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DECOMPOSITION, NUTRIENT UPTAKE AND MICROBIAL COLONIZATION OF SAWGRASS AND CATTAIL LEAVES IN WATER CONSERVATION AREA 2A

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BY

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WEST PALM BEACH, FLORIDA

EXECUTIVE SUMMARY

Phosphorus and nitrogen-enriched water entering Water Conservation Area 2A (WCA-2A) through the S-10 structures has created a zone of nutrient enrichment and assimilation in the vicinity of these structures (Gleason et al, 1974; Davis and Harris, 1978). Davis and Harris showed that the decomposing leaf litter of sawgrass and cattail was at least as important as the growing plants in nutrient uptake in this zone. The mechanism for this uptake by leaf litter was not determined, although it was hypothesized that microbial colonization of the decomposing leaves may have been responsible. This study was undertaken to explore the roles of microbial populations in nutrient uptake during leaf decomposition.

Dead leaf material was placed in litterbags in the marsh at sites subjected to enriched and background P and N concentrations in WCA-2A water. Bags were retrieved at approximately biweekly intervals for three months and at monthly intervals for the remainder of the six month study. Populations of aerobic and facultative bacteria and fungi inhabiting the leaf litter were assessed by plate counts, and algal biomass was estimated from chlorophyll a concentrations. Changes in the weight and P and N content of the decomposing leaf litter were monitored in addition to water quality and water depths at the sites. Fiber analysis of dead leaf material was conducted to determine if decomposition rates were correlated with lignin, cellulose and residual ash contents in freshly dead leaves.

Larger weight loss and nutrient uptake by the leaf litter occurred in the zone of nutrient enrichment as compared with the non-enriched location. These results paralleled the findings of Davis and Harris (1978). However, populations of aerobic and facultative bacteria and fungi inhabiting sawgrass litter were found to be significantly lower at the nutrient enriched sites

compared to the non-enriched site. This did not appear to be the case for cattail litter, in which microbial populations were not found to differ significantly between the enriched and non-enriched sites. In contrast to the litter microflora, planktonic bacteria and fungi were found in higher densities at the nutrient enriched sites compared to the non-enriched location in a sample of surface water.

Larger weight loss and nutrient uptake by cattail litter compared to that of sawgrass in the zone of nutrient enrichment also paralleled the findings of Davis and Harris (1978). This larger uptake by cattail litter was accompanied by significantly larger bacterial populations and by earlier fungal colonization in comparison to sawgrass litter.

Litter decomposition rates tend to be inversely related to initial lignin content. However, fiber analyses for lignin, cellulose and residual ash in freshly dead leaves did not yield significant differences between enriched and non-enriched sites. Thus, lignin concentrations failed to explain more rapid leaf decomposition in the enrichment zone. Higher residual ash concentrations in sawgrass in comparison to cattail may have contributed by slower decomposition by sawgrass. Residual ash in the fiber analysis scheme is not readily broken down, and the higher concentrations found in sawgrass probably indicate a higher silica content.

Dissolved oxygen concentrations in the water just above the litter layer were < 0.2 mg/l on the majority of sample dates at the nutrient-enriched sites compared to values averaging approximately 2.0 mg/l at the non-enriched site. The low D.O. readings at the enriched sites indicate that anaerobic conditions existed in the litter most of the time. Anaerobic conditions provide a likely explanation for the retarded populations of aerobic and facultative bacteria and fungi in sawgrass litter at the nutrient-enriched sites. In contrast, low D.O. at the enriched sites apparently less adversely

affected the facultative microbial populations which inhabited cattail litter. Since facultative litter bacteria and fungi were not more abundant in the enrichment zone, they apparently were not responsible for the reduced D.O. concentrations which were found there. Respiration by planktonic bacteria and fungi, which were more abundant in the enrichment zone, probably contributed to the lower D.O. concentrations there.

It is concluded that the colonization of sawgrass leaf litter by aerobic and facultative bacteria and fungi was inhibited in the enrichment zone of WCA-2A, probably as a result of low D.O. levels. Populations of these organisms on cattail litter were neither inhibited nor stimulated in the enrichment zone. Thus the increased nutrient uptake by leaf litter in the enrichment zone of WCA-2A cannot be attributed to increased colonization by aerobic and facultative microbes in either sawgrass or cattail litter.

The increased rates of leaf decomposition in the enrichment zone, the lack of corresponding increases in aerobic and facultative microbial populations, and the anaerobic conditions which prevailed there suggest an alternative explanation that anaerobic bacteria may have been responsible for most of the leaf decomposition in this area. Although not enumerated in this study, anaerobes have been shown to play a major role in the metabolism of salt-marsh communities (Wiebe et al., 1981). The possibility of a shift from an aerobic and facultative metabolism to an anaerobic one in WCA-2A as a result of nutrient enrichment deserves further investigation and serious consideration in the management of the area.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

ACKNOWLEDGEMENTS

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INTRODUCTION

The Water Conservation Area system encompasses about 3500 km^2 of diked Everglades marshland in southern Florida (Figure 1). These areas are managed by the South Florida Water Management District for a variety of purposes including flood control, water conservation storage, groundwater recharge, preservation of fish and wildlife resources, and recreational benefits. Much of the water entering the Water Conservation Areas is runoff from agricultural lands; this water contains high phosphorus and nitrogen concentrations compared to the water of the interior marshes. The 547 km^2 marsh of Water Conservation Area 2A (WCA-2A) receives a particularly large supply of these nutrients through the S-10 inflow structures because of the large canal system which converges on these inflows (Figure 2). The absence of interior canals in WCA-2A forces this water to flow across the marsh. The potential backpumping of additional canal water into the Water Conservation Areas would further increase supplies of P and N to WCA-2A. Most of the P and N presently entering WCA-2A is assimilated in an enrichment zone in the vicinity of the S-10 structures (Gleason, et al, 1974; Davis and Harris, 1978). However, the ability of the marsh to store the additional nutrient supplies which would result from back-pumping is not known. This study is one of a series of investigations in WCA-2A to determine the effects of increased phosphorus and nitrogen supplies on sawgrass and cattail communities and their effectiveness as nutrient sinks.

