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Cottonwood hybridization affects tannin and nitrogen content of leaf litter and alters decomposition

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Abstract Cottonwoods are dominant riparian trees of the western United States and are known for their propensity to hybridize. We compared the decomposition of leaf litter from two species (*Populus angustifolia* and *P. fremontii*) and their hybrids. Three patterns were found. First, in one terrestrial and two aquatic experiments, decomposition varied twofold among tree types. Second, backcross hybrid leaves decomposed more slowly than those of either parent. Third, the variation in decomposition between F_1 and backcross hybrids was as great as the variation between species. These results show significant differences in decomposition in a low-diversity system, where >80% of the leaf litter comes from just two species and their hybrids. Mechanistically, high concentrations of condensed tannins in leaves appear to inhibit decomposition ($r^2=0.63$). The initial condensed tannin concentration was high in narrowleaf leaves, low or undetectable in Fremont leaves, and intermediate in F_1 hybrid leaves (additive inheritance). Backcross hybrids were high in condensed tannins and were not different from narrowleaf (dominant inheritance). Neither nitrogen (N) concentration nor the ratio of ash-free dry weight to N (a surrogate for carbon:nitrogen ratio) were significantly correlated with decomposition. The N content of leaf material at the end of each year's experiment was inversely correlated with rates of litter mass loss and varied 1.6- to 2.1-fold among tree classes. This result suggests that hybrids and their parental species are used differently by the microbial community.

Key words Condensed tannins · Decomposition · Hybridization · Litter quality · *Populus*

Introduction

In combining the genes of different species, plant hybrid zones often produce substantial genetic variation that can then be used as a tool to understand diverse ecological and evolutionary processes (e.g., Whitham et al. 1999). In some plant groups, hybrids are common (Stebbins 1959; Grant 1971; Stace 1987) and are thought to have played an important evolutionary role (Ellstrand et al. 1996; Rieseberg et al. 1996). For example, throughout North America, hybrid cottonwoods are common to streams where two or more parental cottonwood species cooccur (Eckenwalder 1984), and they form hybrid zones over 100 km in length (Floate et al. 1997). Because cottonwoods are a dominant riparian tree and their hybridization is known to affect diverse taxa and trophic levels of the terrestrial community (e.g., arthropods, birds, fungi; Floate and Whitham 1995; Dickson and Whitham 1996; Waltz and Whitham 1997), we predicted that other important processes such as decomposition and nutrient cycling would also be affected.

A key process shaping stream ecosystems is the input and subsequent breakdown of leaf litter (Kaushik and Hynes 1971; Petersen and Cummins 1974; Suberkropp et al. 1976). Leaf litter can account for 50–99% of the total energy input to streams (Fisher and Likens 1973; Cummins et al. 1989). Recent experimental data show that when terrestrial litter was excluded from a stream, detritivore as well as predator biomass and density were significantly reduced (Wallace et al. 1997).

Microorganisms play an important role in leaf decomposition by breaking down the leaves after initial leaching of soluble components. The most active decomposers and the largest biomass component of the microbial community are the aquatic hyphomycete fungi (Triska 1970; Kaushik and Hynes 1971; Findlay and Arsuffi 1989). Bacteria, oomycetes and some terrestrial fungi are also found on leaves recovered from streams, but little is known about their impact on decomposition or their interactions with other microbes (Bärlocher 1992). Invertebrate detritivores feed on leaf litter only after it has been

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sufficiently “conditioned” by microbes (Triska 1970; Kaushik and Hynes 1971). Thus, microbial conditioning is a key link between litter and the ability of higher trophic levels to utilize this resource.

Leaf decomposition has been intensively studied over the past two decades (see reviews by Webster and Benfield 1986; Boulton and Boon 1991). Both tree species and genera have been examined as factors that influence leaf decomposition rates (e.g., Kaushik and Hynes 1971). However, most work has been conducted in eastern deciduous streams where there is a high diversity of riparian plant species. In contrast, far fewer studies have been conducted in western streams. Most importantly, because cottonwoods commonly hybridize and their litter dominates many streams, we can use these naturally occurring hybrids to examine the effect of genetic variation on decomposition and nutrient cycling.

Plant genetics affect leaf chemistry and may influence decomposition after leaf abscission via secondary compounds that slow leaf decay. Hybrid plants often contain the chemical compounds present in both parents (Rieseberg and Ellstrand 1993). Because the concentration of these compounds, rather than their presence or absence, may be correlated with decomposition, it is important to quantitate how plant hybridization influences chemical concentrations. For example, Orians and Fritz (1995) found that while willow hybrids contained the defensive chemicals of both parent species, the concentrations of both phenolic glycosides and condensed tannins were intermediate in hybrids compared to their parents.

