J. N. Am. Benthol. Soc., 1987, 6(3):147–158

Effects of acidification on leaf decomposition in streams

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Abstract. Effects of acidification on leaf decomposition in streams were studied in four second-order streams in the Great Smoky Mountains National Park. The streams ranged in pH from 4.5 to 6.4 at baseflow. Mass loss of leaves incubated in mesh bags placed in pools in each stream was measured periodically over 15 wk beginning in late August. Measurements were also made of C, N, P, and Al in leaves, microbial biomass (adenosine triphosphate [ATP]) and respiration rate and bacterial production (thymidine uptake) associated with leaves, and the number and biomass of macroinvertebrates in leaf bags.

Rates of leaf mass loss were significantly lower in streams with pH $\leq$5.7 compared with a stream with pH 6.4. Although rate of leaf mass loss among the streams varied directly with pH, differences between streams with pH values between 4.5 and 5.7 were not significant. Microbial ATP and respiration rates and bacterial production rates followed the same pattern as leaf mass loss rate, i.e., low for more acidic streams and highest in the stream with the highest pH. Accumulation of aluminum by the leaf-microbe complex was also greatest in the most acidic streams. The number and biomass of macroinvertebrate shredders found in leaf bags was lowest at the highest pH site and therefore cannot account for the higher rate of leaf mass loss found at this site. Our results suggest that the lower rate of leaf decomposition in the more acidic streams is due largely to low rates of microbial activity.

Key words: acidification, streams, leaf decomposition, microbial activity.

Various studies have shown that decomposition of organic matter is slow at low pH in lakes (Andersson 1985, Francis et al. 1984, Grahn et al. 1974), reservoirs (Carpenter et al. 1983), streams (Friberg et al. 1980, Harrison 1958, Hildrew et al. 1984, Kimmel et al. 1985, Mackay and Kersey 1985, Minshall and Minshall 1978), artificial stream channels (Allard and Moreau 1985, Burton et al. 1985), and microcosms (McKinley and Vestal 1982, Traaen 1980). However, decomposition of organic matter in lake sediments was reported to be unaffected by acidification of the water column, probably because alkalinity-generating processes associated with anaerobic catabolism maintained sediment pH at or above 6.0 (Gahnstrom et al. 1980, Hoeniger 1985, Kelly et al. 1984).

The low decomposition rate of organic matter in highly acidic surface waters could be due either to low microbial activity or to a scarcity of detrivorous macroinvertebrates. Several studies have shown that microbial numbers and/or activity associated with decomposing organic matter are low at low pH, particularly when pH is <5 (Hendrey et al. 1976, Minshall and Minshall 1978, Rao et al. 1984, Traaen 1980). Other studies have shown that low numbers of macroinvertebrates are associated with detritus at low pH (Burton et al. 1985, Friberg et al. 1980). However, none of these studies involved simultaneous measurement of decomposition rate, microbial activity, and macroinvertebrate shredder abundance. Carpenter et al. (1983) reported that decomposition rate of leaf litter at highly acidic reservoir sites was lower than at circumneutral sites and that this difference was primarily due to reduced microbial activity at the acidic sites rather than lower densities of macroinvertebrates. The role of macroinvertebrates in leaf decomposition in streams, however, may be greater than that in lentic reservoir environments.

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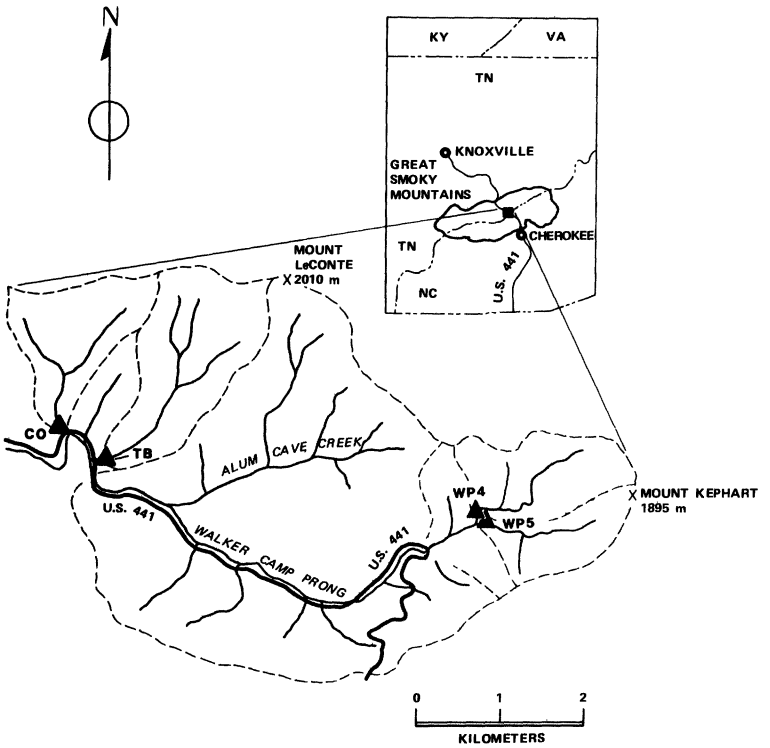

FIG. 1. Map of the Walker Camp Prong drainage showing sampling sites and topographic boundaries.

