were selected for model development because these sites provide the largest abundance of diel dissolved oxygen data with supporting biological information. Additionally, the diel data provide the best characterization of the dissolved oxygen regime. The model developed followed the typical sinusoidal diel cycle evident in 4-II-2. Grab data from areas outside WCA-2 were used to verify that the model was suitable for application to other areas of the Everglades.

Since periodic (*e.g.*, monthly, biweekly) grab sampling is the current and most practical means of dissolved oxygen compliance determination, it is intended that compliance with this SSAC be measured through comparison of grab sample dissolved oxygen measurements with model predicted values. Therefore, the model developed from the diel data was cross-validated with a set of grab reference sites selected to be representative of the three Water Conservation Areas and Everglades National Park. The curve predicted by the model was then adjusted to account for the natural spatial and temporal (seasonal, annual) variability seen in grab samples. This adjusted model is then used to define the lower prediction interval that is proposed to serve as the SSAC.

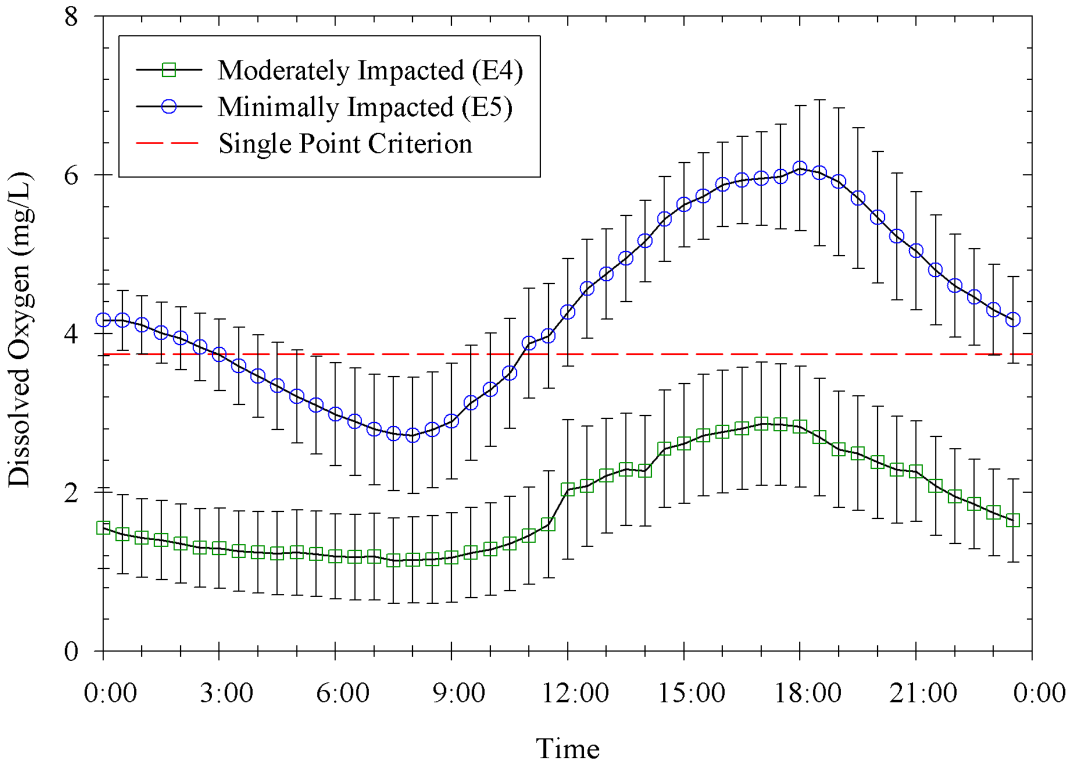

Figure 4-2-10. Mean ± 95% C.I. dissolved oxygen curves at moderately and minimally impacted sites. The red dashed line shows a potential "point" criterion based on the lower 95% C.I. at a set of reference stations.

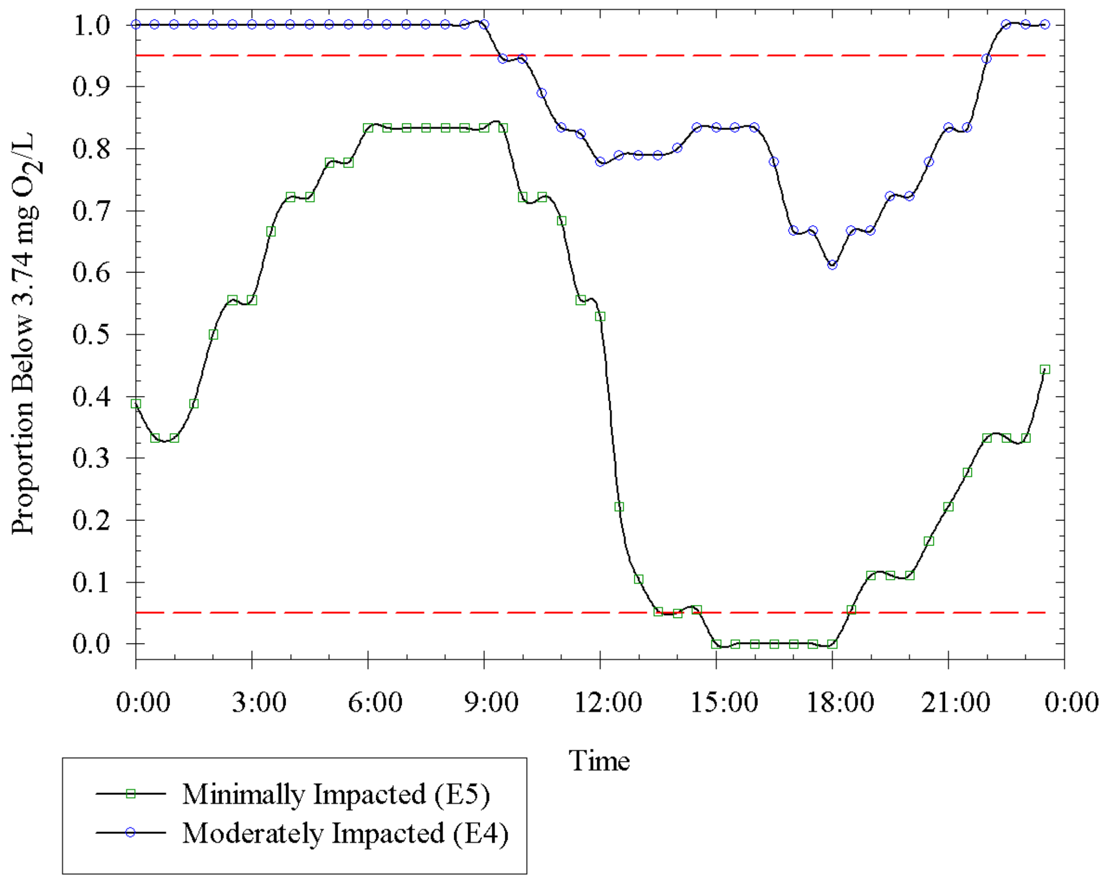

Figure 4-2-11. Proportion of diel dissolved oxygen samples, collected at moderately and minimally impacted sites, less than 3.74 mg/L. The red dashed and black solid lines are reference lines indicating 5 and 95% below the "point" standard.

Section 3.0 Preliminary Data Evaluation

Section 62-302.530 (31) of the Florida Administrative Code (FAC) states that in Class III predominantly fresh waters, dissolved oxygen concentrations "*shall not be less than 5 mg O_2/L*" and "*normal daily and seasonal fluctuations above these levels shall be maintained*". A preliminary review of data collected from 56 interior marsh stations in the Everglades Protection Area (EPA) reveals that dissolved oxygen concentrations routinely fall below the current standard even at minimally impacted background sites. The overall average dissolved oxygen concentration for the grab samples collected at the 56 monitoring sites between 1994 and 1999 was well below the 5.0 mg/L state standard (3.19 ± 2.21 mg/L, mean ± standard deviation) with 81% of the measurements dropping below 5.0 mg/L. The mean dissolved oxygen concentrations at individual monitoring stations ranged from 1.07 to 7.43 mg/L with between 15.8% and 98.1% of the values not meeting the current standard (**Table 4-2-2**).

Spatially, the average dissolved oxygen concentration in Everglades National Park is higher than those observed within the Water Conservation Areas (**Figure 4-2-12**). The higher oxygen concentrations have resulted in a lower excursion rate for the Park (53.7%) than for the Water Conservation Areas (79.4 - 87.0%). This spatial pattern is partially due to differences in the relative proportions of data from impacted sites among the areas. Water Conservation Area 2A, which had the lowest median dissolved concentration (2.09 mg/L) has the largest number of impacted sites (**Table 4-2-2**), followed by WCA-1 with a median dissolved oxygen concentration of 3.02 mg/L. In contrast, none of the sites within the Park were classified as being impacted in regards to dissolved oxygen. Additionally, many of the ENP sampling sites including NE1, P33, P36 and P35 are located within Shark River Slough; a region characterized by large expanses of open water slough, which provides a large oxygen source. The remaining ENP sites are either in Taylor Slough or on the western periphery of Shark River Slough.