Condensed tannins, a heterogeneous group of carbon-based compounds found in the leaves of many plant species, including *Populus*, retard leaf decomposition in terrestrial systems (Harrison 1971) and have been implicated in slowing decomposition in streams (Stout 1989; Gessner and Chauvet 1994; Campbell and Fuchshuber 1995). These secondary compounds may bind with available proteins in leaves and deter decomposition by making the leaves less degradable to microbial and invertebrate decomposers (Suberkropp et al. 1976).

Another chemical characteristic of leaf litter that can affect its use by aquatic decomposers is nutrient status. Irons et al. (1988) found that caddisfly larvae selectively fed on leaves with higher N concentration compared to those with lower N concentration. Several studies show that decomposers use leaf litter high in N more readily than litter low in N resulting in fast decomposition of high-quality litter (Kaushik and Hynes 1971; Taylor et al. 1989). The C:N ratio has also been used to predict decomposition (e.g., Berg and Ekbohm 1983; Hart et al. 1992).

Although riparian plants are known to have important impacts on their associated streams, no studies have considered how genetic variation among closely related individuals might influence decomposition in aquatic or terrestrial systems. We investigated how natural hybridization in cottonwoods (*Populus fremontii* × *P. angustifolia*) affected subsequent leaf decomposition. Our studies addressed the following questions. (1) What effect does

hybridization have on the decomposition rates of leaves in both aquatic and terrestrial systems? (2) What is the pattern of condensed tannin concentration among abscised leaves of cottonwood hybrids and their parents? (3) If hybridization affects decomposition, what leaf chemical characteristics are associated with these differences? Finally, we present indirect evidence that examines how hybridization may affect processing by aquatic microbial decomposers. By focusing on trait variation among closely related individuals, we eliminate much of the background variation that occurs among different plant genera on which most decomposition studies have been conducted.

Materials and methods

Study site

Decomposition of cottonwood leaves was examined on the Weber River, a third-order stream located near Ogden in Weber County, Utah, USA. The riparian vegetation is dominated by cottonwood trees. Fremont cottonwood (*P. fremontii*) is present at lower elevations (1300–1500 m) and narrowleaf cottonwood (*P. angustifolia*) occurs at higher elevations (1400–2300 m). There is a 13-km overlap zone where the two tree species cooccur and hybridize freely (Whitham 1989). The parental types (Fremont and narrowleaf) cross to produce F_1 hybrids which backcross only to narrowleaf cottonwood (i.e., an example of unidirectional introgression; Keim et al. 1989). All four of these tree types grow together in the overlap zone. Because we wanted to examine decomposition of these four tree types in their natural habitat and this is the only area on the Weber River where they all naturally co-occur, our study took place entirely within this hybrid and overlap zone. Leaf fall begins in October and is completed by late November. Decomposition experiments began shortly after leaf abscission in the autumns of 1994 and 1995, and continued 63 and 84 days into their respective winters.

Sites where leaf pack studies were performed had flow rates that ranged from 0.10 to 0.40 m/s. Temperature ranged from 2 to 8°C in 1994 and from 3.7 to 8.1°C in 1995. The stream pH varied in 1994 from a low of 4.6 to a high of 7.4, and from 8.5 to 9.2 in 1995. Total N for the stream was 570 µg/l and total P was 740 µg/l as measured in 1995.

1994 leaf pack experiment

In the fall of 1994, leaf litter was collected in nets placed around branches of each of the four tree types described above. Leaf morphological traits distinguished the different tree types. Molecular genetic analysis on these trees showed a high level of agreement with the morphological classifications (G.D. Martinsen, unpublished data). To reduce the effect of among-tree variance, we constructed leaf packs in which 3–3.5 g dry weight of abscised leaves from 10 to 15 different genotypes of each tree type were placed into nylon mesh bags (3 mm mesh size). Because microsite can have important effects on leaf decomposition, we blocked for microsite differences by placing packs in the stream in sets of four that included one of each tree type. Each of these blocks was attached to an iron rod that was driven into the stream bed. We randomized the order of leaf packs on the iron rod so that any influence of one leaf type being upstream of another was also random. At the same time, we made ten identical leaf packs of each type, weighed and flash froze them on dry ice for later chemical analysis to examine the initial quality of the leaf litter. These packs also served as standards to relate dry mass to ash-free dry mass.

A total of 160 leaf packs were placed into the stream in two reaches where natural leaf litter accumulation occurred. To quanti-

fy decomposition over time, we randomly removed packs from the stream at 1, 7, 20, and 63 days. We immediately rinsed insects from the packs and then froze the leaves for later chemical analysis. After 1 day, 40 packs (ten blocks with four tree types each) were removed and 15 blocks (60 leaf packs) were removed after 7 days. Due to ice and snow, we were only able to find and retrieve 7 blocks (28 leaf packs) after 20 days and 8 blocks (32 leaf packs) after 63 days.