The purpose of our study was to simultaneously measure leaf litter decomposition rate, microbial activity, and macroinvertebrate abundance in four forested headwater streams that ranged in pH from 4.5 to 6.4. We measured mass loss and chemical changes in decomposing leaves, and microbial biomass (ATP) and respiration rates (O_2 consumption), bacterial production rates (thymidine uptake), and abundance of macroinvertebrates associated with decomposing leaves over a 15-wk period in each of these streams. Although our experimental design did not include replication of stream pH, the range in pH among the study streams permits us to make inferences concerning the magnitude of effects over a pH gradient frequently found in regions affected by acid deposition.

Study Sites

Four high-elevation, second-order streams in the Great Smoky Mountains National Park (eastern Tennessee, USA) were selected for the study (Fig. 1). These streams are within 10 km of each other, but vary in mean annual pH from 4.5 to 6.4 at baseflow. Pyrite oxidation at exposed high-elevation outcrops of a pyritic carbonaceous phyllite in some of the stream catchments may explain the differences in pH between the streams (Huckabee et al. 1975). Vegetation is dominated by mature stands of red spruce, Fraser fir, and yellow birch at the higher elevations (>1500 m) in all catchments; beech and hemlock are also important at lower elevations and in riparian zones. *Rhododendron* is abundant, forming a dense subcanopy at high elevations and near each of the streams.

Two of the streams (WP4 and WP5) form the headwaters of Walker Camp Prong; the other two, Cole Creek (CO) and Trout Branch (TB), are tributaries of Walker Camp Prong, joining it approximately 10 km downstream from the WP4-WP5 confluence (Fig. 1). Physical and baseflow chemistry data for each stream are presented in Table 1. The streams are similar in size, are low in acid-neutralizing capacity and total ionic strength (as indicated by specific conductance), and vary in mean annual pH (from

4.5 to 6.4) and in mean annual total monomeric aluminum (from 0.018 to 0.242 mg/L) at baseflow. The most acidic stream (WP4) is elevated in aluminum, manganese, and iron relative to the concentrations in the less acidic streams (Table 1). Concentrations of Cd, Cu, Hg, Pb, and Zn were undetectable at all sites.

Methods

Sugar maple (*Acer saccharum* Marsh.) leaves were collected just after abscission in autumn 1984. The leaves were leached in distilled water for 1 d, and air dried for at least 7 d in a greenhouse. A subsample of the leaves was oven-dried (80°C for 48 hr) to allow conversion from air-dried to oven-dried mass. Nylon mesh bags (20 × 20 cm; 4 mm mesh) were filled with 5.00 ± 0.05 g of air-dried leaves (4.35 g ash free dry mass [AFDM]). Fifty bags were placed in each stream on 27 August (TB and CO) and 29 August 1985 (WP4 and WP5). In each stream, ten bags were strung on nylon line and staked to the streambed in each of five pools, where they were sheltered from high turbulence during storms. At intervals of 7, 13, 28, 49, 70, 84, and 105 d, twenty bags, one from each of the five pools in each stream, were each placed in a plastic bag filled with stream water from the collection site, and transported on ice to the laboratory.

During each collection, water temperature, specific conductance, and pH of the stream were measured, and water samples were collected for analysis of NH_4^+, NO_3^-, soluble reactive phosphorus (SRP), and total soluble phosphorus (TSP). These parameters were also measured monthly as part of a routine sampling program at all sites. Water temperature was measured with a thermistor, specific conductance with a Cole Parmer® Model PCM 1 conductivity bridge, and pH potentiometrically with a glass Ross® combination electrode. Water samples for P analyses were filtered through 0.4-μm filters (Nuclepore®), placed on ice, and returned to the laboratory. SRP was analyzed using the ascorbic acid method and TSP using the ascorbic acid method following persulfate digestion (APHA 1985). Analyses of NH_4^+ and NO_3^- were by phenate colorimetry (U.S. EPA 1983) and ion chromatography (Small et al. 1975), respectively.

In the laboratory, leaf material in each bag was carefully rinsed in stream water to remove adhering sediments and macroinvertebrates. Leaf discs (cut with a cork borer and avoiding major veins) were used for chemical analyses (C, N, P, and Al) and for measurements of ATP, bacterial production, and respiration. Macroinvertebrates separated from the leaf material were identified, grouped into functional categories according to Merritt and Cummins (1984), and counted. The macroinvertebrates were then dried at 60°C for 48 hr and weighed to determine dry mass per bag. Remaining leaf material was placed in a tray, dried at 80°C for at least 72 hr, weighed, combusted at 500°C for at least 16 hr, and reweighed to determine the AFDM.