Along the SFWMD transects in WCA-2A, an increasing trend in oxygen concentrations with distance from canal inflows is evident. The trend is apparent whether diel or grab data are evaluated and clearly shows the difference between impacted and background areas (**Figures 4-2-13** and **4-2-14**). Sites within seven km of the canal (*i.e.*, E1-E4 and F1-F4) exhibit frequently low oxygen concentrations with few measurements above 2.0 mg/L. In contrast, sites located greater than seven km from the canal (*i.e.*, E5, F5 and U1-U3) exhibit wider oxygen ranges with values rarely below 2.0 mg/L. Diel dissolved oxygen measurements collected by the SFWMD at stations along the nutrient gradient transects in WCA-2A were averaged by station and plotted by time of day in **Figure 4-2-15**. This series of diel curves illustrates the typical difference in daily fluctuations between the nutrient impacted and background regions of WCA-2A. Mean concentrations at enriched sites were consistently below the unenriched areas throughout the diel cycle. Sites F1, F2, E1, E2, and E3, which had the highest level of phosphorus enrichment (see **Table 4-2-2**), exhibited prolonged periods of extremely low dissolved oxygen (<1.0 mg/L). While sites (F3, E4 and F4) with moderate enrichment (see **Table 4-2-2**) tended to stay above 1.0 mg/L but below 2.0 mg/L for much of the day. Diel curves at the minimally impacted (reference) sites (E5, F5, and U1–U3) show pronounced daily fluctuations with concentrations tending to remain above 2.0 mg/L throughout the entire cycle and average daily maxima ranging from 4.6 to 7.7 mg/L. Despite being representative of the natural background condition, all five diel sites classified as reference (minimally impacted) sites commonly violate the current Class III standard over at least a portion of the diel cycle. These violations illustrate the inappropriateness of the current standard to the Everglades and the need to establish a Site Specific Alternative Criterion that recognizes the low dissolved oxygen regime and wide diurnal fluctuations that occur naturally within minimally impacted areas of the Everglades.

Table 4-2-2. Summary of dissolved oxygen concentrations (mg/L) at interior Everglades marsh stations from 1994 to 1999. Samples were collected as grabs.

Area	Site	Mean	Standard Deviation	Minimum	First Quartile	Median	Third Quartile	Maximum	N
WCA1	LOX3	5.54	2.12	0.45	3.86	5.97	7.38	9.27	38
	LOX4	3.71	1.81	1.17	2.37	3.54	4.77	8.27	40
	LOX5	5.52	1.70	1.82	4.23	5.56	6.82	9.69	45
	LOX6	3.00	1.72	0.19	1.65	2.82	4.41	8.50	59
	LOX7	3.11	1.79	0.14	1.76	2.69	4.45	7.92	58
	LOX8	3.93	2.17	0.06	2.18	3.75	5.63	9.71	60
	LOX9	4.64	1.73	0.73	3.54	4.55	5.67	8.53	48
	LOX10	4.54	1.74	1.91	3.17	4.28	5.25	8.89	44
	LOX11	3.19	2.00	0.11	1.91	3.11	4.87	7.43	54
	LOX12	3.48	1.89	0.49	2.01	3.17	4.64	8.30	65
	LOX13	3.74	2.05	0.27	2.16	3.62	5.87	7.88	57
	LOX14	2.36	1.43	0.42	1.37	2.13	2.87	7.50	61
	LOX15	3.38	1.60	0.24	2.40	3.19	4.31	7.11	64
	LOX16	2.23	1.52	0.29	1.06	2.03	3.06	8.98	60
	X1	2.01	1.38	0.18	0.77	1.80	3.04	5.19	49
	X2	2.30	1.29	0.33	1.24	2.18	3.24	6.78	53
	X3	3.34	1.63	0.06	2.29	2.93	4.41	7.58	52
	X4	3.49	2.01	0.56	2.20	2.98	4.42	7.94	51
	Y4	3.67	1.87	0.32	2.41	3.09	5.24	8.34	51
	Z1	1.07	1.09	0.07	0.32	0.67	1.55	5.72	52
	Z2	2.00	1.37	0.31	0.93	1.72	2.95	5.94	49
	Z3	3.48	1.75	1.11	2.13	3.16	4.50	8.82	52
	Z4	4.01	1.97	0.51	2.65	3.91	5.43	8.66	52
WCA2A	CA215	4.89	2.22	0.09	3.35	4.55	6.20	12.17	100
	CA27	3.85	1.97	0.41	2.52	3.59	4.82	10.45	106
	CA28	1.37	1.43	0.04	0.49	0.99	1.68	6.99	93
	CA29	3.70	1.98	0.18	2.36	3.45	4.86	9.09	106
	E1	1.98	2.05	0.24	0.67	1.03	2.32	10.51	73
	E2	1.68	1.31	0.11	0.75	1.36	2.11	5.88	66
	E3	1.66	1.38	0.18	0.69	1.32	2.10	7.31	74
	E4	1.82	1.47	0.25	0.87	1.42	2.19	7.51	78
	E5	3.45	1.39	0.53	2.48	3.51	4.34	7.33	81
	F1	1.82	1.95	0.03	0.45	1.09	2.75	14.85	184
	F2	1.58	1.33	0.03	0.60	1.18	2.09	6.90	215
	F3	1.99	1.27	0.30	0.94	1.66	2.59	6.43	91
	F4	1.78	1.24	0.06	1.00	1.38	2.48	6.42	192
	F5	4.12	1.89	0.86	2.53	3.88	5.36	9.74	89
	U1	3.06	1.62	0.52	1.89	2.77	3.92	9.77	91
	U2	3.20	1.58	0.98	2.00	3.07	4.13	10.41	89
	U3	4.32	1.82	0.32	3.01	4.33	5.38	9.61	102
WCA3A	CA311	3.43	1.61	0.10	2.24	3.34	4.37	8.92	127
	CA315	2.86	1.73	0.10	1.47	2.73	3.74	8.59	133
	CA32	4.24	1.64	0.88	3.04	3.88	5.37	8.38	107
	CA33	3.02	1.37	0.44	2.18	2.84	3.54	8.05	102
	CA34	2.76	1.34	0.64	1.78	2.46	3.52	7.60	112
	CA35	3.77	1.40	1.10	2.86	3.75	4.17	8.32	79
	CA36	2.35	1.70	0.04	0.97	1.90	3.41	8.10	115
	CA38	3.14	1.42	0.30	2.06	3.03	3.96	7.45	112
ENP	NE1	3.96	2.04	0.80	2.20	3.50	5.58	10.60	68
	NP201	4.92	2.22	0.70	3.20	4.64	6.20	13.70	66
	P33	4.00	1.84	1.30	2.85	3.66	4.93	10.50	70
	P34	5.57	1.77	0.10	4.70	5.35	6.61	10.70	54
	P35	4.02	1.74	0.60	2.98	3.70	4.91	10.70	58
	P36	4.71	2.23	1.40	3.05	4.30	5.82	15.00	70
	P37	7.43	2.55	1.30	5.75	7.50	8.55	16.10	57
	TSB	6.37	2.88	1.60	3.73	6.30	8.78	11.90	60

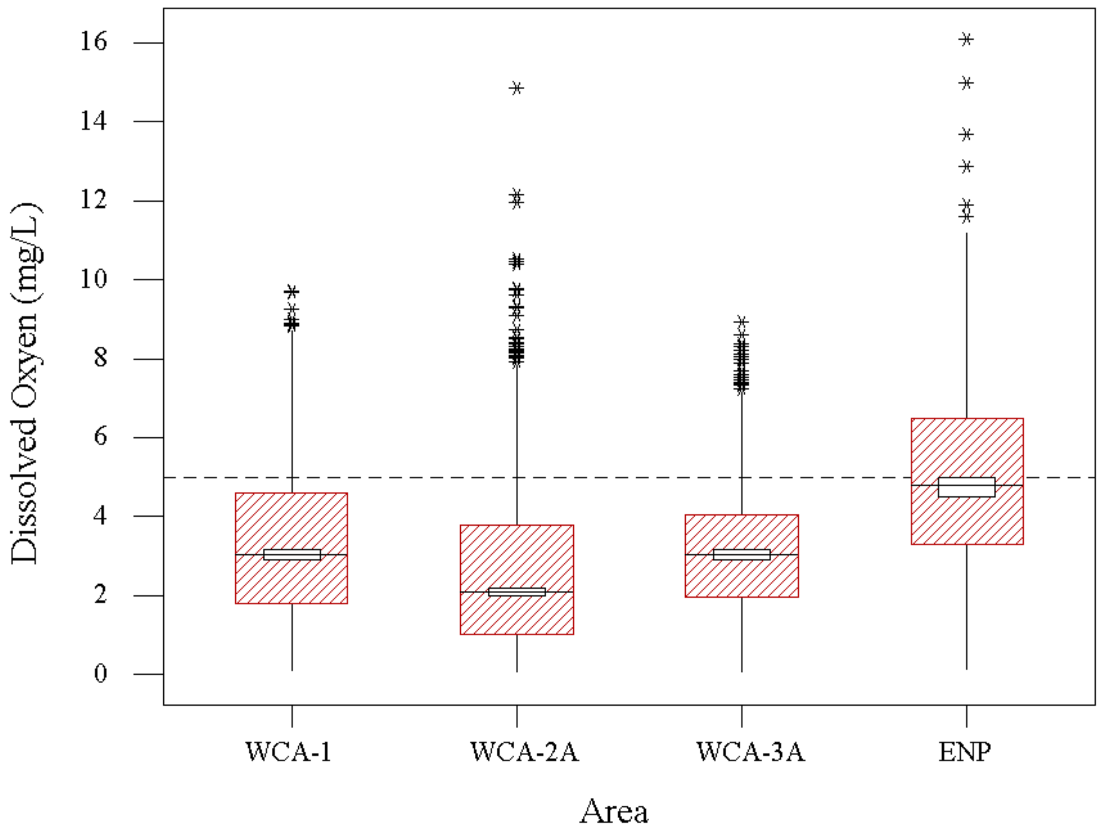

Figure 4-2-12. Boxplot of dissolved oxygen concentrations at monitoring sites in WCA-1, WCA-2A, WCA-3A and Everglades National Park. Data were collected between 1994 and 1999. See Figure 4-II-6 for a description of box and whisker plots.