1995 leaf pack experiment

To replicate the studies from the first year, in 1995 we reexamined the rapidly decomposing Fremont and slowly decomposing narrowleaf and backcross hybrids (intermediate F_1 s were omitted). However, to control for possible site differences, leaves were collected from trees growing in a common garden. The common garden was located on the edge of the hybrid zone and consisted of 5- to 7-year-old trees, established from cuttings of all four tree types. The narrowleaf and Fremont leaf packs were assembled with leaves from eight to ten different genotypes in order to examine leaf decomposition of a population of trees and to control for among-tree variance. The backcross packs were made up of leaves from single trees of known genotypes. These leaves were selected to include high (mean \pm 1 SE=44.8 \pm 2.6 mg/g) and low (28.9 \pm 4.8 mg/g) tannin concentrations relative to one another (t -test, $P=0.023$). Leaf packs were placed in the stream in blocks that included Fremont, narrowleaf, backcross "high" and backcross "low" tannins, so that we were able to compare decomposition among Fremont, narrowleaf and backcross hybrids, and within the backcross hybrids, between high and low tannins.

We investigated decomposition for longer in the second year and sampled less frequently at the beginning of the experiment. We anchored 132 leaf packs to the streambed in two nearby reaches (not the same as the previous year) and removed leaf packs after 14 ($n=10$ blocks, 40 leaf packs), 28 ($n=15$ blocks, 60 leaf packs), and 84 ($n=8$ blocks, 32 leaf packs) days. Leaf packs were treated as described for the 1994 experiment. Hereafter, we refer to the first year of data as the 1994 experiment and the second year of data as the 1995 experiment. We analyzed our decomposition and N dynamic data only within one time period instead of using an overall estimate of decomposition rates because the data were blocked by microsite and we did not have replication of blocks over time. We calculated decay rates (k) using an exponential model (sensu Peterson and Cummins 1974) where $-k=\log_e(\%R/100)/t$ and $\%R$ is the percent of leaf material remaining after time t in order to compare these data to other published studies.

To examine terrestrial leaf litter decomposition of these same tree types, we constructed packs (3 mm mesh) of backcross hybrid, Fremont, and narrowleaf cottonwood leaves in which we pooled the litter from eight to ten genotypes growing in the common garden. We placed the packs underneath trees in the hybrid zone near the stream banks and covered them with some of the leaf material already present. The packs were put out in sets of three, including one pack of each of the above-mentioned tree types ($n=10$ blocks, 30 leaf packs). These packs were retrieved after 84 days of decomposition on the forest floor.

Analytical and laboratory methods

Upon returning to the laboratory, leaves were freeze-dried, weighed, and ground to a homogenous powder using a Wiley mill. Subsamples were analyzed individually for ash-free dry weight (AFDW), N concentration, and condensed tannin concentration. Leaf powder was dried to a constant weight and then ashed to determine AFDW for each leaf pack. Total N was determined on a Technicon Auto Analyzer II using Kjeldahl procedures modified from Isaac and Johnson (1976). Tannins were extracted four times using 70% acetone at 4°C with sonication for 30 min and the extracts were combined. Tannins were assayed biochemically using

the butanol-HCl method (Porter et al. 1986) that employs colorimetry to quantify tannin concentration. Tannins extracted from the actual leaf material were used as standards to estimate concentrations. We extracted tannins from both parental species and ran a subset of assays using both Fremont and narrowleaf cottonwood standards to estimate tannin concentration. We found no significant difference between the two estimates (paired t -test, $t=2.03$, $P=0.18$) and therefore used tannins from the narrowleaf cottonwood throughout the rest of the analyses.

Stream litter composition

To determine the relative contribution of different cottonwood leaf types to the actual litter present in the stream, we sampled the hybrid and narrowleaf zones in the fall of 1995, just after the majority of leaves had fallen. A 30 \times 30 cm square was randomly placed on the streambed in five different locations in the hybrid zone and five different locations in the narrowleaf zone. All of the leaf litter was removed from inside the sampler. Litter was sorted into four categories: Fremont, F_1 hybrid, backcross/narrowleaf cottonwood (it was not possible to distinguish between the litter of these two tree types), and "other" which included all non-cottonwood litter. Each group was dried and then ashed to determine the AFDW for that component. Weights for each group were averaged to estimate the percentage of each leaf type for the two different zones.

Results

Leaf decomposition

Our 1994 experiment showed that litter from the four tree categories decomposed at different rates (Fig. 1A). Fremont leaves decomposed the fastest and backcross hybrid leaves decomposed the slowest. Statistically significant differences among tree types are present after just 7 days (Table 1). At the final time point (63 days), the percentages of leaf mass remaining for backcross hybrids, narrowleaf, and Fremont cottonwoods were significantly different from one another.