Ten pooled leaf discs (1.7 cm diameter each) taken from leaves in each bag were dried at 80°C for 48 hr, weighed to determine dry mass, and ground in a Wiley mill. Approximately 1.5- to 2.0-mg subsamples of the pulverized material were than analyzed for C and N using a CHN Analyzer (Perkin Elmer® Model 240B). To extract total P and Al, oven-dried pulverized leaf material (ten discs pooled from each bag, 1.7 cm diameter each) was digested in concentrated nitric (3 ml) and perchloric acid (1 ml) heated to 120°C for 8 hr. Concentrations of P and Al in the digestate were measured by inductively coupled plasma emission spectroscopy. Analysis of National Bureau of Standards standard pine needles using these techniques gave 89.7 and 87.5% recoveries for P and Al, respectively.

ATP was measured as an indicator of total microbial biomass associated with decomposing leaves. ATP was extracted from five pooled leaf discs (1 cm diameter each, cut from leaves in each bag) in 5 ml of 1 M H_3PO_4 for 24 hr. Subsamples (0.5 ml) of the extract were frozen and stored until analysis. Thawed extract subsamples were diluted with 0.02 M Tris buffer and ATP was measured using a Lumac Biocounter® with internal standardization performed for each subsample.

Tritiated thymidine incorporation into DNA was measured as an indicator of bacterial production on leaves. Five leaf discs (1 cm diameter each) cut from five different leaves from each bag were placed in a 50-ml round-bottom centrifuge tube containing 5 ml of stream water from the site at which the bag was taken. The tubes were equilibrated to the measured stream temperature, and unlabeled (0.64 mol) and tritiated (0.16 nmol) thymidine (specific activity

TABLE 1. Physical and chemical data for the study streams. Chemical values are averages of monthly samples from June 1984 to February 1986 for WP4 and WP5 and from March 1985 to February 1986 for CO and TB (range in parentheses). All values are in mg/L unless shown otherwise.

Parameter	WP4		TB		WP5		CO	
Drainage area (km^2)	1.8		2.8		1.2		1.8	
Average stream width (m)	5		4		4		3	
Specific conductance ($\mu S/cm$)	30.8	(26.7–37.3)	22.9	(21.4–26.2)	19.6	(17.1–22.4)	19.1	(16.8–21.4)
pH	4.5	(4.4–4.6)	5.0	(4.9–5.1)	5.7	(5.5–6.0)	6.4	(6.2–6.8)
ANC ($\mu eq/L$)[a]	−30.8	(−48.2–−18.4)	−15.3	(−26.0–−7.0)	−0.1	(−13.6–+9.8)	19.9	(1.3–54.0)
SO_4^{2-}	4.0	(3.4–5.0)	4.2	(3.7–5.2)	2.8	(2.2–3.5)	2.0	(1.6–2.5)
NO_3^-	4.0	(2.6–5.5)	3.5	(2.5–4.3)	4.6	(3.8–5.4)	3.9	(2.6–4.7)
Ca	1.2	(1.0–1.4)	1.2	(1.1–1.4)	1.7	(1.4–1.9)	1.5	(1.4–1.7)
Al (total monomeric)[b]	0.251	(0.133–0.368)	0.135	(0.080–0.188)	0.025	(0.010–0.081)	0.022	(0.011–0.047)
Al (organic monomeric)[b]	0.054	(0.036–0.078)	0.013	(0.005–0.019)	0.009	(0.006–0.018)	0.020	(0.009–0.047)
Fe	0.050	(0.023–0.090)	0.004	(0.004–0.006)	0.003	(0.002–0.004)	0.006	(0.004–0.010)
Mn	0.056	(0.026–0.093)	0.038	(0.021–0.059)	0.003	(0.001–0.012)	0.001	(0.001–0.004)
SRP ($\mu g/L$)[c]	1.4	(<0.5–3.4)	0.6	(<0.5–1.0)	0.8	(<0.5–3.5)	1.2	(<0.5–1.9)
TSP ($\mu g/L$)[d]	3.4	(0.9–5.0)	1.6	(0.6–3.9)	2.0	(0.5–7.0)	3.2	(1.0–5.0)
Dissolved organic carbon	2.1	(1.4–3.0)	0.7	(0.4–1.4)	0.5	(0.2–0.8)	1.1	(0.7–1.7)

[a] Acid neutralizing capacity.
[b] Fractionation technique of Driscoll (1984).
[c] Soluble reactive phosphorus.
[d] Total soluble phosphorus.

2.886×10^{12} Bq/mmol) were added to each tube. The discs were incubated for 20 min, after which 0.5 ml of 37% formaldehyde was added. The liquid in the tubes was discarded and the leaf discs were rinsed three times with distilled water. The extraction of the labeled DNA from leaves followed the methods of Findlay et al. (1984) and Palumbo et al. (1987a).

Respiration of the microbial community on leaves was measured with a Gilson® differential respirometer. Nine pooled leaf discs (1 cm diameter each) cut from the material in each bag and 5 ml of stream water from that site were placed in each respiration flask. Controls were three flasks containing 5 ml of stream water from each site. After a 2-hr equilibration period, oxygen consumption was measured at constant temperature over a 3-hr period. Respiration measurements for each sampling interval were made at the same temperature for all sites; this temperature was always within 2°C of ambient stream temperature at the time of sampling at each site.