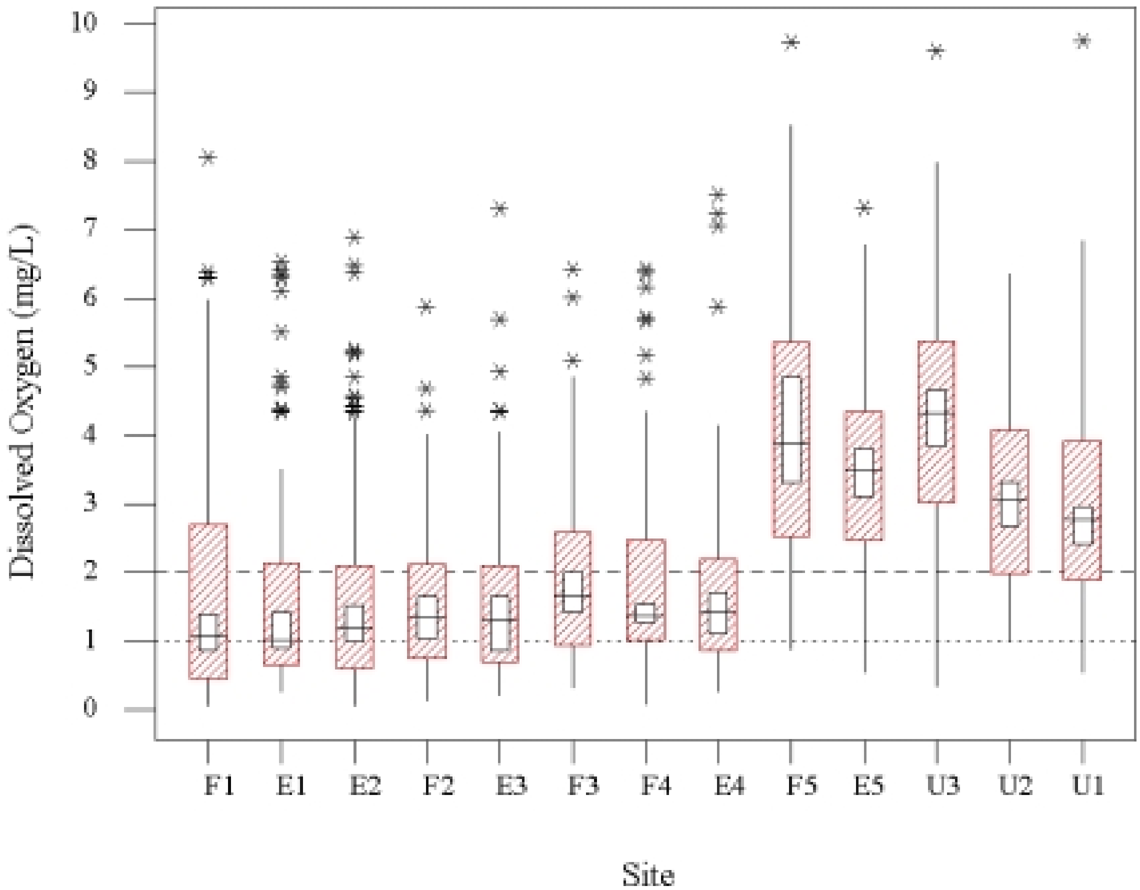

Figure 4-2-13. Diel dissolved oxygen concentrations along the SFWMD WCA-2A nutrient gradient, collected between April 25, 1995 and February 27, 1998. Sites are ordered by increasing distance from the Hillsboro canal. Dashed lines are placed at 1.0 and 2.0 mg/L for reference purposes. See Figure 4-II-6 for a description of box and whicker plots.

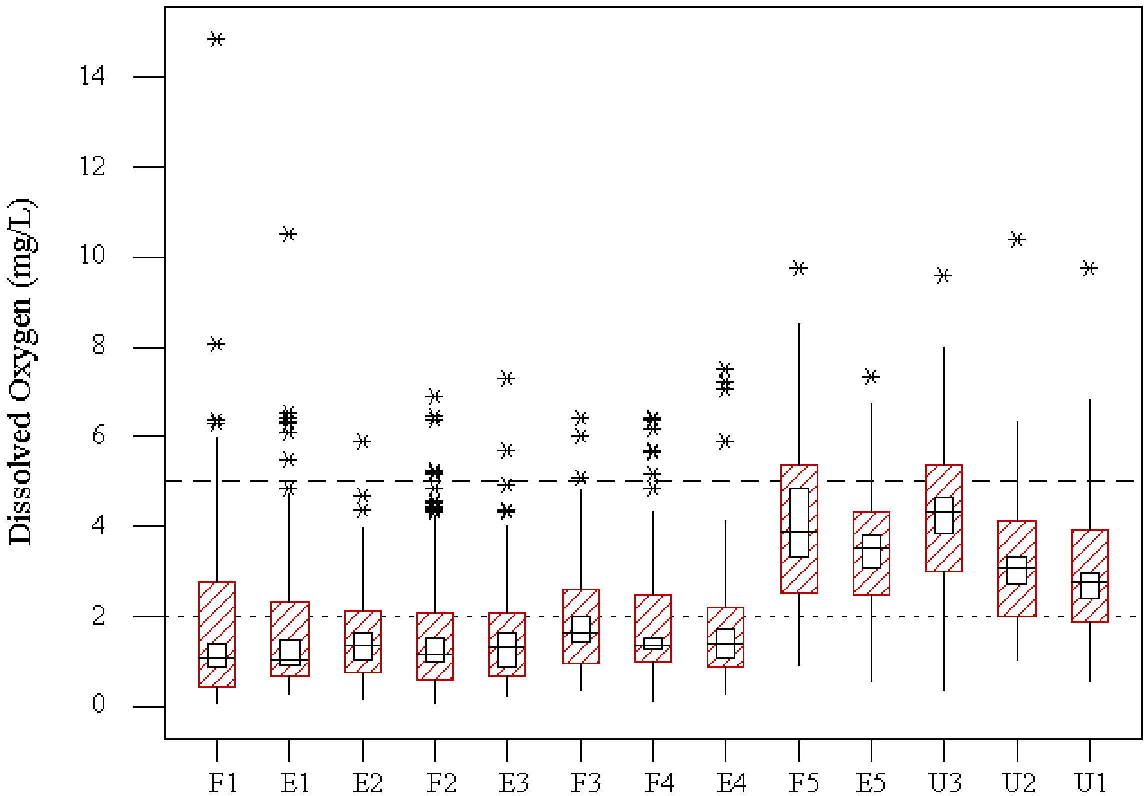

Figure 4-2-14. Summary of dissolved oxygen concentrations measured along the SFWMD WCA-2A nutrient gradient as approximately bi-weekly grab samples between February 14, 1994 and December 28, 1999. Sites are ordered by increasing distance from the Hillsboro canal. Dashed lines are placed at 1.0 and 2.0 mg/L for reference purposes. See Figure 4-II-6 for a description of box and whisker plots.

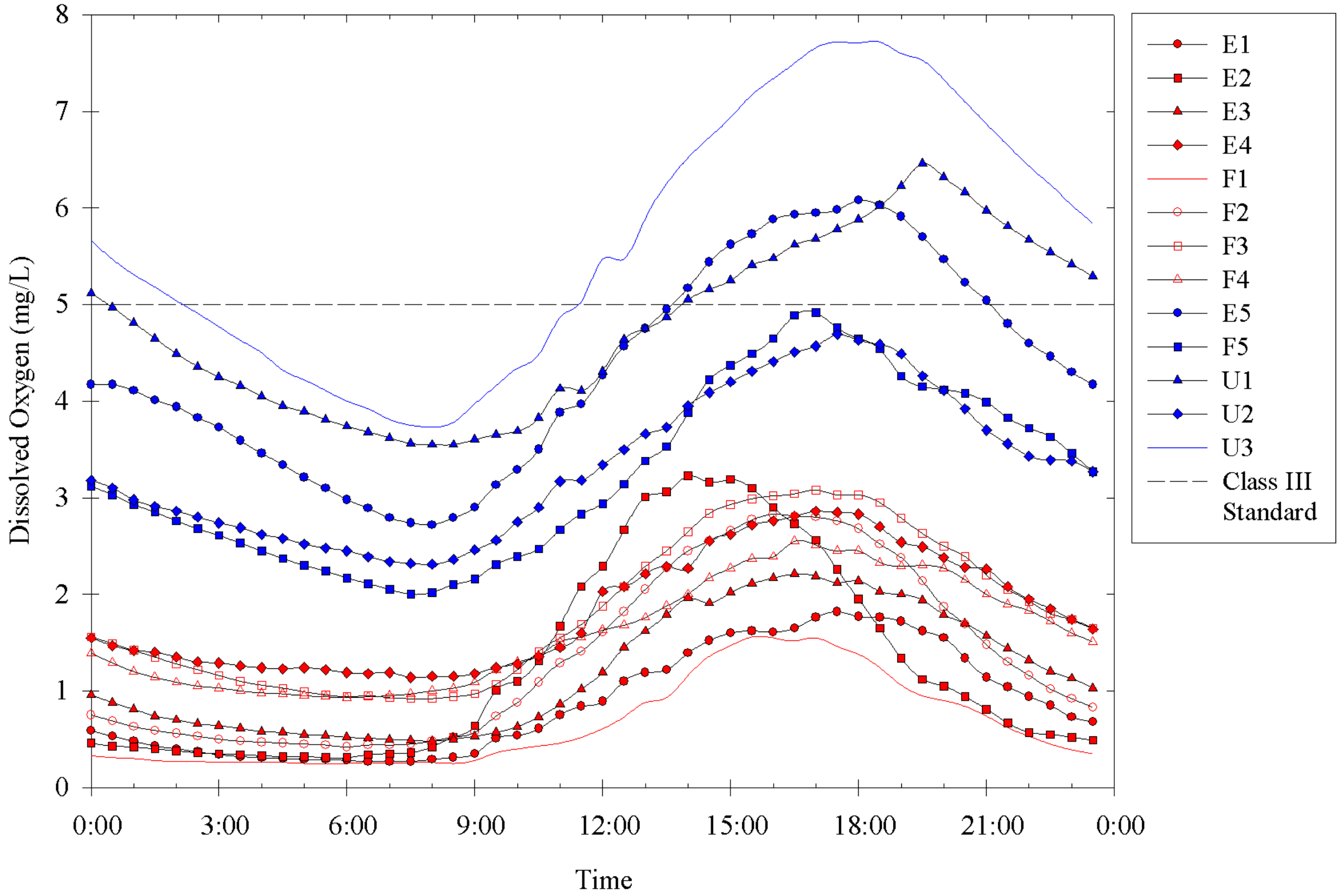

Figure 4-2-15. Mean dissolved oxygen diel curves at SFWMD transect sites in WCA-2A. Samples were collected on five occasions between April 25, 1995 and February 27, 1998. Red lines or symbols denote impacted sites, while blue lines or symbols denote background reference sites. Note: Curves indicate that all reference sites are in violation of the Class III standard for some portion of the day.