Davis and Harris (1978) showed that sawgrass and cattail leaf litter during the first year of decomposition was at least as important as the growing plants in phosphorus uptake in the enrichment zone of WCA-2A. More rapid weight losses and larger P accumulations in leaf litter were found in the enrichment zone compared to non-enriched areas, and in cattail compared to sawgrass litter. Davis (1982) reported similar results for shorter-term

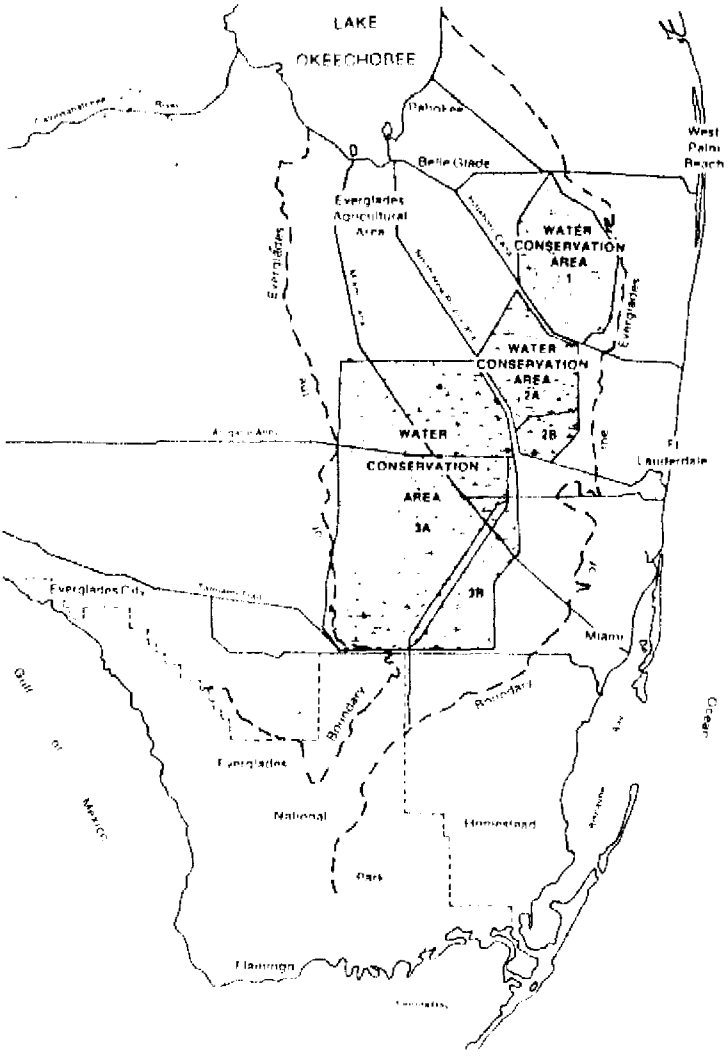

FIGURE 1 LOCATION OF EVERGLADES WATER CONSERVATION AREAS AND BOUNDARY OF ORIGINAL EVERGLADES IN SOUTH FLORIDA.