Total dry mass and AFDM of leaves in each bag on each sampling date were calculated as the sum of the oven-dry mass and AFDM of all of the discs cut plus the remaining bulk material. Dry mass and AFDM of the discs used for the respiration studies were measured after the respirometer incubations. AFDM of the discs used for chemical analyses was calculated using the measured dry mass of the discs and the ratio of AFDM : dry mass measured for the discs used in respiration studies. Dry mass and AFDM of the remaining discs cut from the bulk material were estimated assuming the same mass : area ratio as the respiration studies discs. Rate of decomposition (k) of the leaves was calculated using linear and exponential decay models for leaf AFDM loss over time (t) (Petersen and Cummins 1974):

$$AFDM_t = AFDM_0 - k(t), \quad \text{and}$$
$$AFDM_t = (AFDM_0)e^{-kt}$$

where $AFDM_0$ is the initial leaf AFDM.

Results

Differences in water chemistry parameters directly related to pH were conspicuous among streams (Table 1). In general, the more acidic streams had higher specific conductance, higher

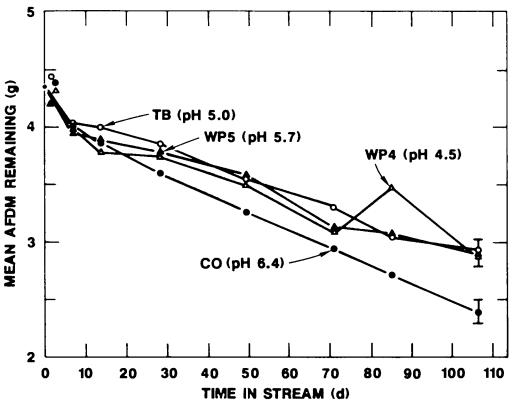

FIG. 2. Time course of leaf AFDM remaining in bags at each site. ±1 SE is shown for the last sampling date (105 d).

dissolved SO_4^{2-}, Fe, Mn, and Al concentrations, and lower ANC and Ca^{2+} concentrations. Depressions in pH and increases in total monomeric Al have been observed in the highest pH streams during large storms (pH as low as 5.5 at CO and 5.0 at WP5). However, such depressions are relatively short (a few days) and infrequent (<10 per year). Values of dissolved inorganic N were high relative to TSP at all sites (N/P > 1000 by weight), and P concentrations were highest at WP4 and lowest at TB. Water temperatures at TB and CO were similar, but temperatures at the higher-elevation sites (WP4 and WP5) were 0.8 and 1.1°C lower, respectively. There was one large storm during the study period (early November), but the magnitudes of pH depressions are unknown.

During the 15-week study, AFDM of leaves declined from an initial 4.36 g/bag at all sites to values ranging from 2.97 (1 SE = 0.11) g/bag at TB to 2.40 (1 SE = 0.12) g/bag at CO, the highest pH site (Fig. 2). Concomitant gains were noted in concentrations of N, P, and Al, and declines occurred in the C/N ratio of leaves (Fig. 3). The rather large AFDM losses during the first week at each site were probably due to leaching of the remaining soluble leaf constituents. Linear and exponential decay models fitted the leaf mass loss data from 7 to 105 d at each site about equally well (r^2 values were within 0.01 for each site). The order of sites according to leaf mass loss rate constants calculated using the exponential loss model was WP4 < TB < WP5 < CO, matching the trend

