

## Leaf litter decomposition of selected urban tree species during mulching

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### Introduction

The arrangement of trees and shrubs along city streets and parks has been a common practice. It has been estimated that about 36% of the urbanized area of the Tokyo metropolitan is covered with trees or other types of urban vegetation ([http://www.sangyo-rofo.metro.tokyo.jp/norin/aramasi/ara-ringyo/ara\\_4.htm](http://www.sangyo-rofo.metro.tokyo.jp/norin/aramasi/ara-ringyo/ara_4.htm)). The area of Tokyo urban forest is 78,698 ha and it is increasing continuously with the urbanization (TOYOHARA *et al.*, 2001). But urban forests generate significant amounts of litters because the maintenance activities are essential in urban area. Recycling of solid wastes from the urban forest would be important from the view point of environment such as global warming, urban heat islands, and air pollution in recent years, and would be free from the pressure of the overcrowded municipal landfills and improve the urban environment. The application of waste as landscape mulching is one of the best way to recycle these materials. Studies of the application of plant residues to woodland in urban area were focused mainly on the effect of soil quality (FRAEDRICH and HAM, 1982), soil microorganism (BADEJO *et al.*, 1995), soil moisture (GICHERU, 1994), and growth of plant (LITZOW and PELLETT, 1983; GREENLY and RAKOW, 1995; FOSHEE *et al.*, 1996). Decomposition rate and chemical changes in the structural components of leaf litter during mulching in urban ecosystems have received little attention. Therefore, recycling of wastes from urban forest is hampered by limited information.

Leaf litters of *Cinnamomum camphora* Sieb., *Zelkova serrata* Makino, *Firmiana simplex* W. F. Wight, and *Ginkgo biloba* Linn., which are often used as street tree species in Japan were collected from the campus of the University of Tokyo. Decomposition rate and changes of chemical structure of leaf litters during 1 year mulching were investigated.

### Experimental

#### Mulching of leaf litter

Mulching experiment was conducted at the experimental field (35°41'N, 139°46'E) of the University of Tokyo. Fallen leaves of *Z. serrata*, *F. simplex* and *Ginkgo biloba*, which are deciduous trees, were collected from Yayoi campus of the University of Tokyo in early winter of

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1999 and those of *C. camphora*, which is an evergreen tree species, were collected in April 2001. Litter bags (20×20 cm) made of nylon net with a mesh size of 0.1 mm were used. Litter bags were recovered with 2-month intervals up to one year.

### Determination of decomposition rate

Leaf litters in the bags were cleaned from foreign materials and air dried for 3 days at room temperature, then weighted. Moisture content of each sample was determined after drying overnight at 105°C. The remaining mass in the litter bags was calculated using the following formula:

$$W_{RM} = W \times (1 - \text{moisture content}),$$

where  $W$  is the air dried weight and  $W_{RM}$  is the oven dried weight of remaining sample. The dry mass loss of leaf litter was calculated as follows:

$$L(\%) = (W_0 - W_m) / W_0 \times 100,$$

where  $L$  is oven dried weight loss, and  $W_0$  and  $W_m$  are oven dried weights of the original and mulched samples, respectively. The litter decomposition rate was calculated based on decay model of Olson (1963):

$$W_t / W_0 = e^{-kt},$$

where  $W_t$  is the oven-dried weight at time  $t$ , and  $k$  is the annual decomposition rate.

### Chemical analysis of leaf litter

Air-dried sample was ground in a Wiley mill to pass a 420  $\mu\text{m}$  sieve. The ground sample was extracted three times with boiling 80% aqueous ethanol (v/v) for 1 h followed with water extraction overnight at room temperature with shaking.

The neutral sugars were obtained from the extract-free sample (solid residue) by hydrolysis with 3% sulfuric acid (w/w) for 1 h at 121°C after treatment with 72% sulfuric acid (w/w) for 1 h at room temperature. The neutral sugars in the hydrolysate were analyzed as their alditol acetates (BLAKENEY *et al.*, 1983) using *myo*-inositol as an internal standard by Shimadzu GC-1700 Gas Chromatograph (TC 17 capillary column: 30 m×0.25 mm id., column temperature 210°C. Carrier gas: He). Uronic acid was determined colorimetrically, using glucuronic acid as a standard (SCOTT, 1979). Klason residue of the extract-free sample was determined gravimetrically by the TAPPI Standard T 222om-88 used for determination of Klason lignin content in woody materials. Ash content was measured after combustion in a muffle furnace for 3 h at 700°C. The contents of carbon and nitrogen of the extract-free sample were determined with a CHN elemental analyzer (Perkin Elmer 240). Methoxyl group content of the extracts free sample and Klason residue was determined according to a procedure modified by GOTO *et al.* (2000). Methoxyl contents of the Klason residues were used to calculate the estimated lignin content by the following formula based on the assumption that lignin C<sub>6</sub>-C<sub>3</sub> unit (equivalent 200) carries one methoxyl group;