After 63 days, the difference in decomposition between hybrid classes (F_1 and backcross) was as great as the difference between cottonwood species. A statistical contrast on the difference between F_1 and backcross hybrids versus the difference between Fremont and narrowleaf cottonwoods after 63 days was not significant at the

Table 1 Differences in decomposition between the leaves of two species of cottonwood and their natural hybrids are present after only 7 days in the stream in 1994 and 14 days in 1995. Results of a one-way blocked ANOVA of the percent leaf mass remaining at each time point in 1994 and 1995 (aquatic), and in 1995 (terrestrial)

	Days	<i>F</i> -ratio	<i>P</i>
1994 aquatic	1	1.57	0.219
	7	10.66	<0.001
	20	23.18	<0.001
	63	15.91	<0.001
1995 aquatic	14	181.2	<0.001
	28	164.9	<0.001
	84	41.6	<0.001
1995 terrestrial	84	6.02	0.013

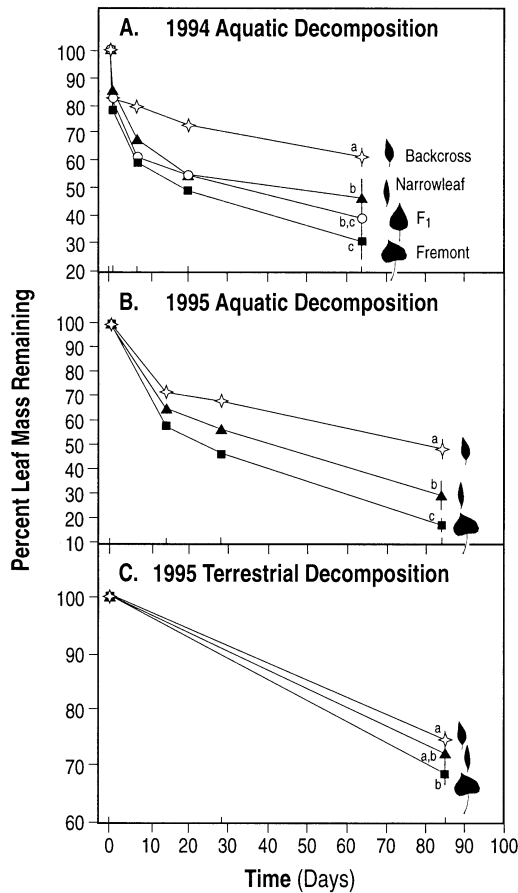


Fig. 1 Leaf pack studies in the Weber River and on the adjacent forest floor, using two cottonwood species and their natural hybrids, showed differences in decomposition. Decomposition was measured as percent leaf material remaining over time (days). In 1994 (A), decomposition was fastest for Fremont cottonwood leaf packs, intermediate for F₁ hybrids and narrowleaf cottonwoods, and slowest for backcross hybrids. In 1995, similar patterns of decomposition are shown for aquatic (B) and terrestrial (C) systems, but the rate of decomposition is about six times faster in the stream than on the forest floor. Lines connecting data points in C are shown only for comparison with A and B and do not imply linear decomposition. Different letters signify significant differences among groups ($P < 0.05$). Leaf silhouettes represent the different leaf morphologies that characterize each tree type and vertical lines represent ± 1 SE around the mean

$P = 0.05$ level, ($F = 0.56$, $P = 0.47$). F₁ hybrid leaf decomposition was intermediate between the two parent species and the mean percentage of leaf mass remaining for F₁ hybrid leaf packs was not significantly different from the mean percentage of leaf mass remaining for the parents (paired t -test, $t = 0.29$, $P = 0.78$). This result is consistent with additive inheritance of traits that control leaf decomposition. In contrast, backcross hybrids had the slowest decomposition rate of all tree types, demonstrating that they possess traits that make their leaves resistant to decomposition.

We observed a 2.4-fold range in decay rates. Calculated decay rates (k) were 0.007 day^{-1} for backcross, 0.012 day^{-1} for narrowleaf, 0.014 day^{-1} for F₁ hybrids, and 0.017 day^{-1} for Fremont cottonwood. Our replicated experiments in

