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# An Assessment of Seasonal Submerged Aquatic Vegetation (SAV) Epiphyte Loading at Blossom Point, Maryland

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## 1. ABSTRACT

Measurements of epiphyte fouling rates were made at one location, approximately one meter mean water depth, along each of three permanent transects (PR5, BP3, and KC1) at Blossom Point, Maryland. Measurements were made in the summer and fall of 1999, as well as the spring, summer, and fall of 2000, using artificial substrates in the form of rectangular Mylar® strips. This technique, indicated fouling rate estimates in the spring of 2000 were extremely low and did not contribute significantly to light attenuation at the leaf surface. As expected, epiphyte fouling rates were highest in the summer. In 2000, mean epiphyte accumulation after one week of exposure reduced the available light through the water column from 18.5% of surface irradiance to 14.9% at the leaf surface. In comparison, epiphyte accumulation rates in the upper mesohaline region of the Patuxent River (SVBA, SV02) reduced the available light through the water column from 8.9 % of surface irradiance to 6.5% at the leaf surface after a week of exposure. These fouling rates are small compared to those found at other locations in mesohaline waters where available light can be reduced by more than 20% of surface radiation after a weeks time. The inorganic component of epiphytic material was significantly higher in 1999 compared to 2000. Dissolved nutrient concentrations in the water column were variable over time and showed significant seasonality. Median concentrations of dissolved inorganic phosphorus (DIP) at all stations remained above the 0.01 mg l<sup>-1</sup> concentration for the oligohaline habitat limits established by the USEPA. While no oligohaline submerged aquatic vegetation (SAV) habitat criteria exists for dissolved inorganic nitrogen (DIN), median concentrations ranged from 0.33 mg l<sup>-1</sup> (transect KC1) to 0.71 mg l<sup>-1</sup> (transect PR5).

## 2. INTRODUCTION

Chesapeake Bay, like many other shallow coastal estuaries worldwide, has experienced declines in submerged aquatic vegetation (SAV) populations during the last half of the twentieth century (Den Hartog and Polderman, 1975; Kemp *et al.*, 1983; Orth and Moore, 1983, 1984; and Cambridge *et al.*, 1986). While overall, coverage of SAV in Chesapeake Bay and tributaries remains well below historic levels (Moore *et al.*, 2000) certain areas have remained vegetated or have even recovered in recent years (Carter and Rybicki, 1986). Consequently, there is keen interest in preserving and protecting SAV populations where they exist.

In response to these changes, a variety of studies have suggested that increased anthropogenic inputs of dissolved nutrients and particulate matter have been primarily responsible for degraded water quality conditions and reduced light availability to rooted macrophyte populations (*e.g.* Sand-Jensen, 1977; Cambridge *et al.*, 1986; Kemp *et al.*, 1983; Twilley *et al.*, 1985; Silberstein, 1986). While light availability is generally agreed to be the most critical resource limiting the extent and distribution of SAV populations, an understanding of what conditions are necessary and sufficient to provide adequate light has proven to be most elusive. For example, a number of studies have demonstrated that epiphytes can substantially reduce the amount of available light reaching the leaf surface (*e.g.* Burt *et al.*, 1995; Stankelis *et al.*, 1999). However, epiphyte loads can be modified to a great extent by a variety of factors such as: epiphyte grazer density (*e.g.* Neckles *et al.*, 1993; Williams and Ruckelshaus, 1993), light availability (Stankelis *et al.*, 1999), nutrient availability (Kemp *et al.*, 1983; Burt *et al.*, 1995), wave action (*e.g.* Koch and Beer, 1996) and leaf turnover rates. The proposed construction of off-shore breakwaters along portions of the shoreline at the Adelphi Laboratory's Blossom Research Facility have the potential to impact healthy SAV populations at this location by altering a variety of these parameters. However, the extent of this impact is uncertain. In order to properly evaluate this impact it is necessary to assess the baseline conditions at this site prior to construction. This portion of the assessment has focused on the contribution of epiphyte accumulation on light attenuation to SAV.

In 1999, three monitoring sites were established along each of three permanent transects (PR5, BP3, and KC1) at approximately one meter mean water depth to evaluate epiphyte accumulation rates. Additional transects (KC02, BP04, and PR06) were established for other studies in this collaborative project and were not used in this study. Epiphyte accumulation rate measurements were made in the summer and fall of 1999 and the spring of 2000, under a separate contract. Data from the 1999 contract have not been included in this report but were included in the analyses. As a part of this contract, water column dissolved nutrients and epiphyte accumulation rates were measured in the summer and fall of 2000. Additional sampling will be conducted in 2001 prior to breakwater construction. This report provides a preliminary evaluation of fouling rates and light attenuation at these sites and compares these results to other regions within the Chesapeake Bay system.

## 3. METHODS

### 3.1. Water Quality Sampling

Water samples were collected independently by the US Army and Chesapeake Biological Laboratory (CBL) to insure that sufficient information was collected for an adequate analysis of the SAV habitat throughout the growing season. The analysis of water column dissolved nutrient concentrations from both sets of samples were completed at the Nutrient and Analytical Services Laboratory (NASL) and are included in this report.

#### 3.1.1. Station Locations

All water quality samples were collected along three fixed transects (KC1, BP3, and PR5) located at the Blossom Point Facility (Figure 3-1). The US Army collected water samples at two locations along each transect within and outside of existing SAV beds. Station codes for these data reflect the distance from shore at which water samples were collected. The Chesapeake Biological Laboratory (CBL) collected water quality samples from a single location along each transect adjacent to each epiphyte collection array at a water depth of approximately 1 meter average water depth (Table 3-1).