$$\text{Assumed lignin content (\%)} = \text{Methoxyl} / 1000 \times (200 / 1000) \times \text{KR},$$

where KR is yield of Klason residue (%) and Methoxyl is the methoxyl content in the Klason residue (mmol/kg).

## Results and discussion

### Chemical composition of original leaves

Total extracts of *C. camphora* and *G. biloba* made up 35.9 and 39.2%, while *Z. serrata* and *F. simplex* gave 17.9 and 16.8% (% of dry matter) of the original leaf litter (Table 1). The major components of 80% ethanol extracts of wheat straw compost are monomeric or oligomeric carbohydrates and inorganic salts (IIYAMA *et al.*, 1995).

Glucose was clearly the predominant neutral sugar component of leaf litters (Table 1). Most of the glucose would originate from cellulose. Hemicelluloses are heteropolysaccharides and relatively easily hydrolyzed by acids to their monomeric components such as D-glucose, D-mannose, D-xylose, L-arabinose, L-rhamnose, and galactose etc (SJÖSTRÖM, 1981). The composition and structure of hemicelluloses in the hardwood differ from softwood. Leaves of *C. camphora*, *Z. serrata*, and *F. simplex* were characterized by high contents of xylose, which is the major component of hemicellulose in hardwood. Galactoglucomannans are the principal hemicelluloses in softwoods. *G. biloba* gave higher mannose than xylose (Table 1). Total yield of neutral sugar was varied from 19.7% (*G. biloba*) to 25.2% (*F. simplex*) leaf litter, which was much lower comparing with wood. The contents of uronic acid of the leaf litters of *C. camphora*, *Z. serrata*, *F. simplex* and *G. biloba* were 4.7%, 6.7%, 6.9% and 6.7% respectively. Uronic acid could originate from pectic substrates such as rhamnoglacturonan and the content generally higher in younger living cells.

Table 1. Chemical composition of original leaf litter (% of original dry sample)

	Angiospermae			Gymnospermae
	<i>C. camphora</i>	<i>Z. serrata</i>	<i>F. simplex</i>	<i>G. biloba</i>
Extracts				
Boiling 80% ethanol	32.7	13.7	12.1	33.2
Water at 40°C	3.2	4.2	4.7	6.0
Total	35.9	17.9	16.8	39.2
Neutral sugars				
Glucose	11.9	9.9	14.0	9.8
Mannose	0.5	1.5	0.4	1.9
Galactose	1.7	2.1	2.1	2.0
Xylose	5.8	4.1	5.8	1.6
Arabinose	1.9	2.7	1.8	2.7
Rhamnose	0.6	1.1	1.0	1.7
Total	22.5	21.4	25.2	19.7
Uronic acid	4.7	6.7	6.9	6.7
Klason residue	24.4	38.3	35.5	22.6
Nitrogen	0.9	0.8	0.7	0.5
Ash	4.5	10.4	7.7	6.4

The term “Klason residue” instead of “Klason lignin” was used because the residue obtained by the Klason procedure seemed to contain significant amounts of non-hydrolyzable products other than lignin. The contents of Klason residue of the original leaf litters ranged from 22.6% (*G. biloba*) to 38.3% (*Z. serrata*).

Nitrogen content ranged from 0.5% (*G. biloba*) to 0.9% (*C. camphora*) and the ash contents varied from 4.5% to 10.4%. The high content of nitrogen and ash in leaf litter comparing to wood would also contribute to the high content of Klason residue.