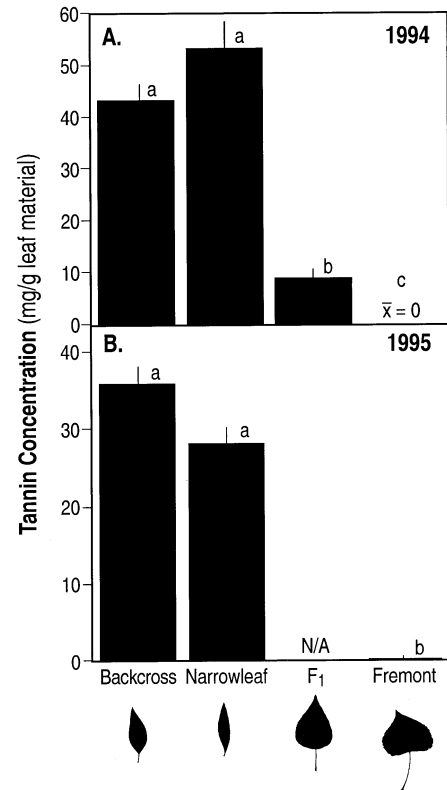


Fig. 2 Condensed tannin concentration in F₁ hybrids exhibits additive inheritance (i.e., intermediate between narrowleaf and Fremont cottonwood parents), and in backcross hybrids exhibits dominance (i.e., not different from narrowleaf cottonwood parent). Initial concentration of condensed tannins in leaf litter as measured by the butanol-HCl method in 1994 (A) and 1995 (B). Analyzed with one-way ANOVA, followed by a Tukey post hoc test for differences between means, $P < 0.05$, $n = 10$ for each tree type. Vertical lines represent ± 1 SE around the mean; different letters signify significant differences among groups

1995 confirmed the 1994 patterns of decomposition (Table 1, Fig. 1A,B). Fremont leaf packs decomposed fastest, narrowleaf were intermediate, and backcross hybrids were slowest to decompose (F₁ hybrids were not included in the repeat experiment). Decay rates for backcross, narrowleaf, and Fremont were 0.009 , 0.014 , and 0.020 day^{-1} , respectively. When using the lines that project decay rates between data points as an approximation, the amount of leaf mass remaining after the same number of days is strikingly similar between years.

The pattern of terrestrial decomposition showed the same pattern as instream decomposition: Fremont litter decomposed the fastest, narrowleaf cottonwoods were intermediate, and backcross hybrids were slowest to decompose (Fig. 1C, Table 1). The leaf mass remaining after 84 days of decomposition on the forest floor was significantly different between Fremont and backcross hybrid leaves. Terrestrial decomposition was clearly slower than aquatic decomposition: for all three leaf types, leaf mass remaining after 84 days on land was similar to that after just 14 days in the stream.

Table 2 Initial concentration of nitrogen, the ratio of ash-free dry weight (AFDW):N (as a surrogate for C:N), and the ratio of tannin to nitrogen concentration (mean \pm 1 SE) in the leaf litter of each tree type for two consecutive years. Results of one-way ANOVA for differences between litter types are included

Tree type	N(mg/g)	AFDW:N	Tannin:N
1994 initial litter quality			
Backcross hybrid ($n=19$)	6.69 \pm 0.10	149.79 \pm 2.21	6.79 \pm 0.98
Narrowleaf cottonwood ($n=10$)	7.61 \pm 0.13	131.73 \pm 2.21	7.00 \pm 1.43
F ₁ hybrid ($n=10$)	8.17 \pm 0.21	123.02 \pm 3.09	1.23 \pm 0.32
Fremont cottonwood ($n=10$)	7.20 \pm 0.13	139.21 \pm 2.54	0.0000
ANOVA	$F=19.4$ $P<0.0001$	$F=16.8$ $P<0.0001$	
1995 initial litter quality			
Backcross hybrid ($n=5$)	7.23 \pm 0.48	142.93 \pm 8.64	4.96 \pm 0.53
Narrowleaf cottonwood ($n=5$)	7.70 \pm 0.42	131.44 \pm 6.66	3.74 \pm 0.70
Fremont cottonwood ($n=5$)	8.15 \pm 0.36	123.53 \pm 5.03	0.036 \pm 0.02
ANOVA	$F=0.996$ $P=0.391$	$F=1.52$ $P=0.248$	

Table 3 Condensed tannin concentrations and the ratios of tannin to nitrogen concentration are the best predictors of decomposition. Having found no effect of year on decomposition, in this analysis we combined the data from both years for a total of seven groups (Fremont, narrowleaf, F₁, and backcross hybrids in 1994; Fremont, narrowleaf, and backcross hybrids in 1995). Coefficients of determination (r^2), P -values, intercepts, and slopes relating percentage original leaf mass remaining to initial litter quality for the leaf tissues are presented

Litter quality variable	Slope	Intercept	P	r^2
Nitrogen (mg/g)	-19.30	185.86	0.075	0.50
Tannin (mg/g)	5.61	24.91	0.032	0.63
AFDW:N	1.07	-103.32	0.067	0.52
Tannin/nitrogen	3.89	26.63	0.029	0.65

Patterns of condensed tannin concentration

In 1994, the condensed tannin concentration was high for backcross hybrid and narrowleaf leaves, undetectable for Fremont, while F₁ hybrid leaves were intermediate (Fig. 2A) (ANOVA: $F=18.64$, $p<0.0001$). A Tukey post hoc test for differences among means revealed that tannin concentrations were different for all tree types except between narrowleaf and backcross hybrid leaves.

Replicate studies in 1995 revealed a similar pattern of condensed tannin concentration (Fig. 2B). Backcross hybrid mean tannin concentration was not statistically different from narrowleaf mean tannin concentration and both were high. Fremont leaves had an extremely low concentration of condensed tannins. However, there were differences in average tannin concentration between the two years for all of the tree types, especially narrowleaf and backcross hybrids. This may have been caused by site differences (leaf material in the second year was collected in the common garden) or may simply represent year-to-year variation in leaf chemistry.