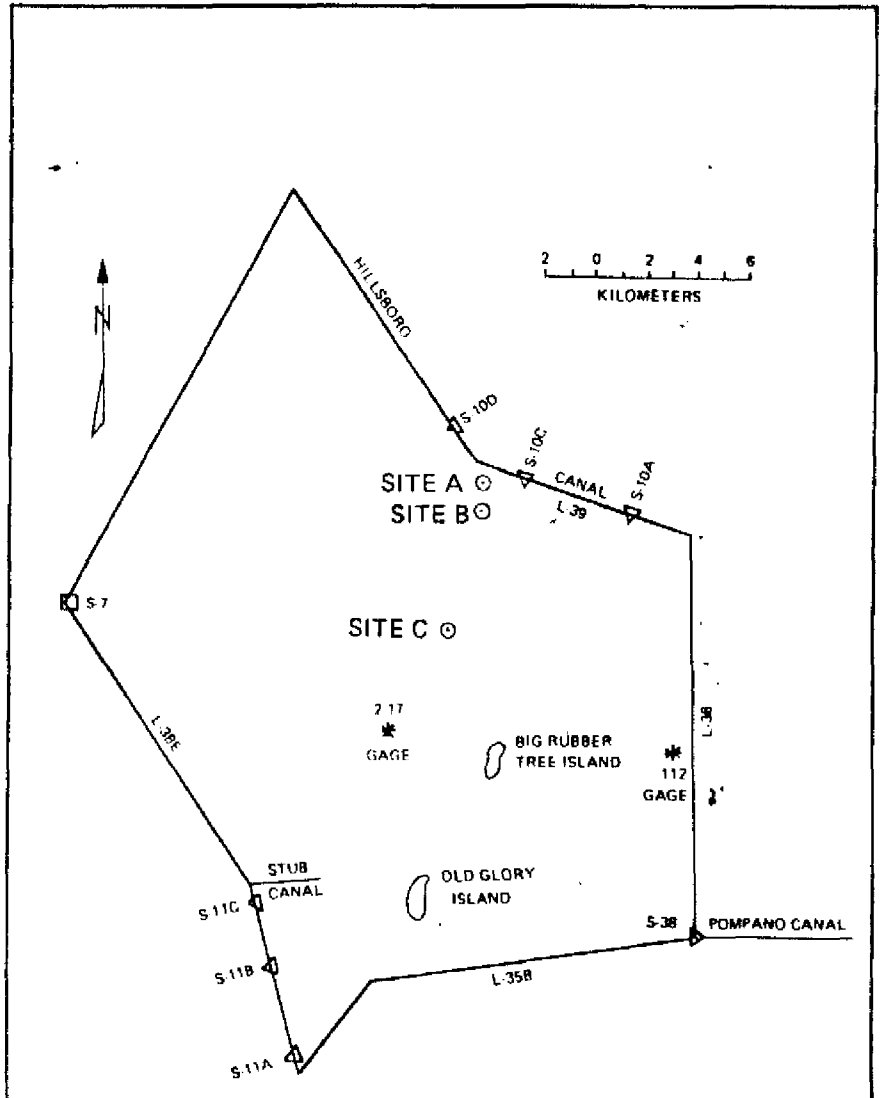

FIGURE 2 LOCATIONS OF MICROBIOLOGICAL SAMPLE SITES IN WATER CONSERVATION AREA 2A

uptake during 10-day radiotracer experiments using ^{32}P; radiophosphorus concentrations in litter were higher at the enriched site compared to the non-enriched one. The mechanism for this uptake by leaf litter was not determined, although it was hypothesized that microbial colonization of the decomposing leaves may have been responsible. In this study, populations of bacteria, fungi, and algae on leaf litter were monitored during 6 months of decomposition to determine if increased weight loss and nutrient accumulation were accompanied by higher microbial densities.

METHODS

Three sampling locations (Figure 2) were selected based on proximity to the S-10 structures and on previously determined concentrations of phosphorus in WCA-2A surface water and dead leaf material (Gleason et al., 1974; Davis and Harris, 1978). Sawgrass (Cladium jamaicense) and cattail (primarily Typha domingensis with some T. latifolia) were the dominant emergent vegetation. Adjacent stands of these species occurred at each site. Site A was located 0.8 km south of L-39 between S-10C and S-10D and was characterized by high levels of nutrient enrichment. An area receiving moderately high enrichment, Site B, was located 1.6 km south of L-39. Site C, 6.4 km south of L-39, was outside the zone of nutrient enrichment.

Litterbag Preparation and Collection

Dead leaves were collected from living sawgrass and cattail plants at each of the three sites in May 1980, cut into 5-10 cm lengths, and oven-dried to constant weight at 45°C for 96 hours. This temperature was selected to insure the viability of spores and resting structures of fungal species that are known to occur in terrestrial as well as aquatic environments (Kaushik and Hynes, 1968, 1971; Barlocher and Kendrick, 1974).

Litterbags were constructed from 30x30 cm squares (6 meshes/cm) of fiber-glass window screening which were loosely folded to contain leaf material, while allowing entry of macroinvertebrates into the bags (Davis and Harris, 1978). Each litterbag contained five grams dry weight of sawgrass or cattail leaf material. Sawgrass litterbags were placed at the three sites on June 12, 1980, cattail on June 19, each bag containing litter from its respective site.

The litterbags were retrieved biweekly through August and at approximately monthly intervals thereafter. Sawgrass collection dates were June 26, July 10,

July 24, August 7, August 21, September 9, October 2, October 30 and November 25. Cattail collection dates were July 3, July 17, July 31, August 14, August 28, September 25, October 23 and December 4. A sample of surface water for counts of planktonic bacteria and fungi was collected from each of the three sawgrass stands in sterile, 100 ml screw cap glass bottles on November 20.

Prior to each litterbag collection, water temperature, pH and dissolved oxygen (D.O.) at the litterbag depth were recorded. The depth of the litterbags within the water column was measured, rather than the depth of the litter layer, because newly-placed litterbags remained buoyant for varying periods of time before sinking to the litter layer. Samples of surface water were collected from both sawgrass and cattail stands and transported, on ice, to the laboratory.

Water samples were analyzed by the South Florida Water Management District Water Chemistry Laboratory for total phosphorus and total nitrogen. Samples for P analyses were digested by autoclaving. Samples for N analyses were digested by the Kjeldahl procedure. Concentrations of P and N were determined using a Technicon AutoAnalyzer II.

On each collection date, one set of five litterbags was retrieved from each of the three sawgrass or cattail stands. One litterbag (for bacterial and fungal plate counts) from the set was transferred aseptically to a sterile plastic bag and tied securely to minimize contamination by airborne microorganisms. A second litterbag, to be used for algal biomass estimation, was placed in a plastic bag containing enough marsh water to cover the litterbag. The remaining three litterbags were used for dry weight determinations and nutrient analyses. All litterbags were placed in a light-tight container for transport to the laboratory.

Bacteria and Fungi Estimates

Upon return to the laboratory, litterbag samples were plated within 6-8 hours after collection. Each sample was aseptically transferred into a sterilized 500 ml flask containing five grams of 3-4 mm glass beads plus 150 ml deionized water. The flask was clamped in a Burrell wrist-action shaker (300 cycles/min., 14 cm oscillation amplitude) and agitated for one hour. Using the Standard Plate Count technique (APHA, 1975), serial dilutions to 10^{-6} were prepared using one milliliter subsamples from the 150 ml to ensure that countable plates (20-300 colonies) would be obtained. Six replicate pour plates were prepared for each dilution. Martin's rose bengal-streptomycin medium (Martin, 1950) plus chlortetracycline was used for culture of fungi. Bacteria were cultured on Jensen's medium (Jensen, 1930) plus cycloheximide.