Section 4.0 Model Development

Historically, numeric water quality criteria have been defined by single point thresholds, annual averages, or simple relationships with other parameters. The current state Class III dissolved oxygen standard is an example of a single point threshold. At first appraisal, it seems reasonable to set an alternative criterion at a new point reflective of background conditions. However, due to wide daily fluctuations in marsh dissolved oxygen concentrations, as discussed in Section 2.3, a single point criterion is not appropriate. Therefore, to realistically represent the natural background dissolved oxygen regime in the marsh the SSAC must account for these daily fluctuations. For this reason, a mathematical model was developed that defines the dissolved oxygen SSAC as a cyclic sinusoidal function of time that accounts for the normal daily fluctuations. Seasonal variations were addressed through cross-validation and calibration of the model with grab data and incorporation of temperature into the final version of the model.

During the development of the dissolved oxygen SSAC, numerous models, using different combinations of sine and cosine functions of various periods (*e.g.*, 720 minutes, 2880 minutes, 8640 minutes) were investigated, in order to establish a balance between fit and simplicity of calculation. The final model was formulated as the summation of two sine functions, one with a period of 1440 minutes and a second with a period of 720 minutes. The regression model is described by the equation;

$$[DO_t] = a - [b \cdot \text{sine}\ (2\pi/1440 \cdot t) + (c \cdot \text{sine}\ (4\pi/1440 \cdot t))] \qquad (1)$$

where t ranges from 0 to 1440 minutes and DO_t is the dissolved oxygen concentration at time t. The a, b, and c appearing in formula (1) represent the unknown coefficients which are to be estimated from the diel reference data in a manner which forces the model to provide "a best fit" to the available data. Values of a, b, and c determine the daily minimum and maximum. Additionally, the value of c influences the timing of minimum and maximum.

In addition to simulating the dissolved oxygen concentration, the use of the saturation deficit was investigated to partially account for the influence of temperature and seasonality. The saturation deficit [SD] is the difference between the measured dissolved oxygen concentration and the concentration at saturation and can be expressed by the formula;

$$[SD] = DO - DO_{sat} \tag{2}$$

where $[DO_{sat}]$ is the saturation concentration which is a function of temperature given by the formula;

$$\ln [DO_{sat}] = -139.34411 + (1.575701 \cdot 10^5/K) - (6.642308 \cdot 10^7/K^2) + (1.2438 \cdot 10^{10} /K^3) - (8.621949 \cdot 10^{11}/K^4) \tag{3}$$

where DO_{sat} is the equilibrium oxygen concentration at 101.325 kPa and K is the water temperature on the Kelvin scale (K = °C + 273.15) (APHA 1992). The formula assumes one atmosphere of pressure at sea level, an assumption that should hold within reason for the Everglades given the low elevation of the region. In addition to temperature, saturation concentration is influenced by chlorinity or salinity. However, the effects of these are negligible for low salinity freshwater such as that found in Everglades marshes.

The model formula for deficit was similar to formula (1), with DO_t replaced by SD_t and is defined by the equation;

$$[SD_t] = a - [b \cdot \text{sine}\ (2\pi/1440 \cdot t) + (c \cdot \text{sine}\ (4\pi/1440 \cdot t))] \qquad (4)$$

where SD_t is the saturation deficit at time *t*.

Concentrations and deficits were initially averaged by sampling time across date (**Table 4-2-3**). A least squares regression model incorporating the relationship of dissolved oxygen or deficit with time of day, expressed as minutes past midnight, was evaluated. The fit obtained for dissolved oxygen concentrations was slightly better than that for DO saturation deficits (**Figures 4-2-16** and **17**). The least squares results in **Figures 4-2-16** and **4-2-17** provide estimates of a, b, and c in formulas (1) and (4), which define diel dissolved oxygen and saturation deficit models. Using the values of a, b, and c from **Figure 4-2-16** provides the equation;

$$DO_t = 4.36 - [1.37 \cdot \text{sine}\ (2\pi/1440 \cdot t) - (0.28 \cdot \text{sine}\ (4\pi/1440 \cdot t))] \qquad (5)$$

as the predictive model for the mean diel dissolved oxygen concentrations at background reference sites. Similarly, the diel saturation deficit at background reference sites at time *t* can be estimating using the equation;

$$SD_t = -3.70 - [1.50 \cdot \text{sine}\ (2\pi/1440 \cdot t) - (0.30 \cdot \text{sine}\ (4\pi/1440 \cdot t))] \qquad (6)$$

when the estimates from **Figure 4-2-17** are used as the values of a, b, and c.

Table 4-2-3. Average dissolved oxygen concentration and saturation deficit used to calculate "best fit" coefficients to formula (1).

Time	Dissolved Oxygen Concentration		Saturation Deficit		N
	Mean	95% Confidence Interval	Mean	95% Confidence Interval	
0	4.11	0.61	-3.98	0.49	18
30	4.00	0.62	-4.10	0.49	18
60	3.89	0.63	-4.22	0.50	18
90	3.80	0.64	-4.33	0.51	18
120	3.71	0.66	-4.42	0.53	18
150	3.62	0.67	-4.53	0.54	18
180	3.52	0.69	-4.63	0.55	18
210	3.43	0.70	-4.74	0.56	18
240	3.33	0.71	-4.84	0.56	18
270	3.25	0.71	-4.93	0.57	18
300	3.18	0.72	-5.01	0.57	18
330	3.11	0.72	-5.08	0.57	18
360	3.04	0.73	-5.17	0.57	18
390	2.98	0.72	-5.24	0.56	18
420	2.94	0.71	-5.28	0.55	18
450	2.97	0.69	-5.25	0.52	18
480	3.05	0.67	-5.16	0.49	18
510	3.20	0.65	-5.01	0.48	18
540	3.33	0.64	-4.86	0.46	18
570	3.46	0.63	-4.71	0.45	18
600	3.66	0.59	-4.49	0.42	18
630	3.76	0.57	-4.34	0.41	19
660	4.01	0.58	-4.08	0.43	21
690	4.25	0.48	-3.79	0.37	19
720	4.64	0.56	-3.43	0.41	20
750	4.65	0.48	-3.33	0.39	19
780	4.89	0.45	-3.03	0.39	20
810	5.16	0.48	-2.75	0.44	18
840	5.32	0.50	-2.57	0.47	18
870	5.46	0.51	-2.41	0.49	18
900	5.62	0.51	-2.24	0.51	18
930	5.79	0.51	-2.06	0.52	18
960	5.89	0.51	-1.96	0.52	18
990	5.94	0.54	-1.91	0.56	18
1020	5.93	0.52	-1.93	0.56	18
1050	5.92	0.54	-1.97	0.58	18
1080	5.83	0.53	-2.08	0.57	18
1110	5.74	0.54	-2.19	0.57	18
1140	5.59	0.53	-2.37	0.53	18
1170	5.42	0.54	-2.56	0.53	18
1200	5.24	0.54	-2.76	0.51	18
1230	5.03	0.54	-2.98	0.49	18
1260	4.87	0.54	-3.16	0.48	18
1290	4.73	0.56	-3.31	0.48	18
1320	4.59	0.56	-3.46	0.47	18
1350	4.46	0.57	-3.60	0.46	18
1380	4.32	0.58	-3.75	0.47	18
1410	4.20	0.59	-3.88	0.47	18

$DO_t = a - [b \cdot \text{sine}(2\pi/1440 \cdot t) + (c \cdot \text{sine}(4\pi/1440 \cdot t))]$

Sum of Residuals = -0.297
Average Residual = $-6.191 \cdot 10^{-3}$
Residual Sum of Squares (Absolute) = 1.114
Residual Sum of Squares (Relative) = 1.069
Standard Error of the Estimate = 0.154
Coefficient of Multiple Determination (R^2) = 0.976
Adjusted coefficient of multiple determination (Ra^2) = 0.975
Durbin-Watson statistic = 0.125

Variable	Value	Standard Error	t-ratio	Prob(t)
a	4.35682	0.02279	191.156	0
b	1.36701	0.03297	41.4579	0
c	-0.2753	0.0315	-8.741	0

Source	DF	Sum of Squares	Mean Square	F Ratio	Prob(F)
Regression	2	43.6002	21.8001	917.8338	0
Error	45	1.06883	0.02375		
Total	47	44.669			