#### 3.1.2. Sampling Frequency

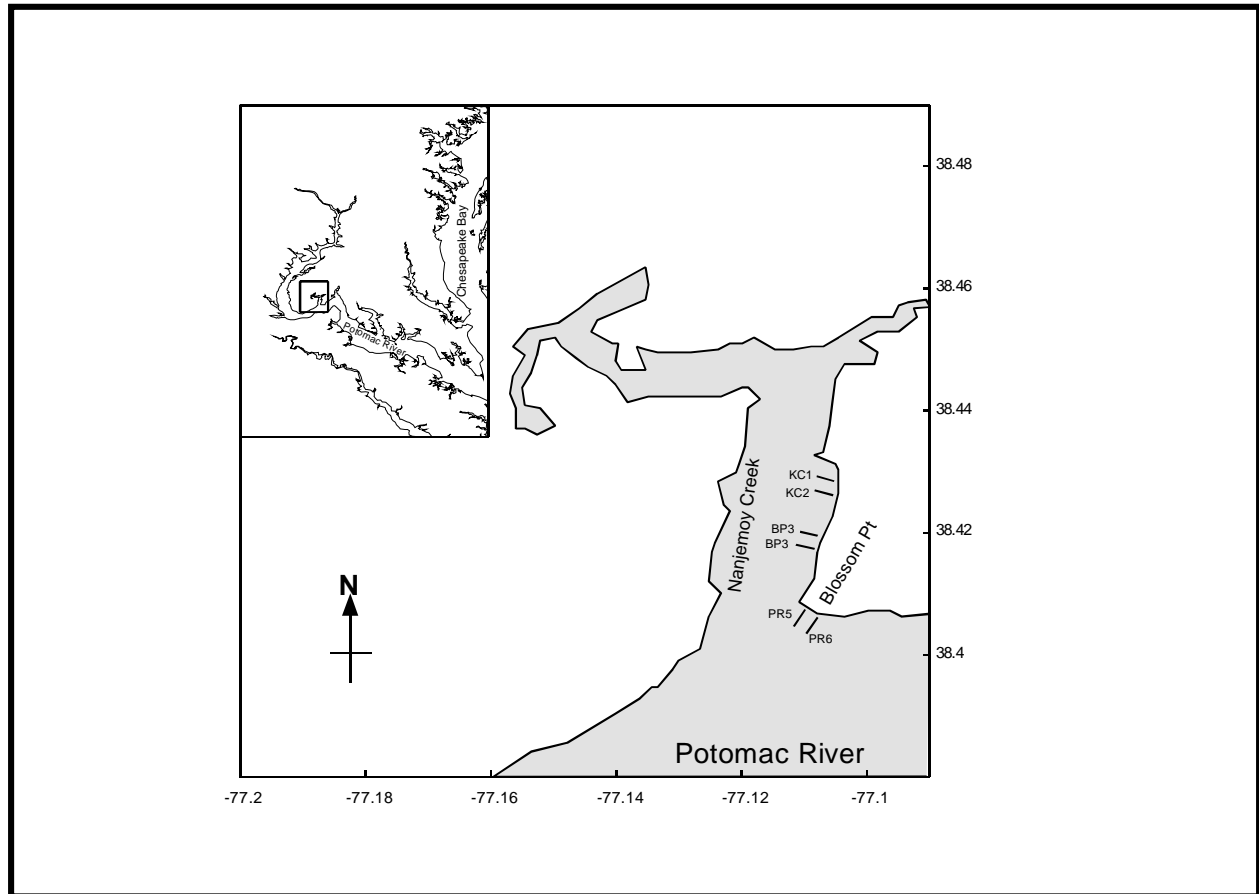
Sampling by CBL was conducted weekly from August 18 to September 1 and from September 22 to October 13, 2000 to coincide with the measurement of epiphyte fouling rates. A total of seven water samples were collected during this time by CBL. The US Army collected water samples approximately bi-weekly March through November 2000.

#### 3.1.3. Water Quality Methods

The following field procedures apply to data collected by CBL only. Water samples collected by the US Army may have followed separate procedures.

##### 3.1.3.1. Physical Parameters

Temperature, salinity, conductivity, and dissolved oxygen measurements were measured with a Yellow Springs International (YSI) 600R or YSI 6920 multi-parameter water quality monitor at 0.5 meters below the water surface. Water column turbidity was estimated with a secchi disk, while water column light flux in the photosynthetically active frequency range (PAR) was measured with a *Li-Cor* LI-192SA underwater quantum sensor. When possible, light flux



**Figure 3-1. Location of Submerged Aquatic Vegetation (SAV) epiphyte monitoring stations at Blossom Point, MD.**

*Latitude and longitude are in decimal degrees.*

**Table 3-1. Blossom Point: Submerged aquatic (SAV) station code and geographical coordinates.**

<b>STATION CODE</b>	<b>LATITUDE (DGPS) NAD 83</b>	<b>LONGITUDE (DGPS) NAD 83</b>
KC1	38° 01.620'	75° 50.509'
BP3	37° 58.249'	75° 52.609'
PR5	38° 08.835'	75° 50.349'

measurements were collected at three discrete water depths in order to calculate water column light attenuation (Kd). Weather and sea-state conditions such as air temperature, percent cloud cover, approximate wind speed and direction, total water depth, and wave height were also recorded.

### **3.1.3.2. Water Column Nutrients, Chlorophyll-*a* and Suspended Solids**

Whole water samples were collected by CBL with a hand pump at approximately 0.5 meters below the water surface. A portion was immediately filtered with a 25 mm, 0.7  $\mu\text{m}$  (GF/F) glass fiber filter. Both the filtered portion and the remaining whole water samples were placed in coolers for transport back to the laboratory for further processing. The filtered portion was analyzed by the Nutrient Analytical Services Laboratory (NASL) for ammonium ( $\text{NH}_4^+$ ), nitrate ( $\text{NO}_2^-$ ), nitrite plus nitrate ( $\text{NO}_2^- + \text{NO}_3^-$ ) and phosphate ( $\text{PO}_4^{-3}$ ). Whole water portions were filtered in the laboratory using 47 mm, 0.7  $\mu\text{m}$  (GF/F) glass fiber filters and were transferred to NASL for analysis of the following parameters: total suspended solids (TSS), total volatile solids (TVS), and total and active chlorophyll-*a* concentrations, where total chlorophyll-*a* includes chlorophyll-*a* plus breakdown products.

### **3.1.3.3. Chemical Analysis Methodology**

Methods for the determination of dissolved nutrients collected by both CBL and the US Army were as follows: ammonium ( $\text{NH}_4^+$ ), nitrite ( $\text{NO}_2^-$ ), nitrite plus nitrate ( $\text{NO}_2^- + \text{NO}_3^-$ ), and dissolved inorganic phosphorus (DIP or  $\text{PO}_4^-$ ) were measured using the automated method of EPA (1979). Methods of Strickland and Parsons (1972) and Parsons *et al.* (1984) are followed for chlorophyll-*a* analysis. Total suspended solids (TSS) and total volatile solids (TVS) were measured with a gravimetric method.

## **3.2. Epiphyte Growth Survey**

### **3.2.1. Epiphyte Station locations and Sampling Frequency**

Epiphyte collection arrays were placed at a single location, in water approximately 1 meter deep average water depth, along three fixed transects (KC1, BP3, PR5) at Blossom Point, Maryland (Figure 3-1, Table 3-1). Three weekly epiphyte fouling rate measurements were collected in late summer 2000, and three rate measurements were collected in fall 2000. Data collected for three consecutive weeks in the summer of 1999, fall of 1999 and spring of 2000 were collected under a separate contract and are not included in this report.

### **3.2.2. Epiphyte Growth Measurement Method**

In order to assess the light attenuation potential of epiphytic growth on the leaves of submerged aquatic vegetation (SAV) artificial substrata, thin strips of Mylar<sup>®</sup> polyester plastic, were deployed at a single location along each transect for a period of 7 to 10 days. The use of transparent Mylar<sup>®</sup> plastic provided a means to estimate light attenuation due to epiphytic growth and sediment accumulation, as well as to quantify the organic and inorganic components of the fouling.

