

Litter Decomposition of *Picea orientalis*, *Pinus sylvestris* and *Castanea sativa* Trees Grown in Artvin in Relation to Their Initial Litter Quality Variables

Temel SARIYILDIZ*

Kafkas Üniversitesi, Artvin Orman Fakültesi, 08000, Artvin - TURKEY

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Abstract: The aim of the present study was to determine the effects of the litter chemistry of spruce (*Picea orientalis*), pine (*Pinus sylvestris*) and chestnut (*Castanea sativa*) grown in the Artvin region on their decomposition rates. Freshly fallen litters from these species were analysed for total carbon, nitrogen, lignin, cellulose and hemicellulose. The litterbag technique was used to determine their mass loss in the field for 2 years. At intervals of 6, 12, 18 and 24 months, 5 litterbags of each species were collected and returned to the laboratory and then analysed for their mass losses. The results showed that differences in mass losses for the 3 tree species were significant for all sampling intervals. Final mean mass losses were 35.9% for spruce, 51.1% for pine and 64.5% for chestnut (increments of 27.0%, 34.7% and 38.6% compared to the initial mass losses at 6 months respectively). Initial lignin concentration was the best predictor ($r^2 = 0.97$) for mass losses from litter species, indicating the relative importance of the initial lignin concentration of these 3 species in affecting their decomposition rates and hence the nutrient cycling process in this forest ecosystem.

Key Words: Decomposition, forest ecosystem, litter quality, lignin, Artvin

Artvin'de Yetişen *Picea orientalis*, *Pinus sylvestris* ve *Castanea sativa* Türlerinin Yapraklarının Ayrışmasında Kimyasal Bileşimlerinin Etkisi

Özet: Bu çalışmada, Artvin'de yetişen ladin (*Picea orientalis*), sarıçam (*Pinus sylvestris*) ve kestane (*Castanea sativa*) yapraklarının kimyasal bileşimlerinin ayrışma oranları üzerine olan etkisi belirlenmeye çalışılmıştır. Bu amaçla, ilk önce orman yüzeyine düşmüş olan o yıla ait yapraklar araziye gidilerek toplanmış ve içerdikleri toplam karbon, azot, lignin, selüloz and hemiselüloz miktarları belirlenmiştir. Daha sonra yaprakların ayrışma oranlarını belirlemek için her bir türden belli miktardaki yaprak örnekleri daha önceden hazırlanmış naylondan paketler içine (20 x 20 cm genişliğinde ve 1 mm² den az ağ gözüne sahip) yerleştirilerek tekrar araziye konulmuştur. İki yıllık süre içinde her altı ayda bir olmak üzere her bir tür için beşer tane örnek araziden alınarak ayrışma oranları belirlenmiştir. Sonuçlar incelendiğinde, yapılan her örnekleme zamanında (6., 12., 18. ve 24. aylar sonunda) bu üç ayrışma oranları arasında önemli derecede farklılıklar olduğu görülmüştür. İlk altı ay sonundaki ayrışma yüzdesi, ladin için % 8.87, sarıçam için % 16.4 ve kestane için % 25.9 olarak bulunmuştur. Bundan sonraki örneklemeelerde de kestanenin daha hızlı bir kütle kaybı gösterdiği bunu sarıçamın ve ladinin izlediği görülmüştür. Yirmi dördüncü ayın sonundaki ayrışma yüzdesi ladin için % 35.9, sarıçam için % 51.1 ve kestane için % 64.5 olarak bulunmuştur. Bu türlerin yapraklarının içerdikleri kimyasal bileşimler ile ayrışma oranları arasındaki korelasyonlara bakıldığında, bu üç türün içerdikleri lignin miktarlarının ($r^2 = 0.97$) onların kütle kayıplarını etkileyen en önemli kimyasal bileşim olduğu bulunmuştur. Bu türlerin kimyasal bileşimleri ve ayrışma oranlarındaki farklılıklar bu türlerden oluşan saf yada karışık orman ekosistemlerindeki besin döngüsü süreçlerinde birbirinden farklı olacağını ve bu ortamda gelişen türlerin besin elementlerinden yararlanmasını önemli derecede etkileyebileceğini göstermektedir.