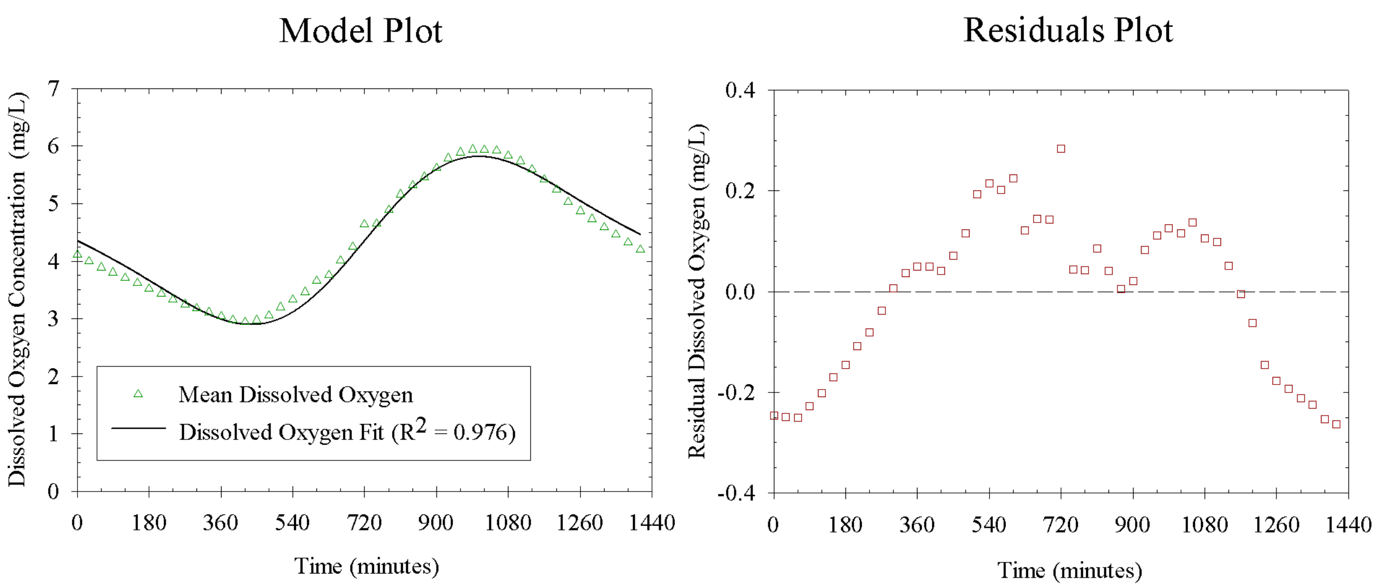

Figure 4-2-16. Fit statistics and plots from least squares analyses performed on mean diel dissolved oxygen concentrations. Least squares analysis was performed using DataFit V. 6.0.10 software for the PC.

$$SD_t = a - [b \cdot \text{sine}(2\pi/1440 \cdot t) + (c \cdot \text{sine}(4\pi/1440 \cdot t))]$$

Sum of Residuals = -0.394
Average Residual = $-8.200 \cdot 10^{-3}$
Residual Sum of Squares (Absolute) = 1.517
Residual Sum of Squares (Relative) = 1.600
Standard Error of the Estimate = 0.189
Coefficient of Multiple Determination (R^2) = 0.969
Adjusted coefficient of multiple determination (Ra^2) = 0.968
Durbin-Watson statistic = $6.56 \cdot 10^{-2}$

Variable	Value	Standard Error	t-ratio	Prob(t)
a	-3.69702	0.027289	-135.478	0
b	1.497328	0.041104	36.42792	0
c	-0.30432	0.038681	-7.86731	0

Source	DF	Sum of Squares	Mean Square	F Ratio	Prob(F)
Regression	2	50.46585	25.23292	709.5284	0
Error	45	1.600333	0.035563		
Total	47	52.06618			

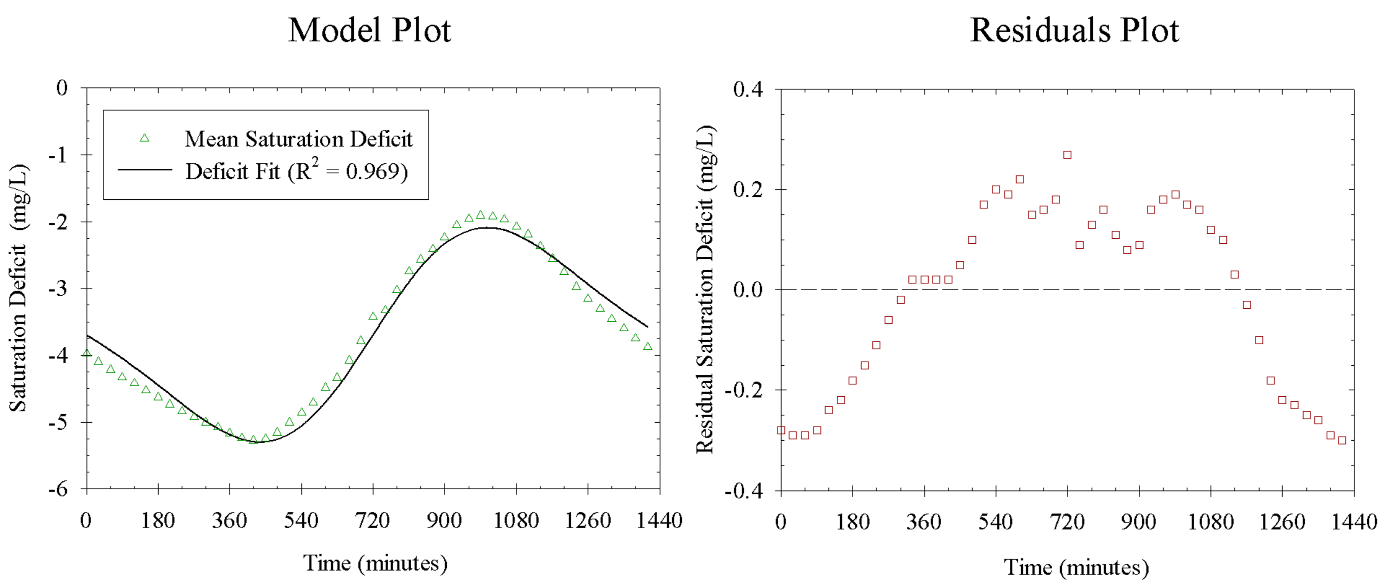

Figure 4-2-17. Fit statistics and plots from least squares analyses performed on mean diel saturation deficit concentrations. Least squares analysis was performed using DataFit version 6.0.10 software for the PC.

The analyses of the residuals (difference between model estimated value and measured value) shown in **Figures 4-2-16** and **4-2-17** show a trend, in that the model consistently under predicts and over predicts average concentration for periods during the day, rather than showing a random distribution around zero. This undesirable trend or pattern suggests that equations (5) and (6) can be improved to provide a better fit to the measured data. However, the practical applications of the model are also an important consideration. Although it is possible to achieve a better fit by using a more complex model, the benefits of the improved fit may not be sufficient to offset the costs of increased computational complexity. For example, the equation

$$[DO_t] \text{ or } [SD_t] = a - [b \cdot \text{cosine}\ (2\pi/1440 \cdot t) + c \cdot \text{cosine}\ (4\pi/1440 \cdot t) + d \cdot \text{sine}\ (2\pi/1440 \cdot t) + e \cdot \text{sine}\ (4\pi/1440 \cdot t)] \quad (7)$$

provides a better fit (DO R^2=0.997; SD R^2=0.998) to the data and reduces large patterns in the residuals. However, given the small magnitude of the residuals and the added computation complexity of formula (7), fits with formulas (5) and (6) were deemed sufficient for the purposes of this model. Because the magnitude of residuals, particularly during the usual period of grab sampling (540-820 minutes) are largely within the error of the current instrumentation (± 0.2 mg/L), additional model accuracy would yield few benefits and is arguably impossible to achieve with current sampling equipment. It was decided that the added complexity was not adequately offset by the small predictive improvement.

4.1 Recalculation of Saturation Deficit Model

The saturation deficit model provided by equation (4) does not directly yield a dissolved oxygen concentration. Rather it requires conversion of dissolved oxygen and temperature measurements into saturation deficits. This extra computation adds complexity and potential error to the application of the model. Therefore, in order to simplify its application, the saturation deficit model can be algebraically reordered to directly yield a predicted dissolved oxygen concentration. As previously defined, the saturation deficit is given by;

$$[SD] = DO - DO_{sat}, \quad (3)$$

where DO_{sat} is defined by equation (2). Additionally, it was demonstrated in Section 4.0 that the saturation deficit at time t (SD_t) can be predicted by;

$$[SD_t] = a - [b \cdot \text{sine}\,(2\pi/1440 \cdot t) + (c \cdot \text{sine}\,(4\pi/1440 \cdot t))]. \quad (4)$$

Therefore, by substituting equation (3) for $[SD_t]$ in equation (4) it may be concluded that;

$$DO_t - DO_{sat,t} = a - [b \cdot \text{sine}\,(2\pi/1440 \cdot t) + (c \cdot \text{sine}\,(4\pi/1440 \cdot t))] \quad (8)$$

or

$$DO_t = a - [b \cdot \text{sine}\,(2\pi/1440 \cdot \mathrm{t}) + (c \cdot \text{sine}\,(4\pi/1440 \cdot \mathrm{t}))] + DO_{sat,t}, \quad (9)$$

where DO_t and $DO_{sat,t}$ are the observed dissolved oxygen concentration and the dissolved oxygen concentration at saturation at time t, respectively. When graphically plotted formula (9) provides a surface that conceptually represents the relationships between dissolved oxygen and time of day and temperature.

The saturation concentration calculation (equation 3) is overly complex for the predictive needs relative to the dissolved oxygen SSAC. In other words, equation (3) provides greater accuracy than can be feasibly measured over the entire range of surface water temperatures (0-100°C). Equation (3) can be simplified when the temperature range is constrained. For temperatures between 6° and 42° C the saturation concentration can be expressed as;

$$[DO_{sat}] = 1/(0.0683 + 0.00198 \cdot C + 5.24{\cdot}10^{-6} \cdot C^2), \tag{10}$$

where C is defined as the water temperature in °C. Formula (10) was determined using a least squares fit against formula (3) for temperatures between 6° and 42° C. When constrained to the specified temperature range, formula (10) provides an excellent approximation ($R^2 \approx 1.00$; $p<0.0001$) of the saturation concentration. The temperature range was selected to bracket the observed range in the 1994 to 1999 grab sample data (8.3 - 39.1° C) with a small additional margin for future extreme values. Substitution of (10) into (9) provides a simplified calculation for DO_t of the form

$$DO_t = -3.70 - [1.50 \cdot \text{sine}\,(2\pi/1440 \cdot t) - (0.30 \cdot \text{sine}\,(4\pi/1440 \cdot t))] + 1/(0.0683 + 0.00198 \cdot C + 5.24{\cdot}10^{-6} \cdot C^2). \tag{11}$$

Using equation (11), the dissolved oxygen prediction becomes a surface defined by time of day and temperature (**Figure 4-2-18**).