### **3.2.3. Description of Epiphyte Collector Arrays**

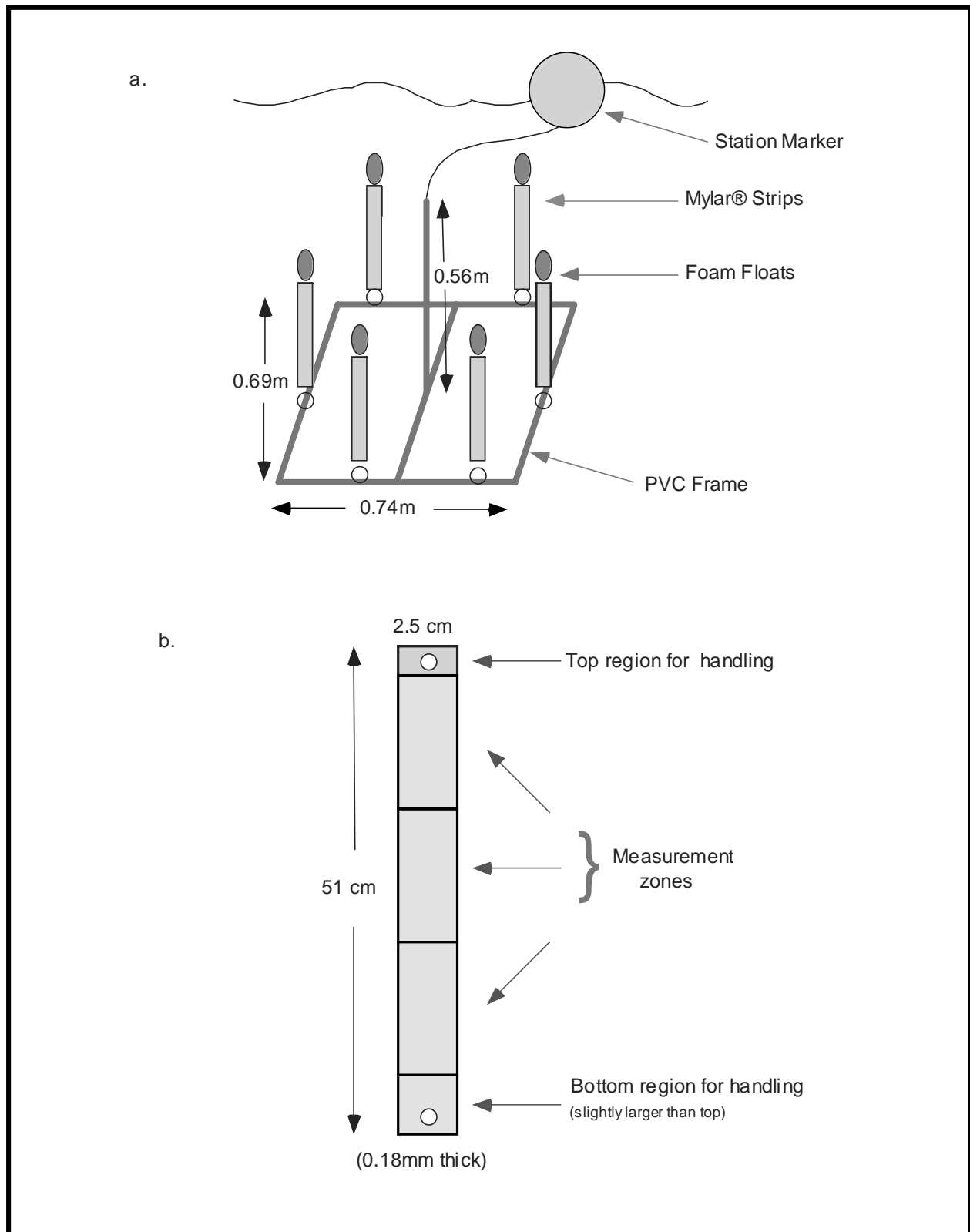
Each collector array (Figure 3-2) consists of a square PVC frame with a vertical PVC shaft in the center of the square. To this shaft is attached a line with a small surface float that allows for easy location of the collector. Each collector array holds up to six strips per deployment. Mylar<sup>®</sup> strips (2.5 cm wide x 51 cm long and 0.7 mil thick) are attached to the frame so that the top is allowed to move freely in the water column. Small foam floats (~3.5 x 3.3 cm) are attached to the top of the strip to help maintain a vertical position in the water column at all times.

### **3.2.4. Sampling the Epiphyte Collector Arrays**

On each sampling date, six replicate Mylar<sup>®</sup> strips were collected. Three strips were analyzed for chlorophyll-*a*, and three for total dry mass/inorganic dry mass. While suspended in the water, Mylar<sup>®</sup> strips were gently removed from the array and cut with scissors to remove the middle 1/3 marked section (64.5 cm<sup>2</sup>, Figure 3-2). This section was once again cut in half and placed in a 60 ml plastic centrifuge tube for transport back to the laboratory. The tube was then placed in a cooler for transport back to the laboratory. The samples were immediately frozen upon arrival at the laboratory prior to further processing.

### **3.2.5. Processing Organic/Inorganic Epiphyte Material**

The Mylar<sup>®</sup> strip sections collected for dry mass/inorganic mass analysis were scraped of all material and rinsed with distilled water. Scraped material and rinse water were diluted to a fixed volume (300 - 500 ml). The solution was mixed as thoroughly as possible on a stir plate until homogenized. A small aliquot (10 to 50 ml) was then extracted with a glass pipette and filtered through a 47 mm, 0.7 μm (GF/F) glass fiber filter. Once filtered, the pads were immediately frozen and delivered to NASL for analysis.