Litter quality as predictor of leaf decomposition: condensed tannins and nutrient status

In addition to tannin concentration, we also investigated N concentrations and AFDW:N ratios as a surrogate

for C:N ratios (sensu Vitousek 1982) to characterize initial litter quality (Table 2). Neither N nor AFDW:N ratios were significantly correlated with decomposition (Table 3) nor were the differences in these two measurements among leaf types very large (Table 2). This contrasts with other studies in which leaves with high initial N concentration decompose more rapidly than those with low initial N concentration (Kaushik and Hynes 1971; Taylor et al. 1989). However, there have been reports of N concentration having little predictive value in other systems (Melillo et al. 1982; Gessner and Chauvet 1994). Also, the range of N concentrations does not vary much among leaf types and this may explain why there is no correlation with decomposition.

Tannin concentration accounted for 63% of the variability in decomposition among the four leaf types in a regression analysis (Table 3). However, we found no difference in decomposition when comparing backcross leaves that were "high" versus "low" in condensed tannin concentration (paired t -test, $t=-0.81$, $P=0.44$). After 84 days, the backcross leaf packs with "high" condensed tannins had a mean \pm SE percentage leaf mass remaining of 46.8 \pm 3.5% and the packs that had "low" condensed tannins had 49.6 \pm 3.5% leaf mass remaining. Because tannin concentration in these leaves ranged between 28.9 and 44.8 mg/g, it is possible that these concentrations are above some threshold at which tannin concentration no longer determines decomposition.

We also measured phenolic glycosides in a subsample of abscised leaves using high-performance liquid chromatography. We found no phenolic glycosides in these leaves. This was expected because these are low-molecular-weight, labile compounds, and are likely translocated back into the tree prior to abscission.

Litter nitrogen content as an indicator of microbial colonization

All leaf types had different patterns in the percentage of leaf pack N remaining over time, indicating different use by microbes (Fig. 3A,B). For example, in 1994, back-

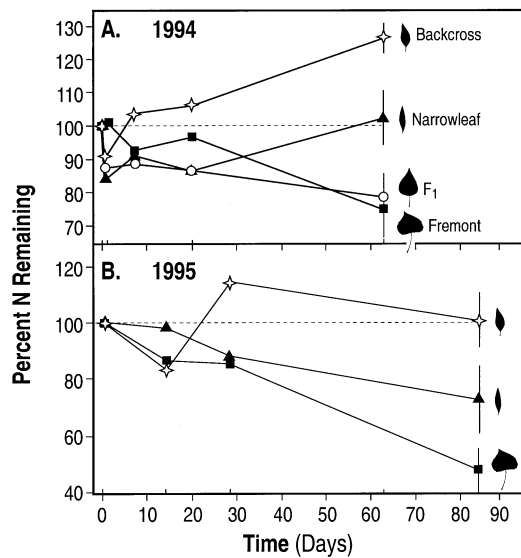


Fig. 3 Two cottonwood species and their natural hybrids showed different N dynamics during decomposition. In 1994 (A), all leaf types showed microbial colonization, as measured by an increase in the %N remaining in leaf material; however, the increase occurred at a different time and a different magnitude for each leaf type. In 1995 (B), backcross hybrid leaves showed the sharpest increase in %N remaining. Different letters signify significant differences among groups ($P < 0.05$). Vertical lines represent ± 1 SE around the mean

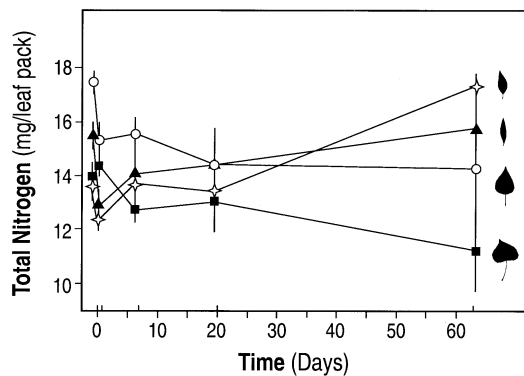


Fig. 4 Total leaf pack N observed for Fremont and narrowleaf cottonwood, F_1 , and backcross hybrids in the hybrid zone shows that a mixture of leaf types results in dispersed N over time. Vertical lines represent ± 1 SE around the mean

Table 4 There are more types of leaf litter in the hybrid zone compared to the pure narrowleaf zone, and cottonwood leaf litter is the dominant litter in both zones. The percentage by weight (mean ± 1 SE) of each tree type of the total litter present in the hybrid zone is compared to the pure narrowleaf zone. Means are based on five samples collected in each zone