4.2 Final Average Background Condition Models

Based on the diel reference data two competing models were selected as providing a combination of good predictive fit and reasonable ease of calculation. Equations (5) and (11) define the dissolved oxygen concentration and the adjusted saturation deficit models, respectively. Based on the diel reference dataset, these two models have nearly equivalent fits. Since the compliance model will be applied to data collected from grab samples, selection of the final model was based upon cross-validation and review of characteristics relative to the grab data. Cross-validation was necessary to confirm that the final model provided an acceptable prediction of dissolved across the entire geographic extent of the Everglades.

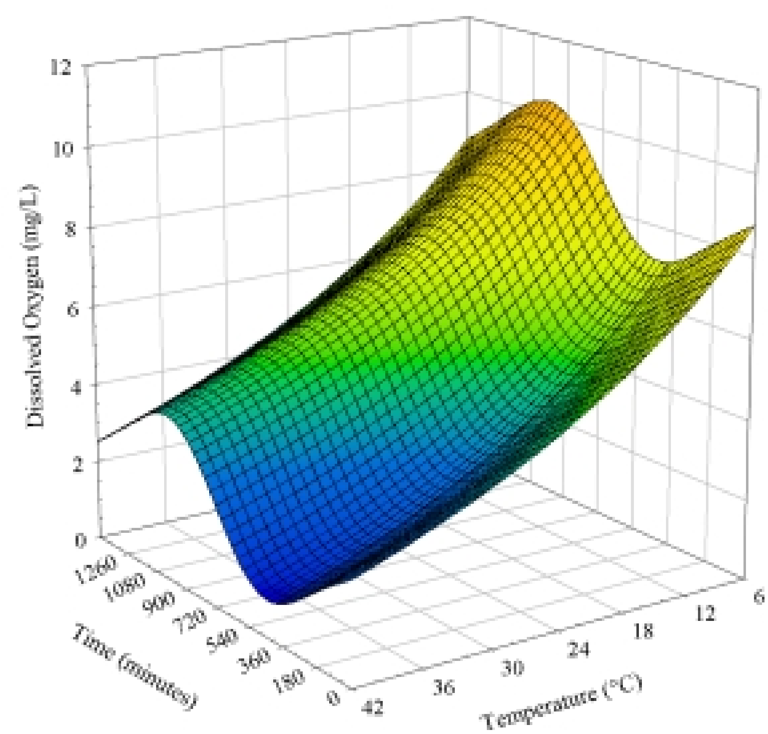

Figure 4-2-18. Plot of predicted dissolved oxygen concentrations provided by the adjusted deficit model.

4.3 Cross-Validation with Grab Reference

The two competing models provide predicted dissolved oxygen concentrations for a specific sample time (t). The saturation adjusted deficit model incorporates some additional complexity by factoring in the influence of temperature (C). Therefore, to compare the values predicted by these equations with measured values, the collection time (t_{ij}) and water temperature (C_{ij}) measurements must also be reported. Using the values of t_{ij} and $C_{ij,}$ predicted concentrations can be generated using equations (5) and (11) such that;

$$DO_{ij} = 4.36 – [1.37 \cdot \text{sine}\ (2\pi/1440 \cdot t_{ij}) - (0.28 \cdot \text{sine}\ (4\pi/1440 \cdot t_{ij}))] \quad (12)$$

or

$$DO_{ij} = -3.70 – [1.50 \cdot \text{sine}\ (2\pi/1440 \cdot t_{ij}) - (0.30 \cdot \text{sine}\ (4\pi/1440 \cdot t_{ij}))] + 1/(0.0683 + 0.00198 \cdot C_{ij} + 5.24 \cdot 10^{-6} \cdot C_{ij}^{2}), \quad (13)$$

where t_{ij} and C_{ij} are the sample collection time and water temperature, respectively, of the i^{th} dissolved oxygen measurement at the j^{th} station. All DO_{ij} are the population of predicted values for the grab reference sites. An example, using data collected at site CA315 during 1999, will help illustrate the comparisons between grab data and the models.

Twenty-two samples were collected at site CA315 in 1999 each with the associated collection time and water temperature (**Table 4-2-4**). For the first sample (i^{th} measurement) collected in 1999 at CA315 (j^{th} station), t_{ij} and C_{ij} were 605 minutes and 11.94° C, respectively. By substituting the value of 605 minutes for t_{ij} into (12), the dissolved oxygen model prediction is calculated as;

$$4.36 – [1.37 \cdot \text{sine}\ (2\pi/1440 \cdot \underline{605}) – (0.28 \cdot \text{sine}\ (4\pi/1440 \cdot \underline{605}))] = 3.47 \text{ mg/L}. \quad (12a)$$

The adjusted deficit model prediction for the same sample at CA315 is expressed as;

$$-3.70 - [1.50 \cdot \text{sine}(2\pi/1440 \cdot \underline{605}) - (0.30 \cdot \text{sine}(4\pi/1440 \cdot \underline{605}))] + 1/(0.0683 + 0.00198 \cdot \underline{11.94} + 5.24\cdot 10^{-6} \cdot \underline{11.94}^2) = 6.12 \text{ mg } O_2/\text{L}. \quad (13a)$$

Table 4-2-4. Dissolved oxygen and temperature measurements taken at site CA315 in 1999 with dissolved oxygen model and adjusted deficit model predicted dissolved oxygen values. Samples collected during Daylight Savings Time were adjusted to Eastern Standard Time.

Date	Reported Sample Time	Minute	EST Corrected Minute	Temperature (°C)	Observed DO (mg/L)	Predicted DO (mg/L)	Adjusted Deficit Model (mg/L)
1/6/99	10:05	605	605	11.94	7.17	3.47	6.12
1/20/99	9:50	590	590	21.18	5.55	3.37	4.11
2/2/99	9:10	550	550	20.73	4.13	3.16	3.95
2/18/99	9:15	555	555	18.67	4.99	3.18	4.35
3/1/99	9:28	568	568	17.66	5.51	3.25	4.62
3/17/99	10:42	642	642	18.15	3.04	3.73	5.05
3/30/99	9:12	552	552	20.83	1.21	3.17	3.94
4/14/99	10:14	614	554	23.06	0.71	3.18	3.58
5/12/99	9:20	560	500	21.67	0.67	2.98	3.59
6/24/99	9:30	570	510	27.30	3.07	3.00	2.74
7/6/99	9:32	572	512	28.18	3.67	3.01	2.62
7/22/99	10:02	602	542	28.75	1.17	3.12	2.67
8/2/99	9:10	550	490	29.70	1.33	2.95	2.35
8/18/99	9:43	583	523	28.52	1.90	3.05	2.62
8/31/99	8:45	525	465	28.52	0.61	2.91	2.47
9/28/99	10:22	622	562	27.22	2.13	3.21	2.98
10/11/99	9:12	552	492	26.59	1.65	2.96	2.79
10/25/99	9:30	570	510	21.81	4.06	3.00	3.59
11/8/99	9:02	542	542	21.94	5.82	3.12	3.70
11/23/99	9:00	540	540	22.50	4.61	3.11	3.60
12/9/99	10:00	600	600	20.73	4.07	3.43	4.25
12/20/99	9:00	540	540	21.33	2.81	3.11	3.80
Annual Mean					3.18	3.16	3.61

The statistical problem of cross-validating the models with the grab samples is equivalent to the classical problems of comparing two populations using t-tests and correlation analysis. If the models are good predictors of grab sample dissolved oxygen concentrations across the Everglades, the population of reference grab samples will be both correlated with and not statically different form the population of all DO_{ij}.

The Student's t-test was used to test the hypothesis that the DO and adjusted deficit models provided mean predictions equal to the mean observed dissolved oxygen concentrations at all grab reference sites on both the entire dataset and as an annual average. On both a point by point (t-test; $p<0.0001$) and annual basis (t-test; $p<0.0001$), the DO model predictions were significantly different from the grab samples (**Table 4-2-5**). Conversely, the adjusted deficit model predictions were equal to the observed grab samples when compared to both the entire data set ($p=0.93$) and as annual average ($p=0.23$) (**Table 4-2-6**). Based on t-test analysis the adjusted deficit model appears to give a good estimate of mean dissolved oxygen for grab samples. However, the DO model is a poor predictor.

Table 4-2-5. T-test results for DO model predictions versus the observed dissolved oxygen concentrations at reference grab sites in the EPA between 1994 and 1999.

DO Measure	N	Mean (mg/L)	Standard Deviation	Standard Error of the Mean	P
Observed DO	2821	3.82	2.07	0.039	
DO Model	2821	3.52	0.59	0.011	<0.0001
Observed Annual Mean	223	3.94	1.29	0.086	
Annual Mean DO Model	223	3.55	0.38	0.025	<0.0001

Table 4-2-6. T-test results for adjusted deficit model predictions versus the observed dissolved oxygen concentrations at reference grab sites in the EPA between 1994 and 1999.

DO Measure	N	Mean (mg/L)	Standard Deviation	Standard Error of the Mean	P
Observed DO	2821	3.82	2.07	0.039	
Adjusted Deficit Model	2821	3.81	1.04	0.02	0.93
Observed Annual Mean	223	3.94	1.29	0.086	
Annual Mean Adjusted Deficit Model	223	3.83	0.38	0.026	0.23

Correlation analysis provides a measure of relationship between two or more variables. In the case of evaluating the models, a statically significant and positive correlation between observed concentrations and predicted values is required. Correlations were calculated between the entire set of reference grab data and both models. Both correlation analyses were statically significant ($p<0.0001$). The relationship between observed and predicted was higher for the adjusted deficit model (R^2= 0.253) than the DO model (R^2=0.196) (**Figure 4-2-19**).