**Figure 3-2. Diagram of SAV Epiphyte Collector Array.**

**a. Epiphyte Collector Array**

**b. Mylar® strips**

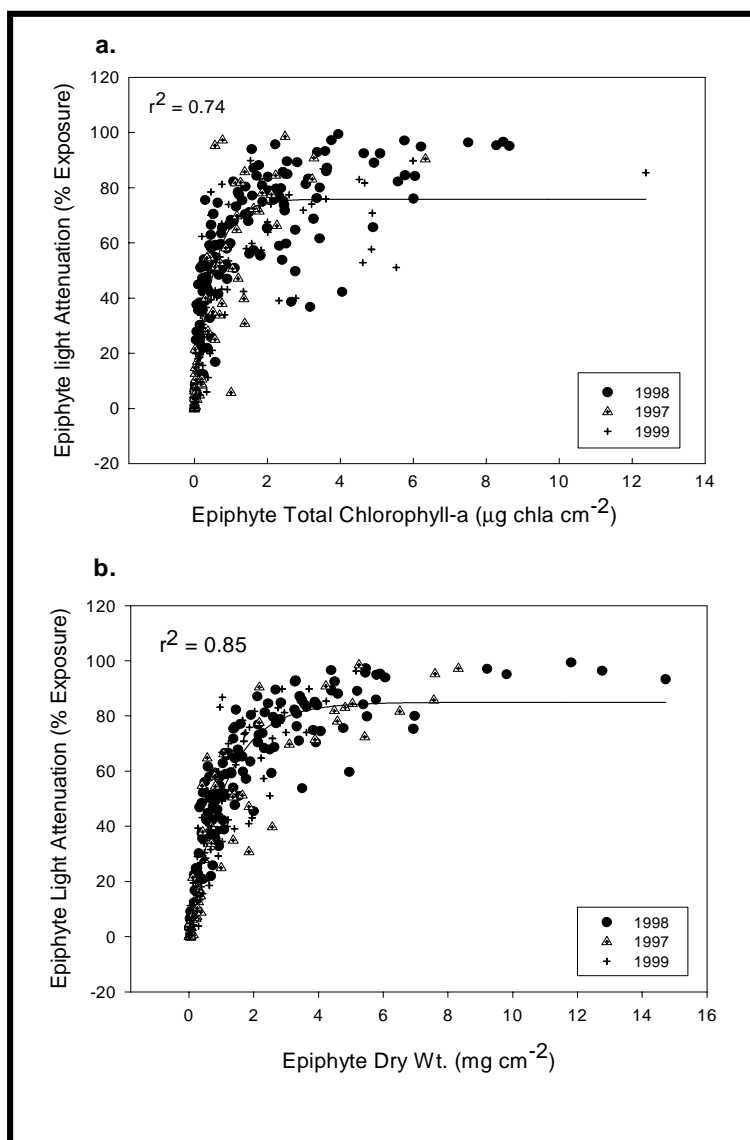


### 3.3. Estimating light Attenuation

There are several trade-offs associated with using artificial substrates for an assessment of epiphyte fouling rates compared to using live SAV. Artificial substrates may not mimic the exact morphology of SAV blades and may not be exactly comparable to the specific species being considered, however they can provide a first order estimate of fouling rates that is much less costly than manipulating live SAV blades. In addition, standardization of these estimates allows more rigorous comparison among other locations and studies.

Estimates of light attenuation were made using measurements of epiphyte dry mass and calculating light attenuation from existing relationships between epiphyte dry mass and light attenuation (Figure 3-3 a, b).

These relationships can be used to calculate the percentage of surface light reaching the depth of SAV blade through the water column (PLW) and the percentage of surface light reaching the blade of SAV through the epiphyte layer at the leaf surface (PLL). These parameters are explained in Table 3-2.



**Figure 3-3. a. Epiphyte light attenuation vs. epiphyte chlorophyll-a, where light attenuation =  $77.36 \cdot (1 - e^{-2.082 \cdot \text{Epi Chla}})$  and b. epiphyte light attenuation vs. epiphyte dry mass where light attenuation =  $84.634 \cdot (1 - e^{-0.963 \cdot \text{Epi drywt}})$ .**

**Table 3-2. Calculation of percentage of surface light reaching the leaf surface (PLL).**

Calculation of % Surface Light Reaching Leaf Surface (PLL)	
$PLW = (I_z/I_0) \cdot 100 = [e^{-kd \cdot Z}]$	Where: $I_z$ = Light flux (PAR) at depth
$PLL = [e^{-kd \cdot Z}] [1 - LA/100]$	$I_0$ = Light flux (PAR) at surface
	$LA$ = Epiphyte light attenuation
	$Z$ = Observation depth (m)

### **3.4. Data management procedures**

All field data were recorded on specially prepared field data sheets. The initials of the person recording the data were entered on each data sheet. The raw data sheets were reviewed for possible missing data values due to sample collection problems prior to data entry. These sheets were filed in the laboratory.

#### **3.4.1. Incorporation of Error Codes in Data Tables**

In order to keep a record of problems experienced during data collection, an alphanumeric code is entered in the data table describing the problem associated with each questionable parameter value (Table 3-3).

#### **3.4.2. Data Tables QA/QC Control**

After data have been entered into spreadsheet files, hard copies of the files were manually checked for errors against original data sheets. Any errors were corrected, and a second printout produced which is re-verified by a different staff member.

#### **3.4.3. Blossom Point SAV Habitat Evaluation Data Sets**

Data file names are a unique alphanumeric code reflecting the type of data and year (yyyy) data were collected.

**WATER QUALITY MEASUREMENTS** Filename: **BPWCNDyyyy**, (Appendix A-1) contains temperature, salinity and dissolved oxygen data measured at 0.5 meters below the water surface.

**WATER COLUMN LIGHT ATTENUATION MEASUREMENTS** Filename: **BPWCLTyyyy**, (Appendix A-2) contains photosynthetically active radiation (PAR) measurements at a minimum of two depths and the subsequent calculated Kd values for each station.

**WATER COLUMN NUTRIENT MEASUREMENTS** Filename: **BPWCNTyyyy**, (Appendix A-3) contains water column dissolved nutrient concentrations, chlorophyll-*a* (active and total) concentrations, and suspended solids concentrations (total and inorganic) in the surface waters at each station.

**EPIPHYTE CHLOROPHYLL-*a* ACCUMULATION MEASUREMENTS:** Filename: **BPECHLyyyy**, (Appendix A-4) contains epiphyte chlorophyll-*a* concentrations (total and active).

**EPIPHYTE DRY MASS ACCUMULATION MEASUREMENTS:** Filename: **BPEDRMyyyy**, (Appendix A-5) contains total epiphyte dry weight and percent inorganic fractions.

**Table 3-3. Analysis Problem Codes**

<b>ANALYSIS PROBLEM CODE</b>	<b>DESCRIPTION</b>
D	Insufficient sample
N	Sample Lost
P	Lost results
R	Sample contaminated
S	Sample container broken during analysis
V	Sample results rejected due to QA/QC criteria
X	Sample not preserved properly
AA	Sample thawed when received
BB	Torn filter paper
DA	Damaged epiphyte array
DS	Damaged epiphyte strip
HH	Sample not taken
JJ	Amount filtered not recorded (Calculation could not be done)
LA	Lost epiphyte array
LL	Mislabeled
NI	Data non-interpretable
NR	No replicate analyzed for epiphyte strip chlorophyll- <i>a</i> concentration
SS	Sample contaminated in field
SW	Shallow water, light flux measured at two points only
TT	Instrument failure
UU	Analysis discontinued
XX	Sampling for this variable was not included in the monitoring program at this time or was not monitored during a specific cruise

## 4. RESULTS

### 4.1. Dissolved Nutrient Concentrations

As expected, dissolved inorganic nitrogen (DIN) and dissolved inorganic phosphorus (DIP) concentrations showed strong seasonal changes ( $p < 0.01$ , Figures 4-1, 4-2, 4-3). There was a significant difference among stations ( $p < 0.05$ ), in dissolved inorganic nitrogen (DIN) concentrations. In addition, DIN concentrations were significantly higher at offshore locations compared to inshore locations ( $p < 0.05$ ). No significant differences in dissolved inorganic phosphorus (DIP) concentrations were found among stations. However, DIP concentrations were significantly higher at off-shore locations compared to in-shore ( $p < 0.01$ ). Dissolved inorganic nitrogen concentrations ranged from a low of  $4.34 \mu \text{Mol N l}^{-1}$  at station KC1-100 (in-shore location) to a maximum of  $101.4 \mu \text{Mol N l}^{-1}$  at the off-shore location of PR5-200 (Table 4-1). Dissolved inorganic phosphorus concentrations ranged from a low of  $0.200 \mu \text{Mol P l}^{-1}$  at station BP3-300 (off-shore location) to a maximum of  $2.040 \mu \text{Mol P l}^{-1}$  at the off-shore location of KC1-300 (Table 4-1).