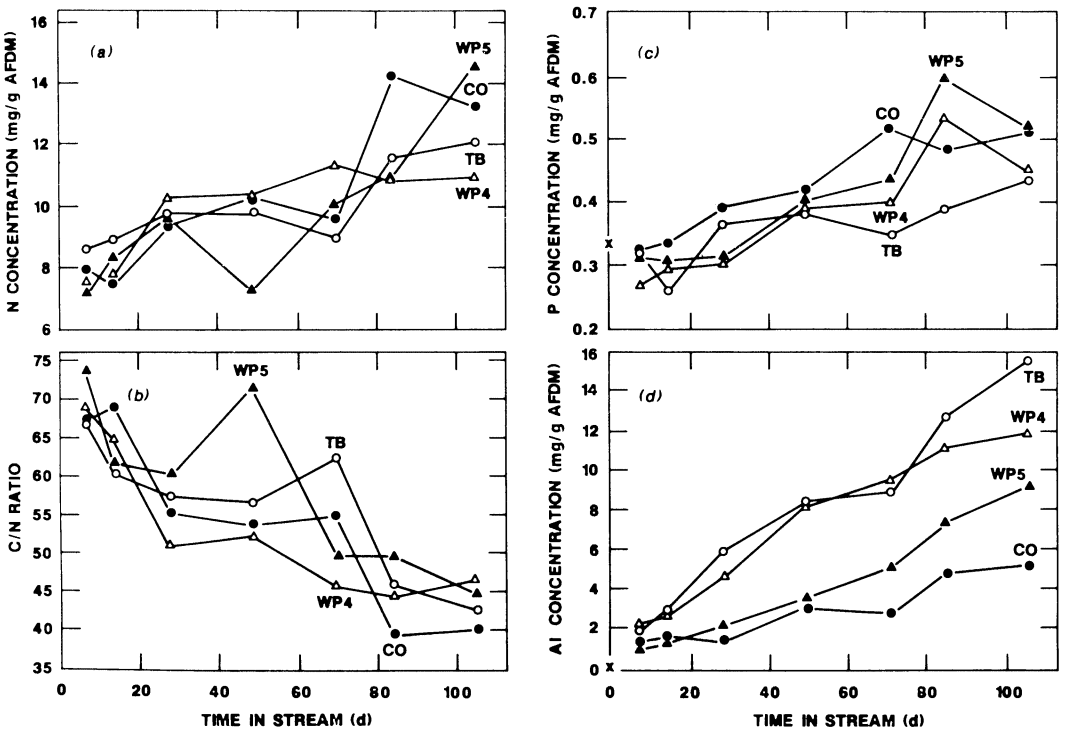

FIG. 3. Time course of (a) N concentration, (b) C/N ratio, (c) P concentration, and (d) Al concentration in leaves at each site.

in pH (Table 2); however, the only statistically significant differences in leaf mass loss rate constants among the sites were between CO and each of the other streams (pairwise *t*-tests of rate constants, $p < 0.05$).

The rate of increase in N concentration in leaves was significantly lower in the more acid streams (WP4 and TB), and the rate of increase in P concentration was lowest in TB but was not significantly different among the other sites (Table 2). Rates of increase in leaf Al were significantly different among all sites and were highest at TB and lowest at CO.

Total microbial biomass, as indicated by ATP on decomposing leaves, was greatest at CO on all sampling dates (Fig. 4a). ATP concentrations increased rapidly at all sites during the first 28 days, and generally leveled off thereafter. ATP concentrations were lowest at TB at 84 d, whereas at WP4 and WP5 they were similar except at 70 d. One-way ANOVAs performed on the ATP data for each sampling date indicated significant differences among streams for all dates (df = 3,16, $p < 0.05$). On all dates ATP was significantly greater at CO than at the other sites and ATP was significantly greater at WP5 than at TB and WP4 on two of the seven sampling dates (Duncan's multiple range test, $p < 0.05$) (SAS 1982).

Bacterial production, as indicated by thymidine incorporation into DNA, was also greatest at CO and least at WP4 on all sampling dates (Fig. 4b). One-way ANOVAs for each sampling date indicated that thymidine uptake was significantly higher at CO compared with the other sites on all but the first sampling date (Duncan's multiple range test, df 3,16, $p < 0.05$). Thymidine uptake rates were generally lowest at WP4 and intermediate between CO and WP4 at WP5 and TB. Rates at WP5 were significantly higher than those at WP4 on four of six dates at the $p < 0.10$ level (see also Palumbo et al. 1987b).

Microbial respiration rate (oxygen consumption rate) was also highest at CO, but only during the first 28 to 49 d (Fig. 4c). Unlike ATP, microbial respiration rates declined in all streams after 28 d (49 d in the case of WP4), and

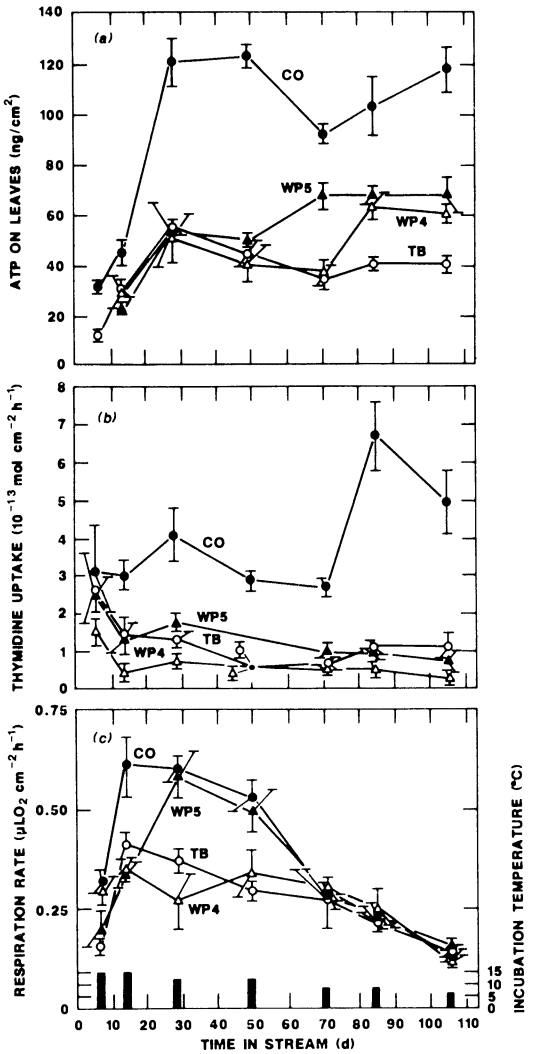

FIG. 4. Time course of (a) ATP, (b) rate of thymidine incorporation into DNA (bacterial production), and (c) respiration rate of leaves at each site (±1 SE). Also given in (c) is the temperature at which thymidine incorporation and respiration rates were measured.

were similar in all streams at 70 d and thereafter. One-way ANOVAs indicated that respiration rates were significantly higher at CO at 13 d and significantly higher at CO and WP5 at 28 d and 49 d, compared with the other sites (Duncan's multiple range test, df = 3,16, $p < 0.05$).