Anahtar Sözcükler: Ayrışma, orman ekosistemi, yaprakların kimyasal bileşenleri, lignin, Artvin

Introduction

In forest ecosystems, litter fall is an important flux of organic matter into soil subsystems and the rate of litter decomposition is influenced by 3 main factors: the physico-chemical environment, the decomposer

population and the resource quality of decomposing material (Swift et al., 1979). However, consistent differences in decay rate persist between litters of different plant species, regardless of environmental conditions, and so litter quality is a primary determinant

* Correspondence to: t_sariyildiz@yahoo.com

of decay rate (Heal et al., 1997; Sariyildiz, 2000). The chemical constituents of litter include concentrations of various classes of organic compounds and mineral elements in relation to plant characteristics and availability. The bulk of litter comprises structural components of plant cell walls (cellulose, lignin and hemicellulose) and hence carbon is always in much larger concentrations than nutrients (Johansson, 1995). In addition to structural polymers, litter also contains water-soluble fractions, such as simple sugars and amino acids, oils, waxes, simple and complex phenolic compounds, cutins and so forth (Alexander, 1977; Swift et al., 1979). Many researchers have demonstrated relationships between these initial litter quality characteristics and decomposition rates for a large number of plant species (e.g., Meentemeyer, 1978; Berg and Staaf, 1980; Sariyildiz and Anderson, 2003a). The nitrogen concentration of the litter or the C:N ratio has been identified as an important controlling factor in decomposition processes (e.g., Anderson, 1973; Berg and Ekbohm, 1983; Taylor et al., 1989). However, other studies have emphasised the importance of the initial lignin concentration in litter decomposition processes (e.g., Berg and Tamm, 1991; Entry et al., 1995; Sariyildiz and Anderson, 2003b), the summed concentrations of lignin and cellulose (Aber et al., 1990), the lignin -N-ratio (Aber and Melillo, 1991; Melillo et al., 1982), the lignin-cellulose index (lignin to (lignin + cellulose); LCL), and the holocellulose to lignicellulose quotient (HLQ) ratios (McClougherty and Berg, 1987). A number of researchers have stated that the influence of initial N and microbial degradation of the holocellulose fraction in the early phase of decomposition is strongly dependent upon lignin concentrations in the litter (e.g., Fogel and Cromack, 1977; Berg and Staaf, 1980) because of their intimate physical association and covalent bonding in the cell wall (Monties, 1994).

Although many studies have been carried out to investigate the effects of litter quality variables on decomposition using different species, it is still not known which litter quality variable is the best indicator of decomposition. Hence, it is important to provide more data from a wide range of environments and so better understand the decomposition processes in a region's forests. The aims of the present paper were therefore firstly to determine the litter quality variables in *Picea orientalis*, *Pinus sylvestris* and *Castanea sativa* tree

species in the Artvin region, and secondly to investigate the effects of these variables on the decomposition rates of these 3 species. To our knowledge, in the literature there is no study on these tree species related to their litter quality and decomposition rates in this region.

Materials and Methods

Sampling site, litter collection, chemical analysis and field incubation

The sampling forest was at high elevations (between 1250 and 1600 m) between the Cerat Tepe and Kafkasör areas, south-west Artvin, Turkey (41°50' N, 41°06' E). The dominant tree in the forest was spruce, followed by pine and chestnut. The trees were approximately 50-70 years old and 25-30 m high. The canopy cover was moderate and the understory was poor in species. The climate in the Artvin region is generally characterised by cold winters and semi-arid summers. A summary of the weather data for 1948-1997 (Artvin Meteorology Station, at 597 m) indicates that precipitation averages 689.4 mm annually, with the highest amounts in January (99.7 mm), and the lowest in August (27.1 mm). Average monthly temperature ranges from 32 °C in August to -2.5 °C in January. However, at upper elevations, average annual precipitation can reach over 1000 mm and mean temperature can drop as low as -16.1 °C. Annual relative humidity is 65%, with the highest amounts in June (71%) and the lowest in March (62%). A shallow sandy loam soil covers the granite bedrock. The soil profile shows distinct A and C horizons; the mineral B horizon is almost absent. The organic soil layer is 5-6 cm thick with a litter layer of 2-3 cm and humus layer of 1-3 cm.

Freshly fallen litter was collected in late September 2000 from ten, 1 m quadrats located at random on the forest floor. The main period of litter fall in this area is short in duration, reaching a peak at the time of sampling. The weather was cold when the litter material was collected and it showed no visible signs of discoloration or fungal colonisation.