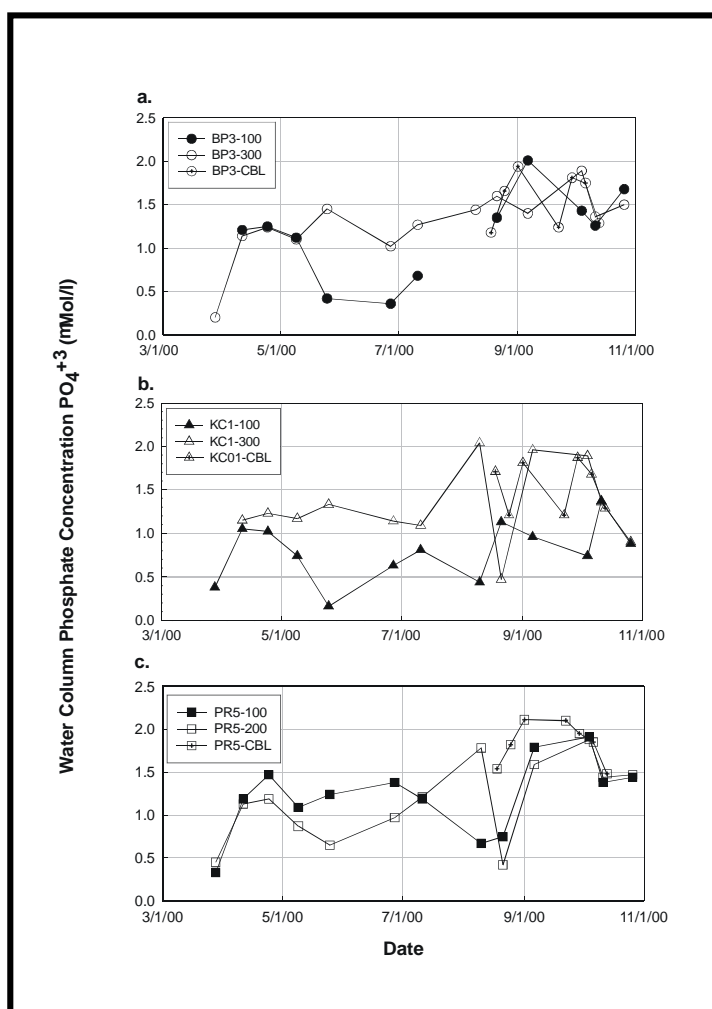


Figure 4-1. Dissolved inorganic phosphorus (DIP) concentrations at Blossom Point in 2000.

Table 4.1 Maximum, minimum and median values of water column nutrient concentrations recorded at Blossom Point in 2000.

		PR5		BP3		KC1	
		50 m	200 m	100 m	300 m	100 m	300 m
Dissolved Nitrate Plus Nitrite ( $\mu\text{mol N}$ )	max	85.8	90.6	87.0	87.7	79.9	83.9
	min	5.35	9.59	18.1	17.2	4.04	5.32
	median	35.1	45.5	28.6	31.2	22.0	40.95
Dissolved Ammonium ( $\mu\text{mol N}$ )	max	13.7	14.0	13.8	13.7	14.4	12.5
	min	0.4	0.5	0.1	0.4	0.3	1.5
	median	3.1	5.4	2.4	4.6	1.3	3.25
Dissolved Inorganic Phosphorus ( $\mu\text{mol P}$ )	max	1.88	1.91	2.01	1.89	1.37	2.04
	min	0.42	0.33	0.36	0.2	0.16	0.47
	median	1.19	1.24	1.25	1.36	0.81	1.2

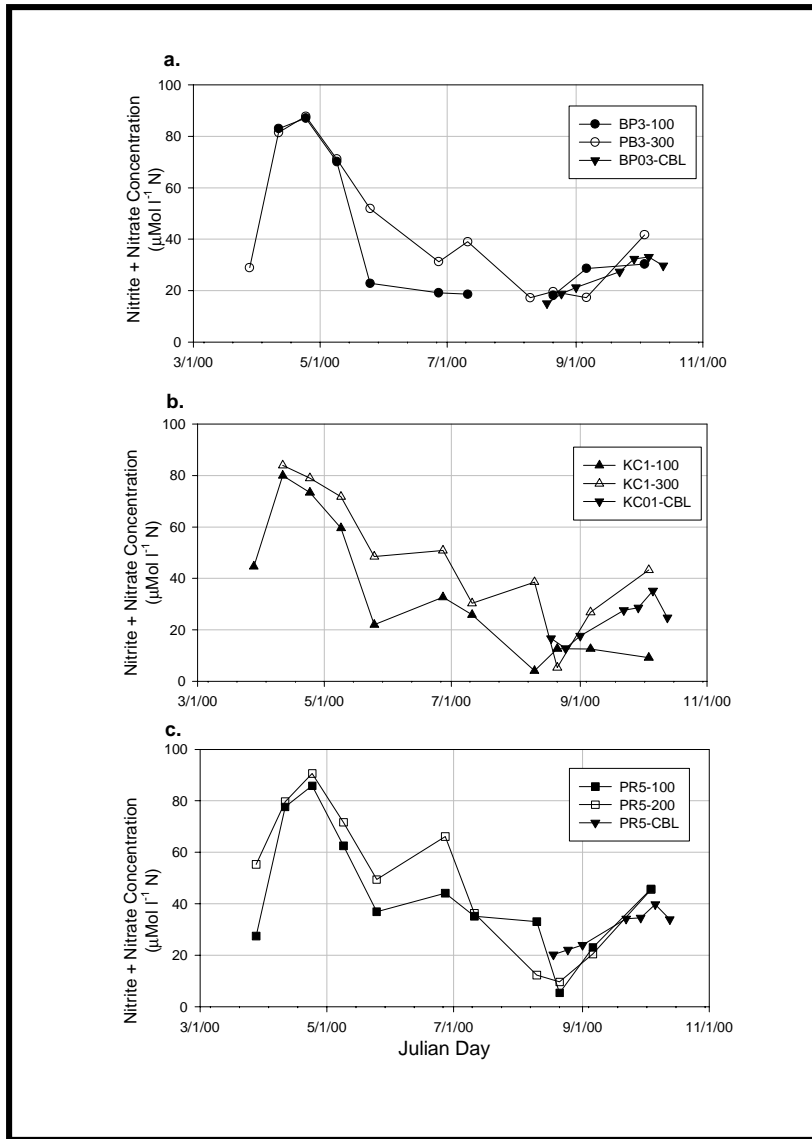


Figure 4-2. Dissolved nitrite plus nitrate concentrations at stations a) BP3, b) KC1, and c) PR5 at Blossom Point in 2000.