Litter type	Hybrid zone (%)	Pure narrowleaf zone (%)
Narrowleaf/backcross	33.15 \pm 3.61	92.40 \pm 0.92
F_1 hybrid	3.00 \pm 1.49	1.16 \pm 0.71
Fremont cottonwood	44.59 \pm 7.73	0
Other	16.05 \pm 5.40	6.44 \pm 1.15

cross hybrid leaf litter showed net N immobilization, while the other tree types generally showed net N release over the course of the experiment. Thus, in addition to the different decomposition rates (Fig. 1), these different N dynamics in leaf litter argue that these leaves were being used differently by the decomposer community. We set the N content determined at time zero equal to 100%. An increase in the N content of leaf litter (i.e., values above 100%) indicates that microbes are colonizing the leaves and immobilizing N from the stream water. N may increase solely because of the numbers of microbes accumulating over time or because of the difference in microbial activity on different leaf types.

Litter composition and nitrogen dynamics in the hybrid versus pure riparian zones

Comparisons of stream litter from the pure narrowleaf and hybrid riparian zones showed that cottonwoods made up the majority of leaf litter in both zones and that the contributions of each tree type varied between the two zones (Table 4). Approximately 92% of all leaf litter in the narrowleaf zone came from narrowleaf cottonwood. However, in the hybrid zone, backcross hybrid, narrowleaf, and Fremont predominated, and F_1 hybrids contributed only a small amount to total leaf litter.

The differences in decomposition and N dynamics across pure and hybrid sites could have important effects on dependent stream biota. For example, at each of the times we measured leaf pack N in 1994, the variation in total N was about four times greater among a mix of leaf types than within any one leaf type alone (Fig. 4). Analyses of our 1995 data showed similar patterns. These results suggest that N dynamics in the 13-km hybrid zone of the Weber River could be very different than in adjacent pure zones.

Discussion

Genetic variation in decomposition rates and tannins

In quantifying herbivore and parasite responses to hybrid plants relative to their parent species, Fritz et al. (1994) described hybrid resistance traits as being additive, dominant, susceptible or resistant. The same characteristics that influence resistance to leaf herbivores and pathogens (e.g., secondary chemistry and N concentration) can also affect the resistance of leaf litter to decomposition, although no adaptive function is implied. When examining decomposition, we found additive genetic effects in F_1 hybrids where their decomposition was intermediate and not significantly different from the mean response of the two parent species. However, backcross hybrids showed resistance to decomposition: they decomposed much slower than either parent. According to Peterson and Cummins' (1974) processing continuum, our calculated decay rates (k) place backcross hybrids in the medium-

decay group and narrowleaf, F_1 hybrids, and Fremont in the fast-decay group. Although genetic resistance to pests and pathogens in natural hybrids is relatively rare (5% of 117 cases: Strauss 1994; 3% of 35 additional cases: Whitham et al. 1999), our study shows resistance to decomposition in backcross hybrids across 2 years, and in both aquatic and terrestrial systems. These differences also emphasize that not all hybrids are alike. Had we pooled F_1 and backcross hybrids, we would have erroneously concluded that hybrids exhibited dominant genetic effects on decomposition, when in fact they are additive and resistant, respectively.

F_1 and backcross hybrids differed with respect to condensed tannin concentration as well as decomposition rate. F_1 s exhibited additive concentrations of condensed tannins, whereas backcross hybrids were dominant. Our findings with abscised leaves parallel those of living leaves in willows (Orians and Fritz 1995) in which *Salix eriocephala* had high levels of condensed tannins, *S. sericea* had low levels, and their hybrids had intermediate, but highly variable concentrations. Because they pooled all hybrids in their analyses, the variation in the concentration of secondary chemicals between backcross and F_1 hybrids could account for the high degree of variability within hybrids.

Previous studies have shown differences in leaf decomposition and the associated stream biota of distantly related taxa in eastern streams (Kaushik and Hynes 1971; Petersen and Cummins 1974; Golladay et al. 1983). Studies of decomposition in western streams where riparian tree species are often closely related (e.g., cottonwoods and willows) may warrant a different approach. The twofold differences in decomposition between cottonwood species and their hybrids suggests that analyses at finer taxonomic levels are important. Although the responses of stream detritivores to this variation are presently unknown, numerous terrestrial herbivores exhibit pronounced responses to the variation between hybrid classes that greatly exceed the variation between parental species. For example, Whitham et al. (1994) showed that the scale insect, *Eriococcus tepperi*, was 87 times more abundant on one hybrid class than another, but at the same time showed no significant differences in abundance on the parental species. Furthermore, Floate and Whitham's (1995) analyses of 15 common insect species showed that the distinct insect communities of Fremont cottonwood, F_1 , and backcross hybrids were just as useful in identifying tree types as were traditional plant morphological traits.