All litters were oven dried at 85 °C and then ground in a laboratory mill to a mesh fraction less than 1 mm and then the initial chemical composition of these litters was determined. Organic carbon was analysed using a Leco HF10 gravimetric carbon analyser (Leco Corporation, St. Joseph, USA). Total nitrogen was determined by Kjeldal

digestion (Allen, 1989) followed by analysis of NH_4^+ by the indophenol method using an auto-analyser (Bemas, Burkhard Ltd, Uxbridge, UK). Cellulose and lignin were determined using the ADF-sulphuric lignin method of Rowland and Roberts (1994). The component sugars of structural polysaccharides (mainly hemicelluloses) were hydrolysed using 4 M trifluoroacetic acid (TFA) and prepared for GC analysis according to the method of Guggenberger and Zech (1994). All analyses were carried out in triplicate.

The decomposition of spruce, pine and chestnut litters was studied using the litterbag technique (Swift et al., 1979). The litterbag was 20 x 20 cm with a mesh size of less than 1 mm². Two grams of samples were included in each litterbag. All bags were numbered and fixed to the ground with metal pegs. Five of these bags (randomly selected) in each site were collected every 6 months for 2 years in order to see the continuum of the decay rates of

litters over time. The litters were oven dried at 85 °C until constant weight. Mean weight loss was then determined.

Variability in litter quality between the 3 species was determined using the Tukey method of multiple pairwise comparisons at $\alpha = 0.05$ using SPSS 9.0 for Windows. Differences in mass losses between the 3 species at each sampling time were tested for significance using analysis of variance, and relationships between litter quality and mass losses were determined using linear regression with MS EXCEL 2000.

Results

Variation in litter chemistry

Initial concentrations of carbon nitrogen, lignin, cellulose and hemicellulose in spruce, pine and chestnut litters are shown in Table 1. Chestnut litter had a higher

Table 1. Resource quality characteristics of spruce (*Picea orientalis*), pine (*Pinus sylvestris*) and chestnut (*Castanea sativa*). All ANOVAs were significant at $P < 0.01$. The Tukey method of multiple pairwise comparison at $\alpha = 0.05$ used to determine significantly different means. Means with the same letter are not significantly different by columns ($n = 3$).

		Carbon (%)	Nitrogen (%)	C:N	Lignin (%)	Cellulos. (%)	Hemicell. (%)	Lignin: N
Spruce	Mean	46.4 ^a	1.16 ^a	40.0 ^a :1	39.9 ^a	25.0 ^a	26.0 ^a	34.4 ^a :1
	Std. Err.	0.05	0.15	0.92	0.44	0.72	0.32	1.43
	Max.	47.3	1.20	41.7	41.3	25.9	27.1	36.9
	Min.	45.2	1.12	38.6	38.3	24.1	24.5	31.9
	Range	2.11	0.08	3.07	3.00	1.81	2.60	4.96
	Std. Dev.	1.08	0.04	1.59	1.58	0.90	1.33	2.48
	Coeff. Var.	1.17	0.02	2.54	2.30	0.83	1.77	6.15
Pine	Mean	46.2 ^a	1.31 ^b	35.3 ^b :1	29.3 ^b	31.6 ^b	28.6 ^b	20.1 ^b :1
	Std. Err.	0.49	0.34	1.07	0.63	0.91	0.21	0.72
	Max.	47.0	1.36	37.0	30.2	33.4	29.2	23.8
	Min.	45.3	1.27	33.3	28.4	29.9	28.3	21.5
	Range	1.70	0.09	3.70	1.80	3.50	0.90	2.24
	Std. Dev.	0.86	0.05	1.86	0.90	1.75	0.49	1.25
	Coeff. Var.	0.74	0.02	3.45	0.81	3.06	0.24	1.58
Castanea	Mean	51.3 ^b	1.35 ^b	38.0 ^c :1	21.1 ^c	31.7 ^b	32.4 ^c	15.6 ^c :1
	Std. Error	0.42	0.42	1.29	0.66	0.46	0.86	0.79
	Max.	53.4	1.38	40.5	22.6	33.6	33.6	16.6
	Min.	49.9	1.32	36.2	19.4	28.9	31.1	14.1
	Range	3.50	0.06	4.29	3.20	4.70	2.50	2.56
	Std. Dev.	1.85	0.03	2.24	1.85	2.48	1.25	1.38
	Coeff. Var.	3.43	0.01	5.00	3.43	6.13	1.56	1.89