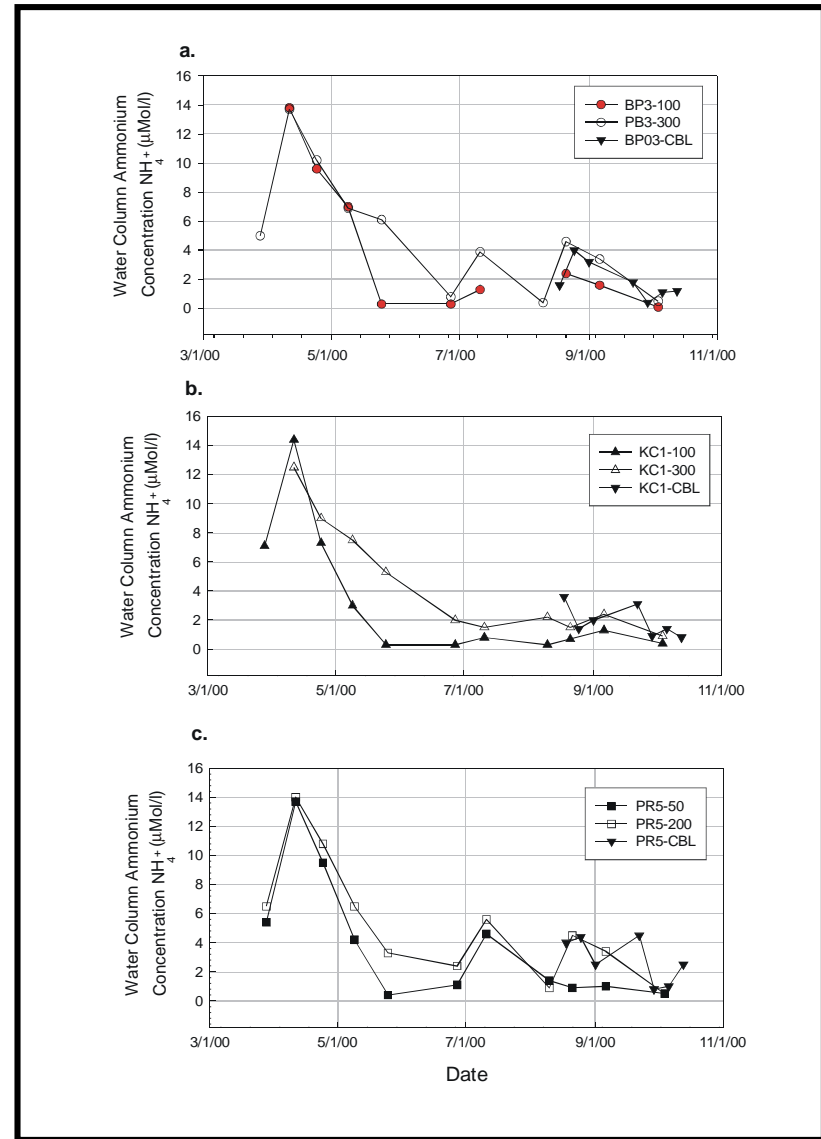


Figure 4-3. Dissolved ammonium concentrations at stations a) BP3, b) KC1, and c) PR5 at Blossom Point in 2000.

## 4.2. Epiphyte Fouling Rates

Epiphyte accumulation rates (epiphyte chlorophyll-*a* and dry mass) in 2000 varied significantly with season and tracked well with water temperature (Figure 4.4). As expected, the highest fouling rates were found during the summer months. Similar results were observed between summer and fall fouling rates in 1999 as well. Summer season epiphyte chlorophyll-*a* fouling rates were higher in 2000 compared to 1999, but were not statistically different from each other (Figure. 4.5, Table 4-2). However, significant differences were found in epiphyte chlorophyll-*a* accumulation rates among stations in both 1999 and 2000 (Kruskall-Wallis Rank test,  $p < 0.001$ , Figure 4-5, Table 4-2). In contrast, summer dry mass accumulation rates were significantly higher in 1999 compared to 2000 (Kruskall-Wallis Rank test  $p < 0.001$ , Table 4-2). The difference in dry mass between years was due to differences in the inorganic component of the epiphytic fouling with significantly higher inorganic accumulation rates in 1999 ( $1.109 \text{ mg cm}^{-2}$ ) compared to 2000 ( $0.112 \text{ mg cm}^{-2}$ ,  $p < 0.001$ ).

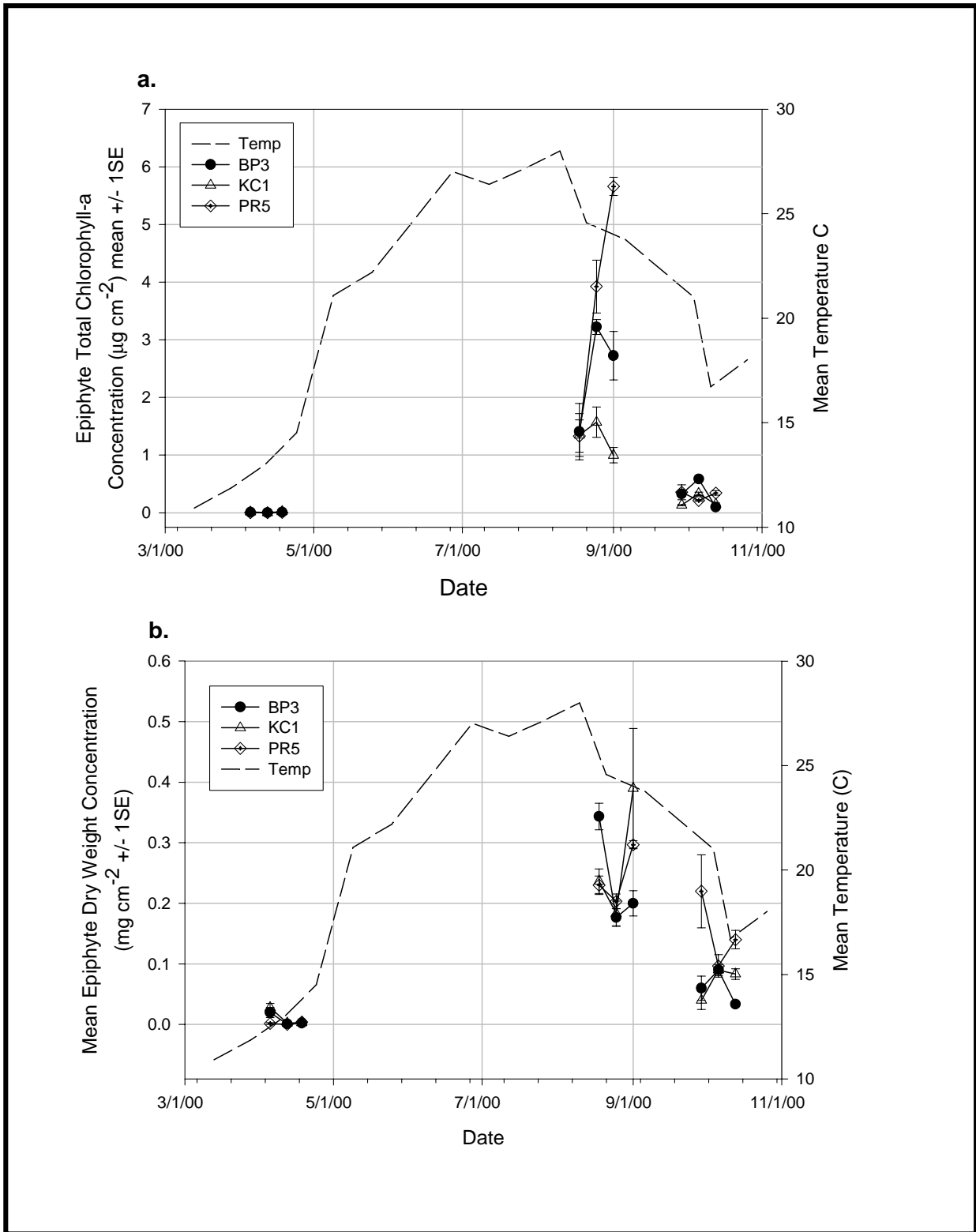
**Table 4-2. Mean summer epiphyte concentrations after approximately one week *in situ* exposure.**