All plates were incubated aerobically at 28°C. Bacterial plates were counted after 15 days. Fungi were counted at eight days, reincubated and checked again at 22 days for any slow-growing species which may have developed on the plates. Plates were examined under 10-20x power of a stereomicroscope. Numbers of bacteria and fungi per litter sample were computed by multiplying the mean colony count of 4-6 replicate plates from the same dilution by 150^n where n = the reciprocal of the dilution used. Multiplication by 150 corrected for the one milliliter subsample pipetted from the 150 ml suspension in the flask. The power n compensated for the serial dilutions of the one milliliter subsample. This count was then divided by the mean dry weight of three replicate litter samples to give numbers of organisms/gram dry weight litter.

Algal Biomass Estimates

Attached algae were removed from leaf litter by scraping. Scrapings were diluted to 200 ml with deionized water. Three replicate 25 ml aliquots were filtered onto Whatman GF/C filters (0.45 micron pore size) and analyzed for chlorophyll *a* concentrations using an acid addition method of correcting

for phaeophytins (APHA, 1975). Percent absorbance was read on a Perkin-Elmer 552 Spectrophotometer at 750 and 663 nm before and after acid addition. The mean chlorophyll a concentration was then calculated, multiplied by 67 for a gross approximation of organic weight (APHA, 1975), and subsequently converted to biomass/g dry weight leaf litter. The remaining 125 ml of sample was sonicated for 30 seconds using a standard microtip at 6.5 KHZ (Heat Systems-Ultrasonics, Inc., Model #W225R), and preserved with 7% formalin for future identification. The predominant species of attached algae were determined by counts and cell volume calculations.

Litter Weight, Nutrients and Fiber

Litterbags for weight determinations and nutrient analyses were oven-dried for 72 hours at 90°C plus 96 hours at 45°C, weighed, and ground in a Wiley mill. Analyses for P and N were made by the South Florida Water Management District Soil Chemistry Laboratory using a Technicon AutoAnalyzer II, after solubilization of P by lithium metaborate fusion and Kjeldahl digestion of N using a block digester. Six replicate samples of freshly dead sawgrass and cattail leaves from Sites B and C were analyzed for lignin, cellulose and residual ash concentration by the University of Florida Agriculture Research and Education Center at Belle Glade according to the procedures of Goering and Van Soest (1970).

Statistical Analysis

Analyses of variance and t-tests were conducted according to Sokal and Rohlf (1981). Single Classification Analysis of Variance followed by the Newman-Keuls test was used to determine whether significant differences existed between sawgrass sites and between cattail sites regarding concentrations of N and P in the water, uptake of N and P by litter, and biomass estimates of bacteria, fungi and algae. The Student's t-test for uncorrelated samples was used to determine whether there were significant differences between

sawgrass and cattail leaves at each of the three sites regarding the parameters mentioned above. Site and species differences in lignin, cellulose and residual ash were tested using nested Analysis of Variance. Prior to these analyses, all data were tested for homogeneity of variance using Hartley's F_{max} test (Winer, 1971). Data were log-transformed in all cases where this assumption was not met. All t and F values of $\alpha \leq .05$ were accepted as significant.

RESULTS

Physical and Chemical Variables

Significantly higher concentrations of phosphorus and nitrogen were present in the water at the enriched sites compared to the non-enriched site (Table 1). Dissolved oxygen concentrations in the water just above the litter layer were $\leq$0.2 mg/l on the majority of sample dates at the nutrient-enriched sites compared to values averaging approximately 2.0 mg/l at the non-enriched site (Table 1). Water temperature ranged from 17 to 32°C, pH from 6.5 to 7.5, and litterbag depth from zero to 48 cm on sample dates.

Litter Decomposition and Nutrient Uptake

Decomposition rates of leaf litter at the nutrient-enriched sites were significantly higher than at the non-enriched site (Figure 3). Weight reductions of five-gram samples of sawgrass litter after 6 months decomposition were 1.66 g at Site A, 1.50 g at Site B and 1.13 g at Site C. Cattail litter lost 2.55, 1.95, and 1.14 g at the three respective sites during this period. Under nutrient enriched conditions (Sites A and B), cattail decomposed significantly more rapidly than sawgrass. However, both cattail and sawgrass decomposed at about the same rate under low nutrient concentrations found at non-enriched Site C.

Nutrient uptake by the leaf litter was directly proportional to decomposition rates. Uptake of both phosphorus and nitrogen after six months of decomposition was significantly higher at the nutrient-enriched sites than at the non-enriched site (Figure 3). Phosphorus concentrations in sawgrass litter (as percent litter dry weight) increased from 0.034 to 0.073% at Site A and from 0.037 to 0.075% at Site B, while phosphorus in cattail litter increased from 0.038 to 0.229% and from 0.026 to 0.150% at these two sites, respectively. In contrast, P concentrations in leaf litter at Site C

TABLE 1. PHYSICAL AND CHEMICAL PARAMETERS AT MICROBIOLOGICAL SAMPLE SITES DURING JUNE-DECEMBER, 1980. VALUES REPRESENT MEANS AND RANGES.