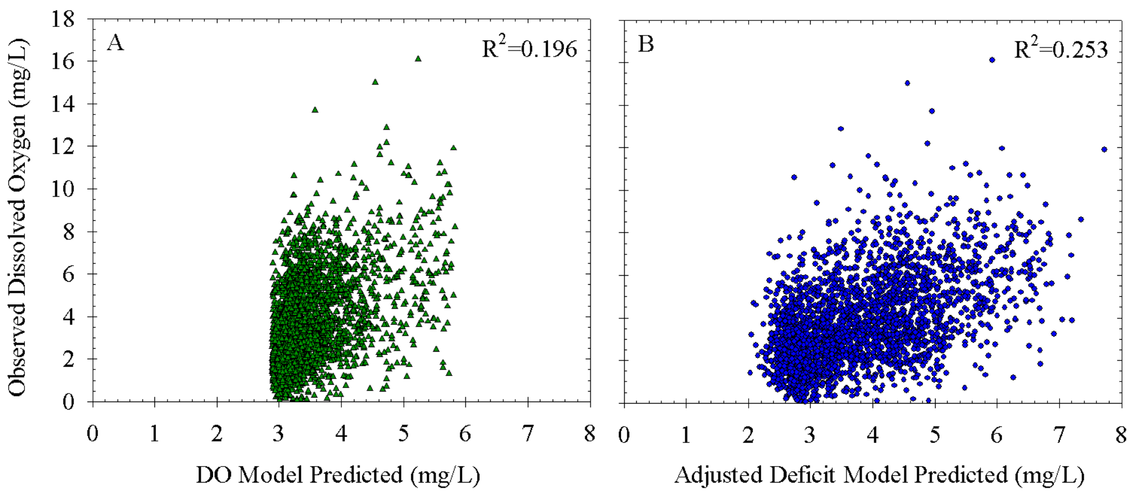

Figure 4-2-19. DO (A) and adjusted deficit (B) model predictions versus observed dissolved oxygen concentrations at grab reference sites in the Everglades Protection Area. Grab samples were collected between 1994 and 1999.

In addition to statistical tests of equivalence and correlation, the model must be free of temporal biases on monthly or seasonal scales. Potential biases of this nature can be revealed through the review of residuals relative to a time series. Residuals are given as

$$ODO_{ij} - DO_{ij}, \tag{14}$$

where ODO_{ij} is the i^{th} dissolved oxygen measured at the j^{th} station and DO_{ij} is the model predicted, using (5) or (11), dissolved oxygen concentration for the given sample. If the model predicts mean dissolved oxygen then the residuals will average to zero; that is the number of measurements above the model will be balanced by those below the model. Additionally, a time series of the residuals should show no pattern. Plots of the cross-validation residuals versus day of the year show the DO model is seasonally biased, while the adjusted deficit model is not biased (**Figure 4-2-20**). The DO model tends to under predict in January, February, March, November and December and over predict in April through September.

The adjusted deficit model proved to be a better predictor of dissolved oxygen concentration in the marsh. Based on the cross-validation process using grab samples, the adjusted deficit model described by equation (11) is the recommended model. It provides an acceptable unbiased prediction of the mean background dissolved oxygen regime in the marshes of the Everglades.

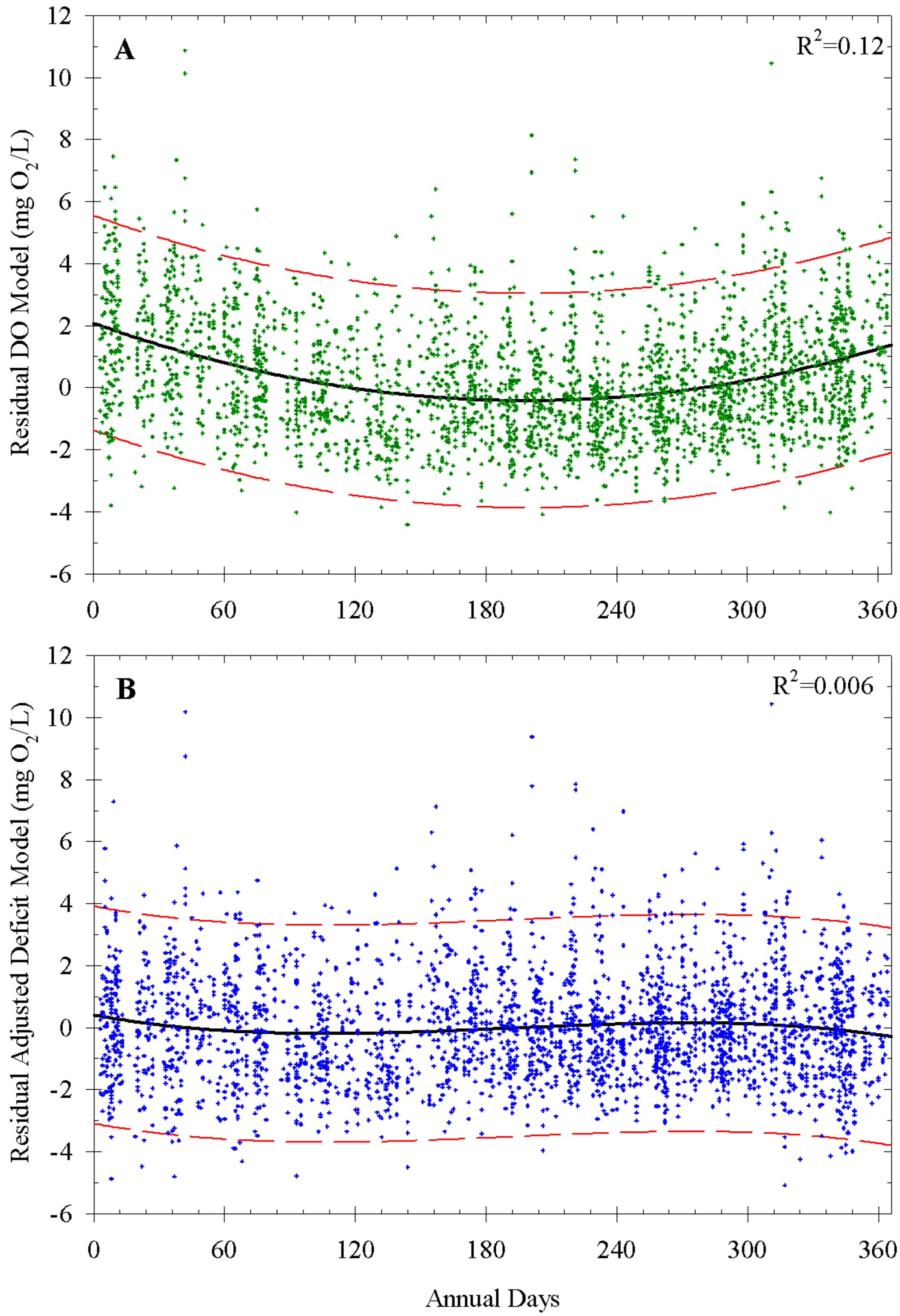

Figure 4-2-20. Cross-validation residuals (grab observed – model prediction) for DO (A) and adjusted deficit (B) models versus day of year. Solid curves indicated third order line of best-fit. The 95% prediction intervals are shown by the red dashed lines.

5.0 Compliance Determination

The recommended predictive model was derived from a single set of data composed of dissolved oxygen measurements collected at a series of reference sites over a number of monitoring years and during various times of the year. Therefore, the resulting model essentially provides a prediction of the average annual dissolved oxygen condition within minimally impacted areas of the EPA. Realistically, over a 12 month period, dissolved oxygen concentrations both above and below the model predicted average annual condition are expected to occur naturally due to factors such as changes in cloud cover, rainfall, water depth, succession of biological communities, etc. Therefore, the issue of compliance becomes a statistically testable hypothesis of whether the difference between observed and predicted dissolved oxygen values would have a reasonable likelihood of occurring due to natural background variability. Thus, to judge compliance with a SSAC based on the recommended model, it is necessary to calculate or define the allowable frequency and magnitude of observations below the curve. In this case, if an observed negative difference is so large as to have less than a 10% chance of occurring under background conditions, then the hypothesis will be rejected and the site will be deemed as out of compliance with the SSAC.

The statistical problem of calculating a 10% lower rejection limit for compliance data is equivalent to the problem of calculating a prediction interval below which there is less than a 10% chance that new observations come from the same distribution as previously collected data. The lower prediction interval (PI_l) can be calculated by the equation:

$$PI_l = \overline{X} - t_{(\alpha,n-1)} \cdot \sqrt{S^2 + (S^2 / n)} \qquad (15)$$

where S and n are the standard deviation and number of the individual observations, respectively, and $t_{(\alpha,n-1)}$ is the one-tailed t-test statistic for a probability of α with n-1 degrees of freedom.

Since the issue of compliance is concerned with the difference between the observed and predicted values, residuals (*i.e.*, observed – predicted values) were used to calculate the lower prediction interval. Residuals provide the distribution of grab samples around the model curve and show how often observations are below the curve. The adjusted deficit model was used to predict the dissolved oxygen concentration for each reference grab sample collected (Appendix 4-IIA). The observed and predicted dissolved oxygen concentrations, as well as, the residuals were then averaged annually for each reference station by year. The average annual residual was 0.11 mg/L, indicating that on an annual basis, the model slightly under predicted the observed values. To compensate for this random error, the deficit model was recalibrated to the grab data by adding a constant 0.11 mg/L to the model predictions (equation 16).