Year	Mean Epiphyte Chlorophyll- <i>a</i> ( $\mu\text{g cm}^{-2}$ )			Mean Epiphyte Dry Mass ( $\text{mg cm}^{-2}$ )		
	BP3	KC1	PR5	BP3	KC1	PR5
1999	1.437	0.822	2.687	1.500	1.002	1.680
2000	2.451	1.305	3.639	0.240	0.271	0.243

## 4.3. Epiphyte Light Attenuation

Epiphyte fouling rates were extremely low during the spring sampling period and did not contribute significantly to light attenuation at any station. While fouling rates were highest during the summer they only contributed marginally to light attenuation (Figure 4-6). After one week of exposure, the maximum observed epiphyte accumulation during a single deployment reduced the light at the leaf surface to 22.9 % of surface irradiance down from 31.4 % without epiphyte accumulation. Overall, mean attenuation from epiphytes reduced light availability from 18.5% surface irradiance to 14.93% at the leaf surface.

A comparison of the relative contribution to light attenuation from epiphytic fouling (PLL vs. PLW) measured at Blossom Point during the summer season to locations in the mesohaline portion of the Patuxent River and Tangier Sound show that epiphytes contribute a smaller proportion of light attenuation at Blossom Point compared to most sites at these other locations. The only other comparable sites were located in the more turbid portions of the Patuxent River (Figure 4-7).



**Figure 4-4. a) epiphyte total chlorophyll-a accumulation rates, and b) epiphyte dry mass accumulation rates measured from Mylar® strips after approximately 1 week of *in situ* exposure in 2000 at Blossom Point .**

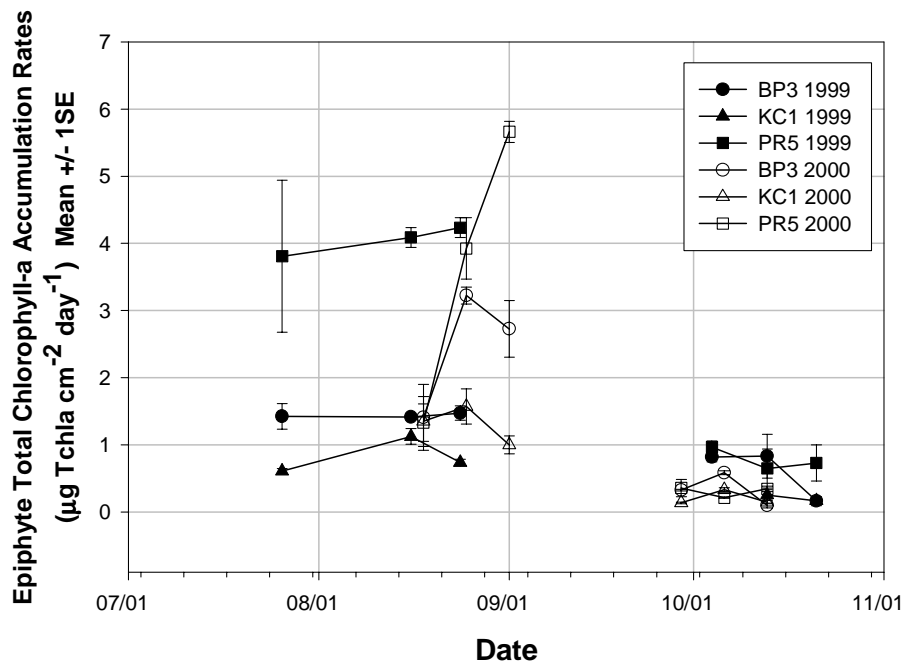
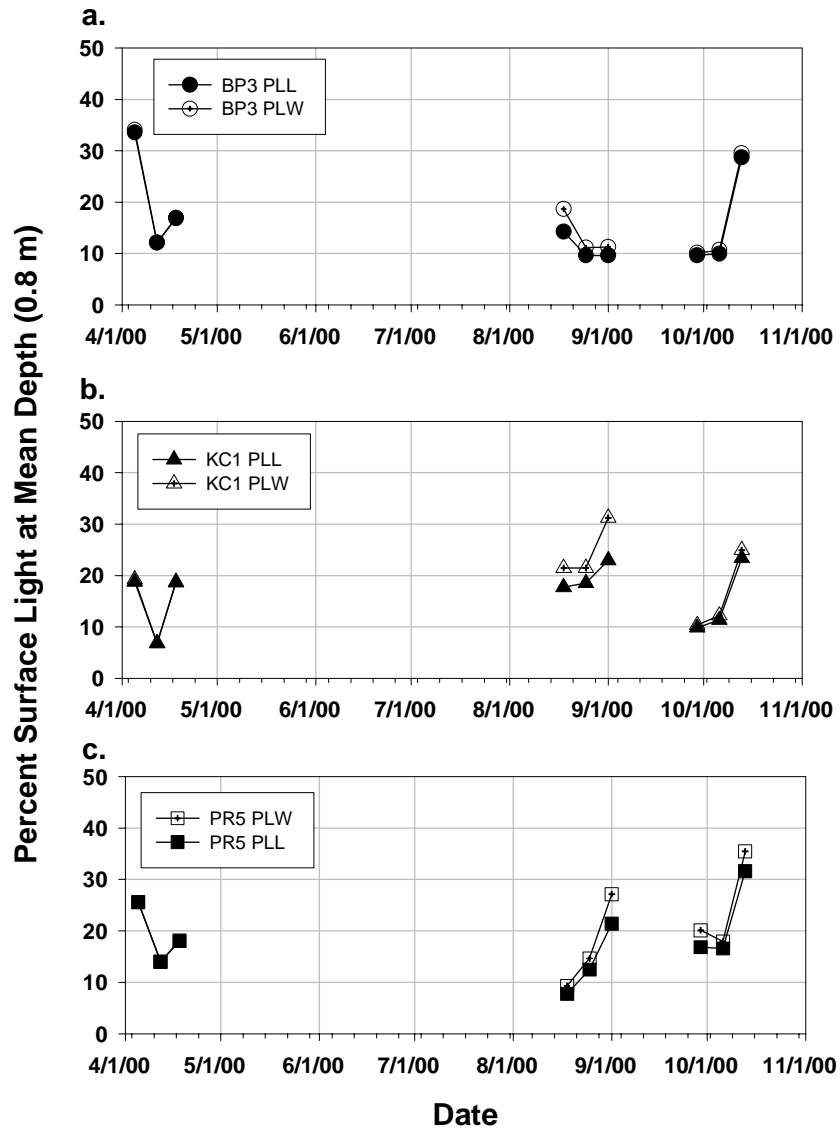


Figure 4-5. Epiphyte chlorophyll-a accumulation rates summer 1999 and 2000 measured from Mylar® strips after approximately 1 week of *in situ* exposure at Blossom Point.