carbon concentration than spruce and pine litter, which showed similar carbon concentrations. Chestnut and pine litters had higher nitrogen concentrations than spruce litter. However, the C-to-N ratio in spruce litter was higher than that in pine and chestnut litters. Spruce litter showed the highest lignin concentration, whereas it showed the lowest cellulose and hemicellulose concentrations. Spruce litter also had the highest lignin-to-N ratio, followed by pine and chestnut litters.

Litter mass losses

Figure 1 shows mass remaining of the 3 tree species sampled over 24 months. The differences in mass losses were significant ($P < 0.01$) between the species for all sampling intervals.

After 6 months mean mass losses were 8.87% for spruce, 16.4% for pine and 25.9% for chestnut. These differences in mass losses between the 3 tree species continued from 6 to 24 months. Final mean mass losses were 35.9% for spruce, 51.1% for pine and 64.5% for chestnut (increments of 27.0%, 34.7% and 38.6% respectively).

Relationship between mass losses and litter quality

Mass losses were plotted against litter quality variables from spruce, pine and chestnut litters. These relationships are illustrated for lignin in Figure 2. The concentrations effectively formed a continuum across species and litter samples. Similar trends were found for

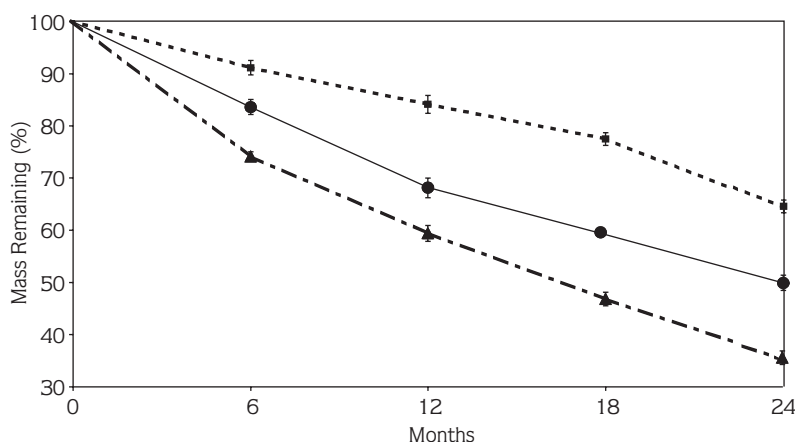


Figure 1. Mean mass remaining (\pm SE) of *Picea orientalis* (■), *Pinus sylvestris* (●) and *Castanea sativa* (▲) litters after 6, 12, 18 and 24 months in the field.

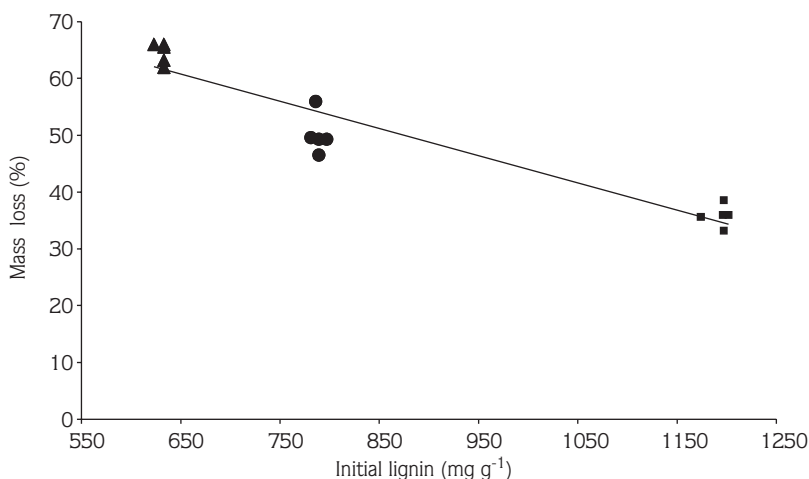


Figure 2. Regression ($P < 0.01$, $n = 15$) of percentage mass loss after 24 months' decay on initial lignin concentrations from chestnut (*Castanea sativa*), pine (*Pinus sylvestris*) and spruce (*Picea orientalis*) litters.

other litter quality attributes, and goodness of fit statistics for the regression of mass losses on litter quality variables are shown in Table 2.