Using residuals, the annual prediction interval of the grab samples was calculated at α=0.10 as summarized in **Table 4-2-7**.

Table 4-2-7. Calculation of lower prediction interval for the adjusted deficit model based on grab samples collected at reference sites in the EPA, between 1994 and 1999.

Statistic	Value
Mean Residual	0.11
Standard Deviation of Residuals	1.157
N	223
n-1	222
$t_{(0.1,222)}$	1.285
Prediction Interval	1.491

$$PI_1 = -1.285 \bullet \sqrt{1.157^2 + (1.157^2 / 223)} = \overline{X} - 1.491 \qquad (16)$$

The lower compliance limit (DO_{LL}) for the model predictions can then be calculated by substituting the recalibrated prediction equation for $\overline{X}$ in equation 15. The lower dissolved oxygen compliance limit, which will be the SSAC, can therefore be expressed as:

$$\begin{aligned} DO_{LL} &= [-3.70 - [1.50 \cdot \text{sine}\,(2\pi/1440 \cdot t) - (0.30 \cdot \text{sine}\,(4\pi/1440 \cdot t))] + \\ &\quad 1/(0.0683 + 0.00198 \cdot C + 5.24{\cdot}10^{-6} \cdot C^2)] + 0.11 - 1.49 = \\ &\quad [-3.70 - [1.50 \cdot \text{sine}\,(2\pi/1440 \cdot t) - (0.30 \cdot \text{sine}\,(4\pi/1440 \cdot t))] + \\ &\quad 1/(0.0683 + 0.00198 \cdot C + 5.24{\cdot}10^{-6} \cdot C^2)] - 1.38, \qquad (17) \end{aligned}$$

where C is water column temperature in °C and t is sample collection time standardized to EST (**Figure 4-2-21**).

Compliance with the SSAC will be determined using the average annual residual (*i.e.*, difference between the observed dissolved oxygen concentrations (measured) and the predicted lower DO compliance limit provided by equation 17) for each site. If the annual average residual is greater than or equal to zero, there is reasonably likelihood that conditions are within natural background variability and the site is in compliance. Likewise, if the annual average residual is less than zero, then the annual average measured dissolved oxygen concentration is below the range explained by natural background variability (at α=0.1) and the site is considered as out of compliance with the SSAC. An example compliance calculation is given in **Table 4-2-8** for site CA315 for the 1999 monitoring year. The average annual difference between the measured dissolved oxygen levels and the predicted lower compliance limit (equation 17) is 0.94 mg/L. Therefore, the annual average measured dissolved oxygen concentration is above the minimum level that can be reasonably expected to be accounted for by natural background conditions (at α=0.1), so site CA315 was in compliance with the SSAC during 1999.

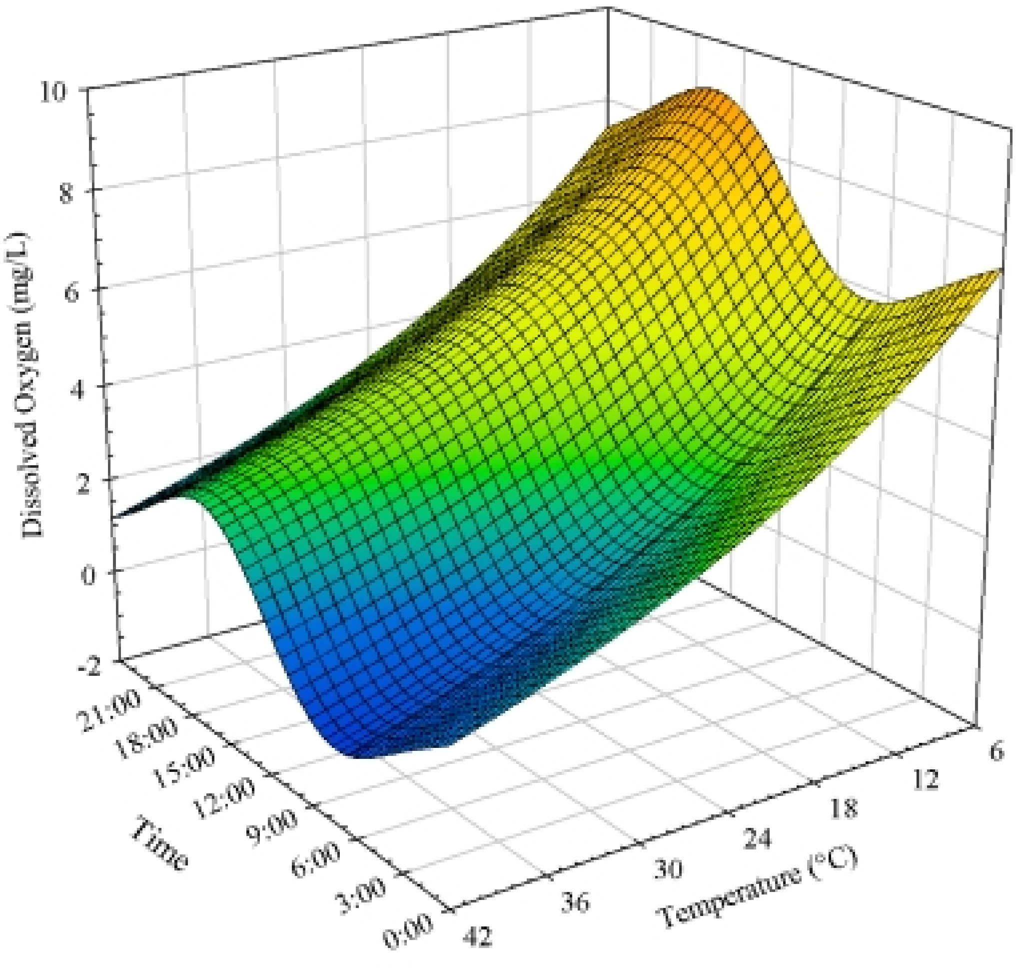

Figure 4-2-21. Annual average dissolved oxygen compliance limit for interior marsh stations in the Everglades Protection Area. Note: Time is standardized to Eastern Standard Time.

Table 4-2-8. Example annual dissolved oxygen threshold calculation using site CA315 during 1999. Note: That the average annual difference between the observed dissolved oxygen concentrations and the predicted lower dissolved oxygen compliance limit is 0.95 mg/L, therefore the site is considered within compliance.

Date	Time	EST Corrected* (min)	Temperature (°C)	Observed DO (mg/L)	Lower DO Compliance Limit (DO_{LL})	Residual (Observed – DO_{LL})
1/6/99	10:05	605	11.94	7.17	4.74	2.43
1/20/99	9:50	590	21.18	5.55	2.73	2.82
2/2/99	9:10	550	20.73	4.13	2.57	1.56
2/18/99	9:15	555	18.67	4.99	2.97	2.02
3/1/99	9:28	568	17.66	5.51	3.24	2.27
3/17/99	10:42	642	18.15	3.04	3.67	-0.63
3/30/99	9:12	552	20.83	1.21	2.56	-1.35
4/14/99	10:14	554	23.06	0.71	2.20	-1.49
5/12/99	9:20	500	21.67	0.67	2.21	-1.54
6/24/99	9:30	510	27.3	3.07	1.36	1.71
7/6/99	9:32	512	28.18	3.67	1.25	2.42
7/22/99	10:02	542	28.75	1.17	1.29	-0.12
8/2/99	9:10	490	29.7	1.33	0.98	0.35
8/18/99	9:43	523	28.52	1.90	1.24	0.66
8/31/99	8:45	465	28.52	0.61	1.09	-0.48
9/28/99	10:22	562	27.22	2.13	1.60	0.53
10/11/99	9:12	492	26.59	1.65	1.41	0.24
10/25/99	9:30	510	21.81	4.06	2.22	1.84
11/8/99	9:02	542	21.94	5.82	2.32	3.5
11/23/99	9:00	540	22.5	4.61	2.22	2.39
12/9/99	10:00	600	20.73	4.07	2.87	1.2
12/20/99	9:00	540	21.33	2.81	2.42	0.39
Annual Average				3.18	2.23	0.95

*Samples collected during Daylight Savings Time were adjusted to Eastern Standard Time

The SSAC and the associated compliance measurement methodology should balance type I and II errors. That is, reference sites should be classified as complying with the SSAC while impacted sites are identified as out of compliance. Applying the recommended SSAC and compliance test to the grab sample data finds 89.5 % and 7.6 % of the impacted and reference site-years, respectively, out of compliance (Appendix 4-IIB). None of the "unknown" sites were determined to be out of compliance for the 1994 to 1999 monitoring years.

These results suggest that the predicted lower compliance limit is functioning very close to the specified sensitivity of 10% (α=0.1) with 92.4% of the reference site-years being correctly identified as in compliance. Therefore, the compliance methodology appears to provide a reasonable balance of errors.

The SSAC can be applied to open water interior marsh stations within the Everglades. In order to account for the inherent variability in dissolved oxygen, compliance is meant to be determined as an annual average. As was demonstrated by the proceeding evaluation of type I and II errors, the current sampling regime (frequency and number) provides sufficient power to differentiate between impacted and unimpacted conditions. Additionally, since the variability of the grab samples was used to define the lower prediction limit, future compliance monitoring should maintain the current sampling methodology. Therefore, it is recommended that compliance with the SSAC be based on data collected during grab sampling conducted at least monthly. Although it is expected that in practice, monitoring data may fall short of 12 samples per year, a review of grab data shows that the SSAC and compliance methodology are not highly sensitive to missing data.

5.1 Impacts to Designated Use

The proposed dissolved oxygen SSAC is not expected to have any adverse impacts on surface waters of the Everglades. It was developed to be representative of natural background dissolved oxygen conditions in the marsh and will therefore be protective of indigenous flora and fauna as well as all designated uses. Similarly, designated uses of adjacent waters will not be adversely affected by the implementation of the proposed dissolved oxygen SSAC.

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