Cumulative leaf mass loss resulting from microbial respiration alone was estimated for each

TABLE 2. Calculated AFDM loss rate constants (per day), and rates of increase in leaf N, P, and Al concentration (mg/g AFDM/d) between 7 d and 105 d in each stream. 1 SE and r^2 are given in parentheses. Significant differences in rates among sites are denoted by different capital letters following the rate values (pairwise t-tests, $p < 0.05$).

Stream	pH	AFDM Loss Rate	Leaf N Increase	Leaf P Increase	Leaf Al Increase
WP4	4.5	0.00283 (0.00056, 0.84) A	0.0350 (0.0078, 0.40) A	0.00229 (0.00038, 0.52) B	0.1062 (0.0079, 0.84) C
TB	5.0	0.00327 (0.00014, 0.98) A	0.0312 (0.0078, 0.33) A	0.00122 (0.00039, 0.23) A	0.1330 (0.0112, 0.81) D
WP5	5.7	0.00362 (0.00024, 0.96) A	0.0593 (0.0087, 0.59) B	0.00271 (0.00048, 0.49) B	0.0828 (0.0081, 0.76) B
CO	6.4	0.00509 (0.00015, 0.99) B	0.0637 (0.0088, 0.62) B	0.00205 (0.00032, 0.55) B	0.0409 (0.0070, 0.67) A

TABLE 3. Estimated carbon loss resulting from microbial respiration during the study, and the respiratory AFDM loss to total AFDM loss ratio for leaves in each stream (assuming an RQ of 1.0 and a carbon content of 50% of leaf AFDM).

Stream	pH	Respiratory Carbon Loss (g/bag)	Respiratory AFDM Loss/Total AFDM Loss
WP4	4.5	0.271	0.62
TB	5.0	0.263	0.51
WP5	5.7	0.330	0.56
CO	6.4	0.393	0.51

site using the oxygen consumption data (computed per unit leaf AFDM) and assuming a respiratory quotient (RQ, moles CO_2 respired/moles O_2 consumed) of 1.0 and a leaf organic carbon content of 50% of AFDM. Use of an RQ of 1.0 is appropriate for metabolism of carbohydrate but may be somewhat high for metabolism of other organic molecules (e.g., RQ for protein metabolism is 0.8). Because we do not know the composition of leaf material metabolized but believe it to be dominated by carbohydrates, we have assumed an RQ of 1.0 and realize this may give slightly high metabolic carbon loss values. Respiration rates measured at laboratory temperatures were converted to rates expected for ambient stream temperatures measured at the time of leaf collection (a conversion of ≤ 3°C for each site on each sampling date) using a Q_{10} value of 2. To calculate cumulative respiration between sampling dates, the respiration rates were assumed to be constant over the period midway between the preceding and the following sampling dates. Carbon loss resulting from microbial respiration alone was greatest at CO and least at TB and WP4, and could account for 51–62% of total loss of AFDM (Table 3).

Number of shredding macroinvertebrates per leaf bag increased markedly at all sites during the first 28 d (Fig. 5a). At all but the most acidic site (WP4), shredder numbers declined somewhat thereafter. Shredder biomass per leaf bag generally increased throughout the study at all sites (Fig. 5b). Shredders were most numerous, and generally greatest in biomass, at TB. One-way ANOVAs performed on log-transformed

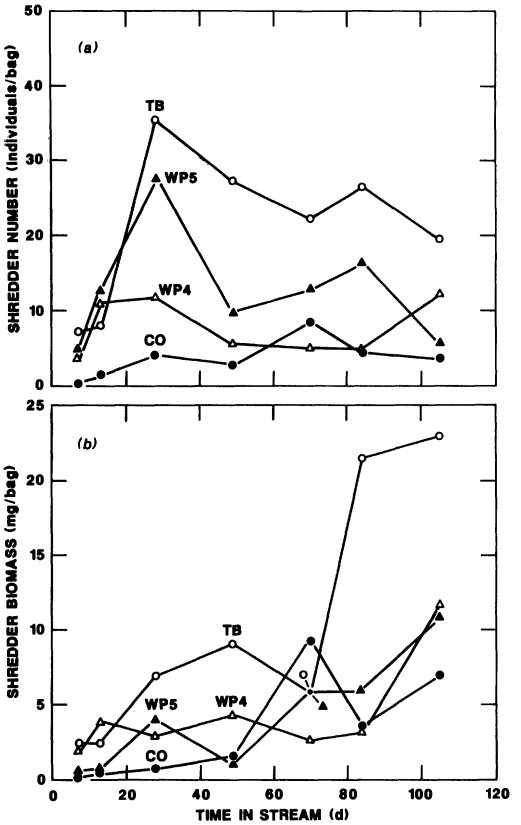

FIG. 5. (a) mean number and (b) mean biomass (dry mass) of shredders per leaf bag at each site during the study.