Mass losses from spruce, pine and chestnut after 24 months showed the highest negative correlation with initial lignin ($r^2 = 0.97$). Lignin-to-N ratio was the second best predictor of mass losses ($r^2 = 0.95$). Nitrogen and hemicellulose concentrations explained less variance ($r^2 = 0.93$ and 0.94 respectively) than lignin and lignin-to-N ratio, but they showed a positive correlation with mass losses.

Discussion

Many studies (e.g., Berg et al., 1995; De Santo et al., 1993) have adapted the litterbag approach in the field to determine the effect of litter quality on decomposition rates. In these studies they have stated that rates of litter decomposition are influenced by a hierarchy of interacting physical, chemical and biotic factors. Climate sets the general limits of the litter decomposition process through physiological constraints on the activity of organisms, while the chemical quality of the resource then defines the rates at which organisms can operate within these constraints (Couteaux et al., 1995). The general conclusion has been that the physical climate dominates rates of decay on board regional scales, whereas litter quality is of more importance in determining decay rates at small scales (i.e. within site).

In small scale sites, it has been found that when the lignin concentration in a given litter increases above about 20% it can dominate litter decomposition rates irrespective of other constituents. Below this level the C:N ratio, polyphenol:N ratio and simple N concentrations may be indicative of decomposition potential depending on which constituents limit microbial activities (Entry et al., 1995; Heal et al., 1997).

The present study was carried out in a small scale site and mean lignin concentrations in the 3 tree litters were 21.1% for chestnut, 26.1% for pine and 39.9% for spruce. Hence, regression of the initial litter quality variables with observed decay rates indicated the relative importance of initial lignin concentration in affecting decay rates all sampling intervals (Table 2).

In decomposing litter the chemical component may be degraded in a certain sequence reflecting a succession of micro-organisms with different saprotrophic abilities (Minderman, 1968; Berg and Staff, 1980; Berg et al., 1982; McClaugherty and Berg, 1987). This means that the heterogeneous group of micro-organisms that invades the litter initially decomposes the water solubles in the non-matrixed part of the cellulose and hemicelluloses (as energy sources), and finally the complex of interwoven holocellulose and lignin is attacked by cellulolytic and lignolytic fungi (Frankland, 1992; Cox et al., 2001). However, in the present study, the results indicated that the relative proportions of these fractions

Table 2. Goodness fit for linear regression of mass losses at 6, 12, 18 and 24 months against the litter quality variables for spruce, pine and chestnut litters. Correlation coefficients for the regression were significant ($P < 0.01$, $F = 13$).

Litter quality variables	6	12	18	24 months	correlation
	r^2	r^2	r^2	r^2	
N	0.80	0.81	0.92	0.93	+
C	0.52	0.47	0.59	0.69	+
Lignin	0.84	0.86	0.95	0.97	-
Cellulose	0.66	0.74	0.82	0.86	-
Hemicellulose	0.82	0.83	0.92	0.93	+
C:N	0.34	0.43	0.43	0.50	-
Lignin:N	0.83	0.84	0.93	0.95	-

in the cell wall may affect the response of the substrate degraded by micro-organisms. For chestnut, the cell wall contained higher concentrations of hemicellulose (sugars), whereas lignins in the cell wall were lower in concentration compared to those in spruce, which had higher lignin and lower hemicelluloses. The low lignin concentration means that its retarding influence was much lower than for the high lignin concentrations, which strongly retarded the degradation of cellulose or hemicellulose by micro-organisms (Sariyildiz and Anderson, 2003b). In the cell wall of chestnut the hemicelluloses may have been partly integrated with lignin and partly free. Therefore, the hemicelluloses not matrix-bound to lignin represented a more available and easily hydrolysed substrate to micro-organisms compared with spruce litter. This part of the cell wall in chestnut was attacked by micro-organisms and responded in different ways compared with the fraction in which lignin regulated the mass loss in spruce litter, and hence initially resulted in different mass loss rates between these 2 trees. Cell walls in pine needles contained intermediate concentrations of hemicellulose and lignin, which were reflected by intermediate rates of mass loss between chestnut and spruce.

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