	SAWGRASS			CATTAIL		
	Site A	Site B	Site C	Site A	Site B	Site C
Total Phosphorus, mg/l	.164 (.083-.409)	.144 (.030-.388)	.008 (.003-.023)	.324 (.067-.850)	.145 (.048-.305)	.008 (.002-.015)
Total Nitrogen, mg/l	5.41 (2.50-8.05)	5.83 (2.94-10.52)	4.11 (2.28-6.13)	6.30 (2.62-10.70)	5.94 (2.69-9.57)	4.14 (1.77-6.88)
Dissolved Oxygen, mg/l	≤0.2 (≤0.2-0.4)	0.5 (≤0.2-1.6)	2.1 (≤0.2-4.8)	0.2 (≤0.2-0.4)	0.4 (≤0.2-3.4)	1.7 (≤0.2-3.6)
pH	7.0 (6.5-7.5)	7.0 (6.5-7.5)	7.0 (6.5-7.5)	7.0 (6.5-7.5)	7.0 (6.5-7.5)	7.0 (6.5-7.5)
Water Temperature, C	25 (18-30)	26 (18-29)	25 (18-28)	25 (18-30)	26 (19-30)	25 (17-32)
Water Depth, cm	57 (42-94)	42 (26-72)	46 (28-71)	58 (42-94)	32 (16-62)	59 (35-86)
Litterbag Depth, cm	15 (0-44)	16 (0-46)	21 (10-39)	12 (0-45)	13 (0-37)	23 (0-48)

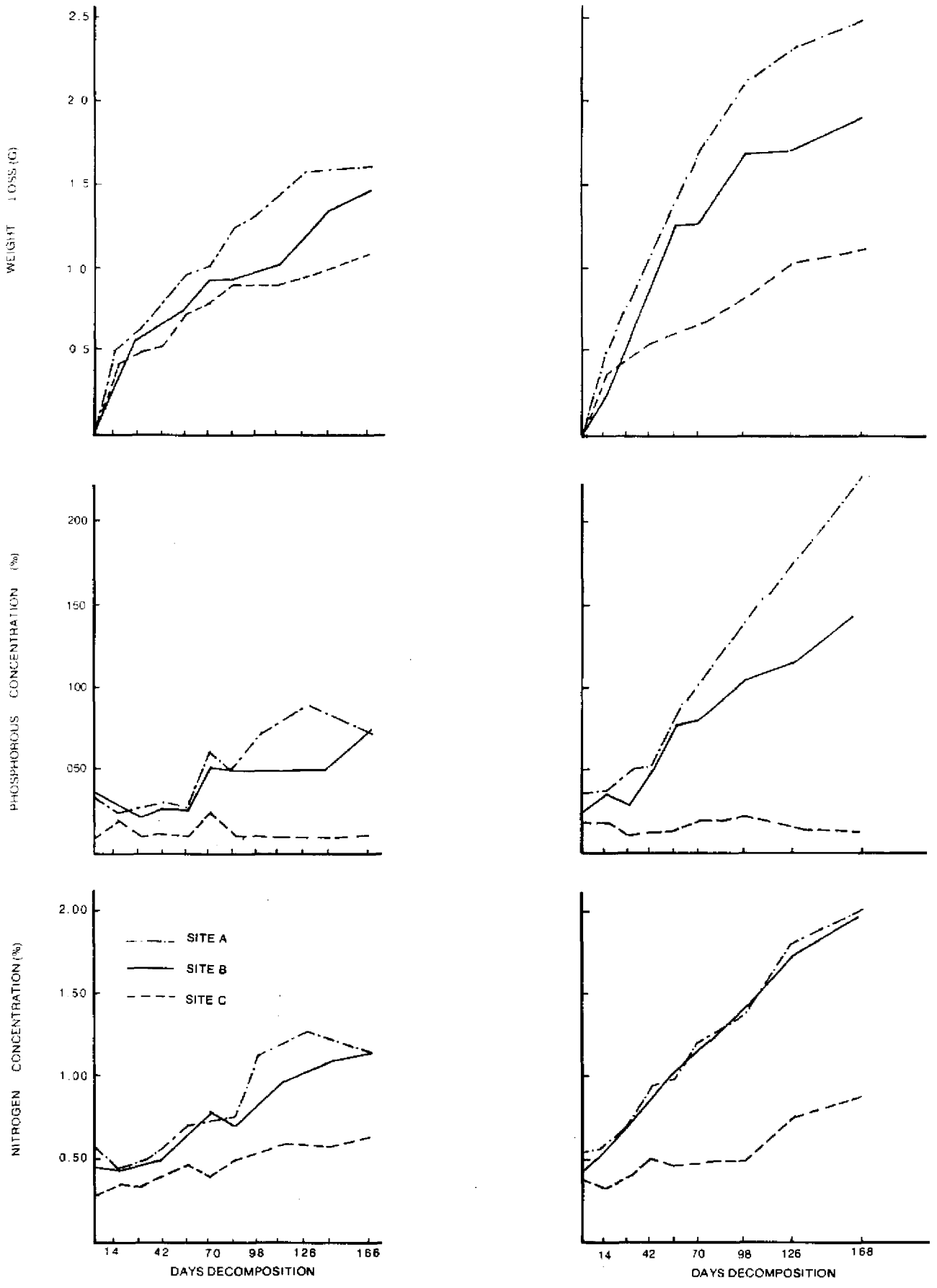

Figure 3 WEIGHT LOSS AND NUTRIENT CONCENTRATIONS OF DEAD LEAF MATERIAL PLACED IN LITTERBAGS IN WATER CONSERVATION AREA 2A

changed little. Nitrogen concentration increases at Sites A, B, and C were 0.58-1.15%, 0.46-1.15%, and 0.28-0.64% in sawgrass litter and 0.54-2.02%, 0.42-1.98%, and 0.37-0.87% in cattail litter. Cattail leaf litter accumulated significantly more P and N than sawgrass in the enrichment zone, but not at the non-enriched location.