Implications for the decomposer community and nutrient cycling

Variation in decomposition rates has the potential to influence the ecology of the biotic community in two major ways. First, leaves that decompose rapidly are likely to be exploited by mobile consumers that move from one resource patch to another as resources are depleted. Due

to their persistence, slowly processed leaves may be used for shelter by invertebrates and as substrate by microbes. The fact that N dynamics differs among pure and hybrid types suggests that these leaf types may be used differently by the microbial community. This community can be diverse: a single decomposing leaf can support 5–12 species of aquatic hyphomycetes (Ingold 1942). As many as 7 species of fungi have been found on 1 cm² of a skeletonized leaf (Greathead 1961) and colonies of different species may overlap extensively (Chamier et al. 1984).

Second, these differences in decomposition rates could lead to more diverse decomposer and shredder communities in the hybrid compared to the narrowleaf zone. At higher elevations where ~92% of the stream leaf litter is derived from narrowleaf cottonwood, all litter will be processed synchronously. However, in the hybrid zone where multiple cottonwood litter types are present, their differential decomposition should temporally disperse resource availability in the stream. We predict that dispersed decomposition in hybrid zones should provide a wider dietary continuum and support greater microbial and invertebrate diversity than in pure zones where similar litter will result in a single pulse of decomposition.

Our predictions about pure and hybrid zones may be analogous to decomposition studies of monospecific versus polyspecific leaf packs. Leff and McArthur (1989) found increased invertebrate biomass on mixed-species leaf packs relative to single-species packs. Similarly, we expect that leaf packs in a pure stand of narrowleaf or Fremont cottonwoods would support less invertebrate biomass than leaf packs in a hybrid zone composed of both species and all hybrid classes. Because dominance patterns of fungi are altered by changes in the plant species composition of leaf packs (Suberkropp and Klug 1976), we suggest that the variation in cottonwood leaf types available in the hybrid zone may have similar impacts on the microbial community.

The above effects of plant hybridization on aquatic communities may also extend to the decomposer community in terrestrial systems. Findlay et al. (1996) argued that studies of aquatic decomposition could be used to model what happens to litter in terrestrial systems. If true, then the differences we have experimentally quantified may be important in terrestrial N cycling. For example, slow decomposition allows more litter to accumulate and slows the return of nutrients via mineralization, whereas fast decomposition may have the opposite effect. Therefore, terrestrial nutrient release from litter may be less temporally variable in the hybrid zone, whereas in the pure zones, nutrients may be released synchronously.

Predictors of decomposition rates

In our analyses of two cottonwood species from different sections of the genus *Populus* (Eckenwalder 1984) and

their hybrids, condensed tannin concentrations explained about 63% of the variation in decomposition rates. Other studies have found similar relationships between decomposition and tannin concentration. When species high in lignin were removed from their analysis, Gessner and Chauvet (1994) found a significant negative correlation between condensed tannin concentration and decomposition rates of seven species. Among *Eucalyptus* species in an Australian stream, Campbell and Fuchshuber (1995) found that condensed tannin concentration influenced decomposition rates, but only when seven other genera of riparian plants were excluded. Thus, condensed tannin levels predicted decomposition, but only among closely related species.

Our attempts to incorporate other predictors of decomposition into a more comprehensive model were not successful. Neither N alone nor a surrogate of C:N were significantly correlated with decomposition. There are at least three reasons why N concentration may not predict decomposition. First, other factors (e.g., lignin, cuticle thickness) may interact with N such that N concentration alone does not predict decomposition. Second, microbes can make use of N in both the leaf litter and the water, so they do not have to rely strictly on the N present in leaf material in order to colonize and decompose the litter. Third, although there were significant differences in litter N concentration, the range was small. Other studies have shown as little as a twofold (Mellilo et al. 1982) and as large as a fourfold (Gessner and Chauvet 1994) difference in N concentration among six to seven species, although N did not necessarily correlate with decomposition.

It appears that there may be a threshold beyond which tannin concentration is no longer important and nutrient status or some other factor (e.g., lignin) controls the rate of leaf decomposition. While tannin concentration was a good predictor for three of the tree types we studied, it was not a good predictor of decomposition within the backcross hybrid leaves. When we separated backcross hybrids into individual trees high and low in tannins, we found no difference in their decomposition, suggesting that all were above the threshold in which further increases in tannins had no effect on decomposition.

In light of the rapid demise of riparian habitat throughout the United States (Noss et al. 1995), it is important to understand the basic processes involved in their functioning as well as their restoration. As poplars represent a major source of stream litter in the west (e.g., up to 92% in our study), they must also represent a major source of nutrients through decomposition. Because natural hybrid zones are found in nearly all western streams (Eckenwalder 1984) and vary in size from ~5 to 100 km (Floate et al. 1997), the processes we describe are likely to be widespread. Furthermore, as hybrid poplars are widely used in commercial plantations (Stettler et al. 1996), their differing decomposition rates could have unforeseen impacts, both positive and negative, on dependent community members.

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