data for each sampling date indicated significant differences among streams on all dates for numbers, but only on 7 d, 49 d, and 84 d for biomass ($df = 3,16, p < 0.05$). Shredder numbers were significantly higher at TB than at the other sites on all but one of the sampling dates, and were significantly lower at CO compared with the other sites on the first four sampling dates (Duncan's multiple range test, $df = 3,16$, $p < 0.05$). Shredder biomass per leaf bag was significantly greater at TB compared with the other sites on 49 d and 84 d (Duncan's multiple range test, $df = 3,16$, $p < 0.05$).

Although there were substantially more shredders at TB than at WP4, the shredder communities at these highly acidic sites were similar and were dominated by the isopod *Lirceus* sp. Shredder communities at the higher pH sites of WP5 and CO were generally more diverse, and were dominated by *Lepidostoma* sp., *Leuctra*

sp., *Paracapnia opis*, *Peltoperla* sp., and the limnephilid caddisflies *Pycnopsyche* (*P. guttifer*, *P. sonso*, *P. divergens*, and *P. scabripennis*) and *Hydatophylax argus*. Nonshredders found in leaf bags at these sites were primarily collector-gatherer species of mayflies and the predaceous stonefly *Isoperla similis*.

Number and biomass of total macroinvertebrates per leaf bag were also generally highest at TB and were lowest at WP4. However, one-way ANOVAs on log-transformed data indicated that differences among sites were only significant on three of the seven dates (Duncan's multiple range test, df = 3,16, $p < 0.05$).

Discussion

The leaf AFDM loss rates measured in this study, with the exception of that at CO, were below the range (denoted by one standard error of the mean) given for maple leaves by Webster and Benfield (1986) in their review of leaf breakdown in aquatic ecosystems. The AFDM loss rate at CO, however, was slightly higher than this range.

Leaf AFDM loss rate was significantly higher at CO (pH of 6.4) than at the sites with pH $\leq$5.7 (Table 2). The low decomposition rates at TB and WP4 (sites with pH $\leq$5.0) appear to be at least partially the result of comparatively low rates of microbial decompositional activity as indicated by low levels of microbial ATP and low rates of bacterial production and microbial respiration at these sites (Fig. 4). Approximately one half of the difference in leaf AFDM loss per bag between CO and WP4 and between CO and TB could be accounted for by greater microbial respiration at CO. Only about one third of the difference in AFDM loss per bag between CO and WP5 could be attributed to greater microbial respiration.

The relatively low rates of microbial respiration and leaf decomposition at TB and WP4 compared with the rates at CO cannot be attributed to differences in temperature and phosphorus. Average temperature was 0.8°C greater at CO than at WP4, but this difference is small compared with the 1.8 times greater rate of leaf AFDM loss and the approximately twofold greater rate of respiration. The average temperature at TB was identical to that at CO. Although phosphorus concentrations were greater in CO than in TB (Table 1), phosphatase activity of the epilithic community and in the water was similar for these two sites (Mulholland et al. 1986) indicating no major differences in degree of phosphorus limitation. Phosphorus concentrations at WP4 were greater than those at CO, and phosphatase activity at WP4 was similar to that at CO (Mulholland et al. 1986).

The lower rate of microbial respiration and leaf decomposition at WP5 relative to that at CO could be due to slightly lower water temperature (on average 1.1°C lower) and lower phosphorus concentrations at WP5. Assuming a Q_{10} of 2 for microbial respiration over the temperature range of this study (6–15°C), the decomposition rate due to microbial respiration could have been as much as 8% lower at WP5 than that at CO owing to water temperature differences alone. However, the respiratory carbon loss was calculated to be 16% lower at WP5 (Table 3). Additionally, microbial ATP and bacterial production (thymidine uptake) were consistently much lower at WP5 than at CO, indicating that lower temperature alone was unlikely to be responsible for observed differences in microbial respiration. Although the epilithic community at WP5 appeared more phosphorus-limited than that at CO (as indicated by phosphatase activities; Mulholland et al. 1986), data from a short-term phosphorus enrichment study showed that bacteria associated with decomposing leaves were not P-limited. Bacterial production associated with decomposing leaves at WP5 at 84 d was not affected by additions of PO_4-P (20 μg/L) 2 hr before ^{3}H-thymidine uptake measurements were made.

There were no consistent differences among streams in leaf N concentration and C/N ratio during the study (Figs. 3a, b), despite the differences in microbial ATP and rates of bacterial production and microbial respiration, particularly between CO and the other streams (Fig. 4). This suggests that N accumulated primarily in non-microbial pools during leaf decomposition, and that low pH had little effect on N accumulation.