Lignin, Cellulose and Residual Ash in Dead Leaves

Lignin, cellulose and residual ash concentrations in freshly dead leaf material (Table 2) did not differ significantly between nutrient-enriched Site B and non-enriched Site C. Freshly dead sawgrass leaves did differ significantly from those of cattail in these constituents. Lignin and cellulose concentrations were slightly higher in cattail in comparison to sawgrass. Residual ash concentrations showed a more pronounced difference between the two species; residual ash concentrations in sawgrass dead leaves were 7 times those in cattail at nutrient-enriched Site B and 12 times those of cattail at non-enriched Site C.

TABLE 2. MEAN LIGNIN, CELLULOSE AND RESIDUAL ASH CONCENTRATIONS (% DRY WEIGHT) IN FRESHLY DEAD LEAVES COLLECTED FROM LIVING PLANTS. N=6.

	Sawgrass		Cattail	
	Site B	Site C	Site B	Site C
Lignin	20.43	21.67	22.72	23.49
Cellulose	29.91	31.12	36.58	34.03
Residual Ash	3.02	4.84	0.44	0.39

Bacteria and Fungi

Counts of bacteria and fungi inhabiting sawgrass litter (Figure 4) were significantly lower at the two nutrient enriched sites compared to the non-enriched location (Table 3). Counts of fungi within the enrichment zone

TABLE 3. SITE DIFFERENCES IN LITTER MICROBIAL DENSITIES. VALUES REPRSENT MEANS FOR SIX MONTHS ± S.D. SIGNIFICANT DIFFERENCES IN MEANS ARE INDICATED BY * ($p < 0.05$) AND ** ($p < 0.01$).

	SITES			Test on Difference Between Means
	A	B	C	
SAWGRASS				
Algae (mg)/g dry wt litter	2.8 ± 3.2 n = 9	2.5 ± 2.1 n = 9	1.4 ± 1.4 n = 9	N.S.
Bacteria ($x10^7$)/g dry wt litter	3.4 ± 1.4 n = 9	5.1 ± 3.8 n = 9	17.7 ± 9.5 n = 9	** C > A, B
Fungi ($x10^4$)/g dry wt litter	7.8 ± 6.0 n = 9	33.0 ± 20.7 n = 9	101.6 ± 70.8 n = 9	** ** C > B > A
CATTAIL				
Algae (mg)/g dry wt litter	3.3 ± 2.0 n = 8	1.9 ± 1.0 n = 8	1.5 ± 1.2 n = 8	N.S.
Bacteria ($x10^8$)/g dry wt litter	2.5 ± 1.5 n = 8	2.8 ± 1.6 n = 8	2.3 ± 0.9 n = 8	N.S.
Fungi ($x10^4$)/g dry wt litter	5.6 ± 2.8 n = 7	18.0 ± 23.7 n = 8	12.2 ± 10.2 n = 8	N.S.

were significantly higher at Site B than at Site A. The site differences in microbial population densities which were found for sawgrass litter were not evident for cattail litter. Counts of bacteria and fungi inhabiting cattail litter (Figure 4) did not differ significantly between the nutrient enriched and non-enriched sites.

Comparison of microflora densities between litter types indicates that cattail yielded significantly larger counts of bacteria than did sawgrass litter at the nutrient enriched sites but not at the non-enriched location. However, sawgrass litter tended to support larger fungal populations. Fungal counts in cattail leaves at the enriched sites on the fourteenth day of decomposition were much higher than counts on sawgrass leaves; however, after 14 days, counts declined rapidly in cattail leaves at all three sites. Comparisons of mean values for the six month study period showed that fungal densities overall were significantly higher in sawgrass litter than in cattail at Sites B and C.

In contrast to the litter microflora, planktonic bacteria and fungi were found in higher densities at the nutrient enriched sites compared to the non-enriched location in a sample of surface water. Colony-forming units of bacteria/ml at Sites A, B, and C were 2.2×10^4, 1.3×10^4, and 2.0×10^3, respectively. Counts of fungal colonies averaged 190/ml at Site A, 70/ml at Site B, and 7/ml at Site C.

Attached Algae

Biomass estimates of attached algae (mg/g dry weight of leaf litter) fluctuated considerably at all three sites (Figure 4). When averaged over the six month decomposition period, algal biomass was higher on litter at the nutrient enriched sites than at the non-enriched site in both sawgrass and cattail (Table 3); however, site differences were not statistically significant. The predominant species of attached algae on both sawgrass and