**Figure 4-6. Estimated percentage of light through the water column (PLW), and percentage of light at the leaf surface (PLL) at Blossom Point in 2000.**

*Fouling rates were estimated using Mylar® strips deployed for one week of in-situ exposure.*

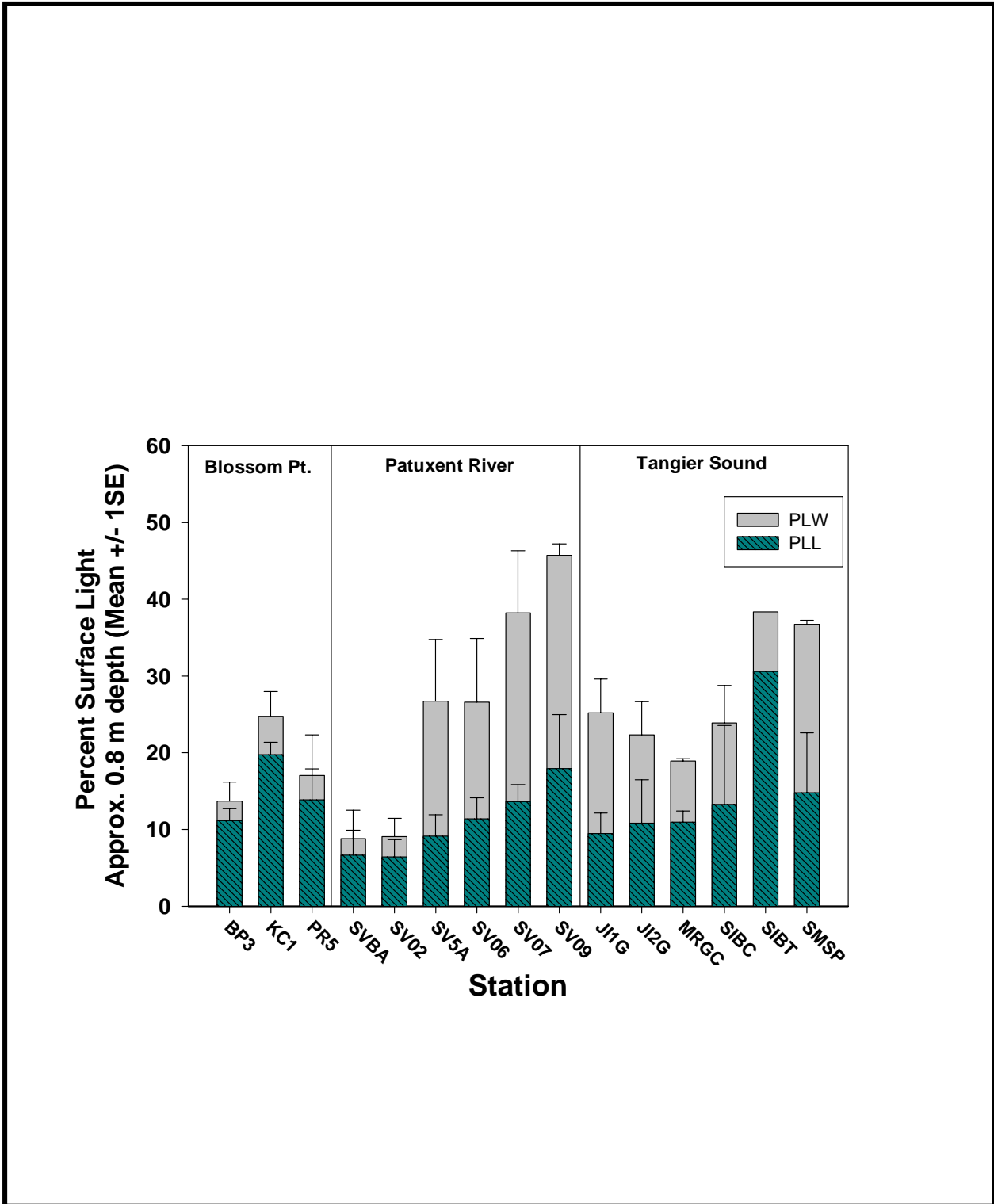


Figure 4-7. Estimated seasonal mean percent light through the water column (PLW), and percent light at the leaf surface (PLL) at Blossom Point in the summer of 2000 compared to other locations in the mesohaline region of the Patuxent River and Tangier Sound.

*Fouling rates were estimated using Mylar® strips deployed for one week of in-situ exposure.*

## 5. DISCUSSION

A number of parameters interact to determine the impact of epiphytic accumulation on SAV. These include the light and nutrients available, water temperature and the growth rate of the SAV. Fouling rates may also be affected by short-term weather events and as a result, fouling rates at a single location and within a single season are typically quite variable. This conclusion is reinforced by the large difference in epiphyte fouling rates and inorganic composition found between 1999 and 2000. While the estimates of biomass accumulation using rectangular strips of Mylar® may not be exactly equivalent to fouling rates on species of SAV with drastically different morphologies, these measurements are useful nevertheless. Artificial substrates provide a means to standardize accumulation rates among locations and seasons without regard to plant growth rates or morphologies. Summer fouling rates at Blossom Pt. were somewhat lower than fouling rates at other locations measured during similar time periods. For example, at Blossom Pt., mean dry mass accumulation after a week of fouling was only  $0.25 \text{ mg cm}^{-2}$ , while the lowest fouling rates found in the Patuxent River (upper mesohaline) during a similar time-period were  $0.49 \text{ mg cm}^{-2}$ . However, since light availability through the water column at the Patuxent River stations (8.9% surface light) was less than at Blossom Pt (18.5%), the overall epiphyte contribution to light attenuation was lower in the upper Patuxent. Despite relatively turbid water at all the locations sampled at Blossom Point, the SAV appears to be healthy and thriving. While the extent and density of these beds shift substantially on an annual basis, several SAV species have persisted there for considerable time in spite of epiphytic fouling. An additional year of fouling data to be collected in 2001 will provide even stronger baseline information by which to judge the impact of the upcoming shoreline revetment and its potential impact on the fouling rates of SAV at Blossom Point.

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