The major differences in water quality among our study sites were in mean pH (4.5–6.4) and in mean total monomeric aluminum concentration (0.018–0.242 mg/L; Table 1). Low measures of microbial activity (ATP, respiration, bacterial production) and low mass loss rates at WP4 and TB were likely a result of high concentrations

of H^+, Al^{3+}, or both. Maintenance of a cytoplasmic environment that is far less acidic than the external milieu is a severe problem for bacteria at low pH sites (Krulwich and Guffanti 1983). Rao et al. (1984) showed in laboratory studies that low-pH stress (pH $\leq$ 5) resulted in morphological changes in lacustrine bacteria and caused respiration and substrate uptake rates to decline. Rimes and Goulder (1986) reported that the metabolic activities of suspended bacteria in acidic streams (pH 4.6–5.3) were much lower than those in calcareous streams (pH 7.5–7.8) in Great Britain. Carpenter et al. (1983) reported less heterotrophic activity at reservoir sites influenced by acid mine drainage (pH $\leq$ 5.7) than at a control site (pH 6.3). High concentrations or altered speciation of metals could also reduce microbial activity at low pH. Babich and Stotzky (1982), for example, showed that nickel is more toxic to many microorganisms when pH is reduced from 7–8 to 5.5 and below.

In our study, the leaves at the sites with pH $\leq$5.0 (WP4 and TB) accumulated considerably more aluminum (about 4×) compared with sites with higher pH. In another study (Palumbo et al. 1987a), decomposing leaves that we transplanted from TB to CO retained relatively high levels of Al, indicating incorporation of Al by microbial biomass. Microbial activity associated with the transplanted leaves remained depressed for four weeks following the transplant compared with leaves incubated entirely in CO (Palumbo et al. 1987a). Such results suggest that improvement in water quality in acidified streams (increases in pH, decreases in Al) may not cause an immediate or rapid response in microbial activity, perhaps owing to inhibition by Al accumulated at cell surfaces or incorporated into cell biomass. Alternatively, lower microbial activity at low pH may be the result of high H^+ alone. In this case, the long lag in response after transplanting the leaves may be due to a long lag phase of growth at the low phosphorus concentrations in these streams.

The correlation between low rates of leaf decomposition and low numbers of macroinvertebrates that has been reported for several acidified streams (Burton et al. 1985, Friberg et al. 1980, Kimmel et al. 1985) was not observed in our study. Macroinvertebrate shredders were least abundant at CO which had the highest pH and greatest rate of leaf mass loss, and most abundant at TB where the rate of leaf mass loss was low. Other researchers have also found greater numbers and/or biomass of shredders at sites with low pH (i.e., those with pH values <6; cf. Hildrew et al. 1984, Mackay and Kersey 1985).

Another factor that could account for lower rates of leaf mass loss at the acidic sites is a difference in abiotic fragmentation. Such a difference was probably not important, however, because the leaf bags were incubated in sheltered locations within pools, and we saw no obvious differences in leaf fragmentation among sites.

Although microbial respiration was similar at all sites at 70 d and thereafter, ATP levels and thymidine uptake on leaves at CO remained high throughout the latter part of the study, showing higher microbial biomass and bacterial growth at this site. Hence, it is possible that the laboratory measurements of respiration rate did not reflect total microbial respiration in situ. It is possible that the laboratory respiration measurements were low because of nutrient limitation in the 5-hr batch-type incubations. Nutrient limitation would be most acute in incubations of leaves with high microbial biomass (i.e., in the later sampling periods and particularly at CO). Therefore, it is possible that differences in microbial respiration accounted for most or all of the differences in leaf mass loss among the sites that we observed.

In summary, our results clearly suggest lower rates of leaf mass loss in southeastern high-elevation streams with pH $\leq$5.7 when compared with an otherwise similar stream with a pH of 6.4. The lower rate of leaf mass loss was accompanied by low ATP levels and low rates of bacterial production and total microbial respiration. Although it remains unclear whether lower microbial respiration rates completely account for the lower rates of leaf mass loss at low pH, macroinvertebrate shredder abundance was not lower at low pH and hence, reduced shredder activity was not a cause of the lower rates of leaf mass loss noted in this study.

Acknowledgements

We thank Leigh Ann Ferren and Donna Genung for help in the field and laboratory, and Mary Anna Bogle and Ralph R. Turner for water chemistry data. The manuscript benefited greatly from critical reviews by A. J. Stewart and R.

B. Cook, T. M. Burton, E. F. Benfield, and an anonymous reviewer. We are grateful for the cooperation of personnel of the Great Smoky Mountains National Park. Research sponsored by the Electric Power Research Institute under Contract RP2326-1 with the U.S. Department of Energy, under Contract No. DE-AC05-84OR21400 with Martin Marietta Energy Systems, Inc. This paper is Publication No. 2921, Environmental Sciences Division, Oak Ridge National Laboratory.

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Received: 6 January 1987
Accepted: 16 June 1987