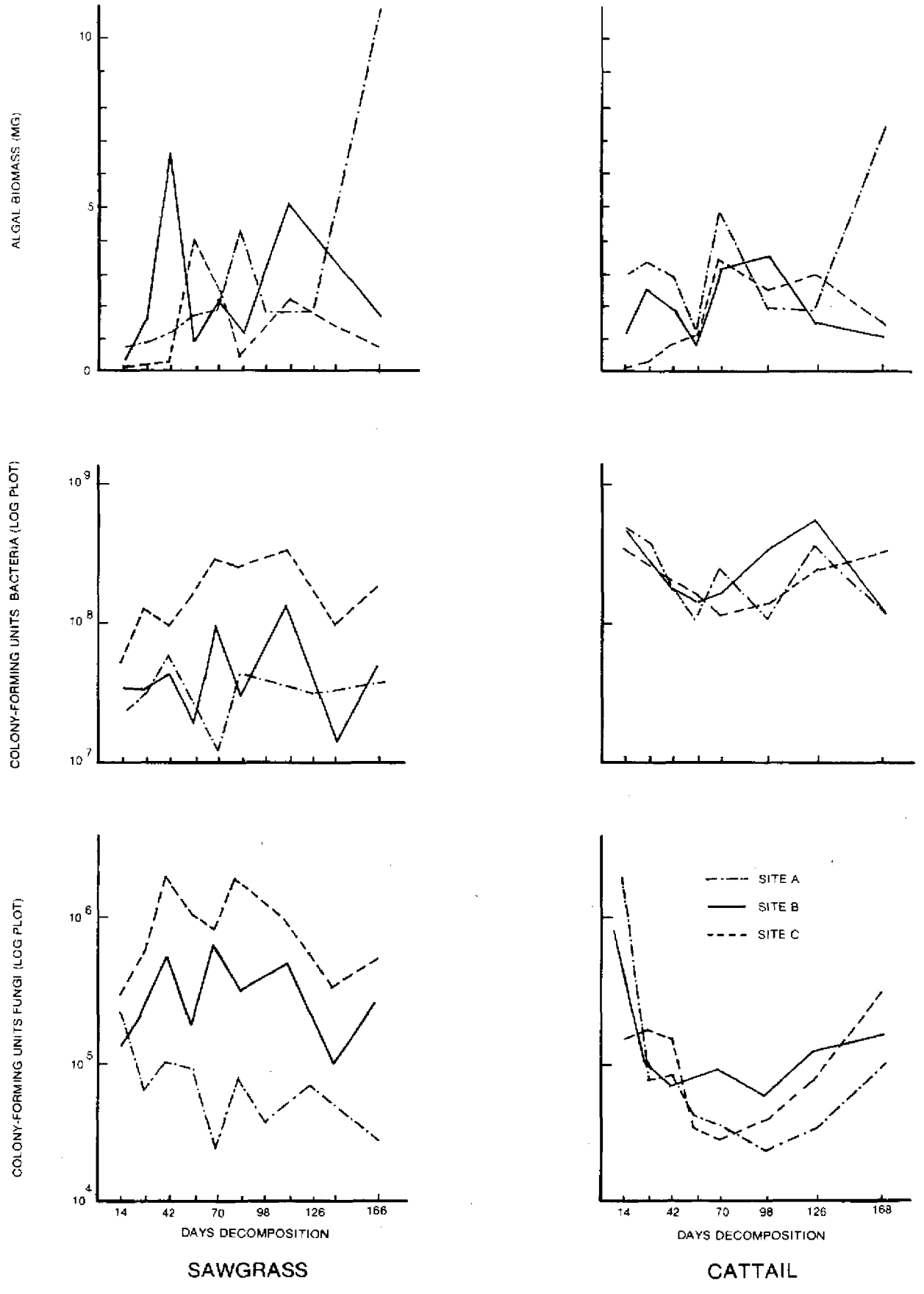

Figure 4 BIOMASS ESTIMATES PER GRAM DRY WEIGHT LITTER OF ALGAE, BACTERIA AND FUNGI IN WATER CONSERVATION AREA 2A

cattail litter at all three sites were the filamentous blue-greens Microcoleus sp. and Schizothrix calcicola, the coccoid blue-green Microcystis sp., and the diatoms Nitzschia spp. and Synedra sp.

DISCUSSION

Greater weight loss and nutrient uptake by leaf litter in the zone of nutrient enrichment than at the non-enriched location paralleled the findings of Davis and Harris (1978). Leaf decomposition and nutrient uptake have also been shown to be related to nutrient supply by Kaushik and Hynes (1971), Howarth and Fisher (1976), Coulson and Butterfield (1978), Fell and Master (1980), and Fell et al. (1980). Larger weight loss and nutrient uptake by cattail litter compared to sawgrass in the enrichment zone also resembled the results of Davis and Harris (1978). Differences between leaf species in decomposition rate and nutrient uptake have also been reported by Kaushik and Hynes (1968, 1971) and Coulson and Butterfield (1978). The higher rates of leaf decay in the litter containing higher P and N concentrations (i.e. cattail compared to sawgrass and enriched compared to non-enriched) agrees with the findings of Witkamp (1966), Kaushik and Hynes (1971), Coulson and Butterfield (1978), and Puriveth (1980).

Techniques for sampling sediment-inhabiting microorganisms are still in the early stages of development, and caution must be exercised in extrapolating estimates of biomass to ecological activity. The dilution plate technique was chosen for enumeration of bacteria and fungi in this study despite its limitations for ecological studies (Garrett, 1951; Witkamp, 1963; Parkinson et al., 1971; Jannasch, 1972; Parkinson, 1973) because it has been shown to be a valid method for comparing microbial populations relative to environmental differences (Witkamp, 1963, 1966; Schmidt, 1973; Maltby, 1975). Correlations between plate counts and respiration rates have also been reported by Stevenson (1956) and Witkamp (1966). The plate count technique was found by Seyfried and Owen (1979) to yield significantly higher rates of microbial recovery

from aquatic sediments than the most probable number technique, which has been used for the majority of bacteriological assessments of aquatic sediments.

The rapid colonization of both sawgrass and cattail leaves by fungi during the first two weeks of decomposition at the enriched sites follows a pattern typical of many aquatic systems (Garrett, 1951; Kaushik and Hynes, 1971).

The smaller populations of aerobic and facultative micro-organisms on sawgrass litter in the enrichment zone were an unexpected result, since we had hypothesized that these organisms were partially responsible for the increased decomposition and nutrient uptake occurring there. The low D.O. readings at the enriched sites indicate that anaerobic conditions probably existed in the litter most of the time. Anaerobic conditions provide a likely explanation for the lower populations of aerobic and facultative microflora in sawgrass litter at the enriched sites. Pugh (1961) postulated that low counts of fungi recovered from salt marsh muds were caused by low redox potentials and toxic effects of accumulated sulphides.

Since aerobic and facultative bacteria and fungi and attached algae were not more abundant on leaf litter in the enrichment zone, the question remains as to what caused the oxygen depletion, increased decomposition rates, and increased nutrient uptake in the litter there. Respiration by the larger populations of planktonic bacteria and fungi that were found in the surface water at the enriched sites would be expected to partially contribute to oxygen depletion. Oxygen depletion was probably also enhanced by chemical oxidative processes occurring within the litter as well. Hargrave (1972) found that oxygen uptake by subsurface lake sediments below two centimeters was due exclusively to chemical oxidation.

Previous work on nutrient uptake by lake sediments suggests that some of the additional phosphorus uptake during leaf decomposition in the

enrichment zone of WCA-2A may have been through physical adsorption rather than biological processes. Controversy concerning the importance of sorptive processes in phosphorus uptake by detritus revolves around the role of organic matter in these processes, since the decomposing leaf material is largely organic. Harter (1969) provided evidence that organic matter is important in the initial bonding of phosphorus by soils and suggested that this mechanism may explain the nutrient status of lakes, since sediments of nutrient-rich lakes are usually high in organic matter and have a very high phosphorus adsorption capacity. Jervis (1969) proposed that the colloidal clay fraction of inorganic soil has an equally effective counterpart in organic material for phosphorus adsorption. This conclusion was based on the findings of Albrecht (1941) that organic colloids increase with total organic matter, the results of Millar et al. (1936) that organic colloids increase with decomposition, and the demonstration by McGeorge (1930) that organic colloids fix exchangeable ions on a chemically equivalent basis. Thus, as detritus decomposes, adsorption processes would be expected to become more important in phosphorus uptake due to increased surface areas and with the formation of organic colloids. More rapid decomposition rates in the zone of nutrient enrichment of WCA-2A would therefore be expected to be accompanied by increased physical adsorption of phosphorus; however, the cause of the more rapid decomposition rates in this zone remains unclear.

Litter decomposition rates have been shown to be inversely related to the initial lignin content of the dead leaf material (Melillo et al., 1982; Cromack, 1973; Fogel and Cromack, 1977). However, the differences in decomposition rate between sawgrass and cattail in this study cannot be attributed to lignin. The more rapidly decomposing cattail litter contained slightly higher initial concentrations of both lignin and cellulose in comparison to the more slowly decomposing sawgrass. The lower residual ash

concentration in cattail dead leaves may have contributed to its more rapid breakdown in comparison to sawgrass. Residual ash in the fiber analysis scheme is not readily broken down, and the higher concentrations found in sawgrass probably indicate a higher silica content. Since lignin, cellulose and residual ash concentrations in dead leaves did not differ significantly between the nutrient-enriched and non-enriched sites, they failed to explain the more rapid leaf decomposition rates in the enrichment zone.

This study was undertaken to test the hypothesis that increased decomposition and nutrient uptake by leaf litter in the enrichment zone of WCA-2A resulted from increased microbial populations colonizing the litter. Implicit in this hypothesis was the assumption that the litter microbial communities in WCA-2A were largely aerobic or facultative. The hypothesis that these organisms are responsible for the increased rates of decomposition and nutrient uptake is rejected. Lignin, cellulose and residual ash concentrations in dead leaves also failed to explain increased decomposition rates.

In view of the anaerobic conditions which apparently prevailed in the litter in the enrichment zone, an alternative explanation is that obligate anaerobic bacteria were responsible for much of the litter decomposition there. Although this explanation remains untested, it is very plausible, since the importance of anaerobic bacteria in salt marsh communities is well documented (Wiebe, et al., 1981). Parkes et al. (1979) found a positive correlation between anaerobic cellulolytic bacteria and rates of cellulose decomposition in estuarine sediments. Jorgensen (1977) reported that sulfate-reduction alone accounted for 53% of the mineralization of organic matter in brackish water sediments. Anaerobic bacteria can become dominant when such sediments are subjected to excess organic matter, and their activities can control the degradation of these compounds (Poole et al., 1977). In contrast

to salt marshes, the role of anaerobic bacteria in the metabolism of freshwater wetlands is poorly understood.

Cappenberg (1972) found obligate anaerobic sulfate reducing bacteria in the hypolimnion of a freshwater pond. Cappenberg provided evidence that facultative heterotrophic bacteria and methane oxidizing bacteria created the anerobic conditions favorable to the sulfate reducers. The elevated numbers of aerobic or facultative bacteria in the water column and the reduced oxygen concentrations in the litter layer at the nutrient-enriched WCA-2A sites parallel Cappenberg's findings.

A shift from aerobic and facultative micro-organisms to obligate anaerobes as the primary mode of leaf decomposition in nutrient-enriched areas would probably affect Everglades ecology in many ways. Detritus characteristics, food webs and nutrient uptake pathways would be likely areas of impact. Such a shift is currently supported only by indirect evidence. The dominance of anaerobes can be verified only by direct measurements of density or metabolism. The role of anaerobic bacteria in the ecology of the Everglades deserves further consideration in environmental research and possibly in the assessment of management alternatives for the Water Conservation Areas.

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