### Mass loss

Annual decomposition rate ( $k$ ) is commonly used for studies to elucidate litter decomposition rates among species or various environments. Mass loss of the leaf litters ranged from 30.5% (*Z. serrata*) to 52.7% (*G. biloba*) after 1 year mulching (Fig. 1). Annual decomposition rates calculated according to a decay model of OLSON (1963) were 0.63, 0.36, 0.48 and 0.75 for the leaf litters of *C. camphora*, *Z. serrata*, *F. simplex* and *G. biloba*, respectively. *G. biloba* leaves have been used as an insecticide from ancient times (FUJIHARA and KATO, 1990), therefore it was generally thought that *G. biloba* leaves resist biodegradation. However the decomposition rate of *G. biloba* leaf litter was higher than that of *C. camphora*, *Z. serrata* and *F. simplex* under same environmental conditions (Fig. 1).

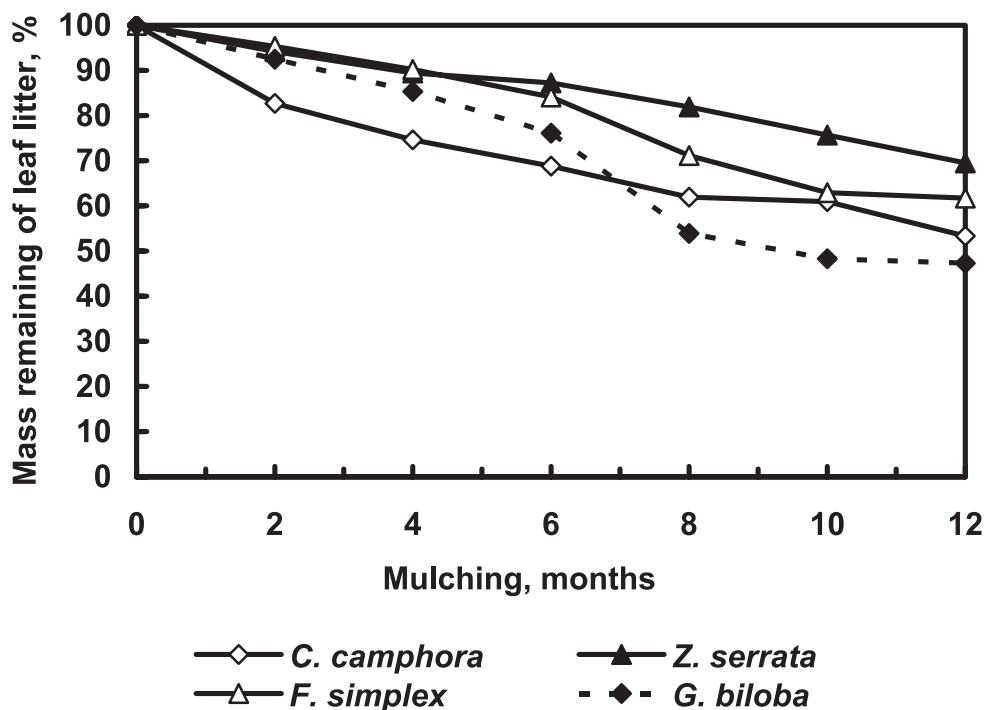


Fig. 1. Mass remaining of leaf litter during mulching

It has been suggested that the decomposition rate is dependant on chemical composition of original plant litter (SWIFT *et al.*, 1979). The initial chemical composition of plant litters affects on its decomposition rate as the chemical changes occurring during decomposition (MCCLAUGHERTY and BERG, 1987). Soluble substances, which could extractable with solvents, are easily decomposed and positively correlate with decomposition rate. Lignin, polysaccharides covalently associated with lignin and nitrogen regulate decomposition rate in late stages and are negatively correlated with decomposition rate (MEENTEMEYER, 1978; MELILLO *et al.*, 1982). *G. biloba* and *C. camphora* leaf litters were characterized by low Klason residues and high content of extracts, and these characteristics would result high decomposition rate.

### Extracts

Chemical components of litters can be divided into three broad groups, namely soluble substances, polysaccharides (cellulose and hemicellulose), and acid insoluble aromatic compounds including lignin and polyphenolic compounds other than lignin. Each group begins its net mass loss at a different stage of decomposition (MCCLAUGHERTY and BERG, 1987). Soluble substances were lost rapidly during early stage of decomposition (MCCLAUGHERTY *et al.*, 1985; BERG, 1986). Extracts from *C. camphora*, *Z. serrata* and *F. simplex* were decreased fast at the early stage of decomposition, while extracts from *G. biloba* leaf litter decreased slowly at the

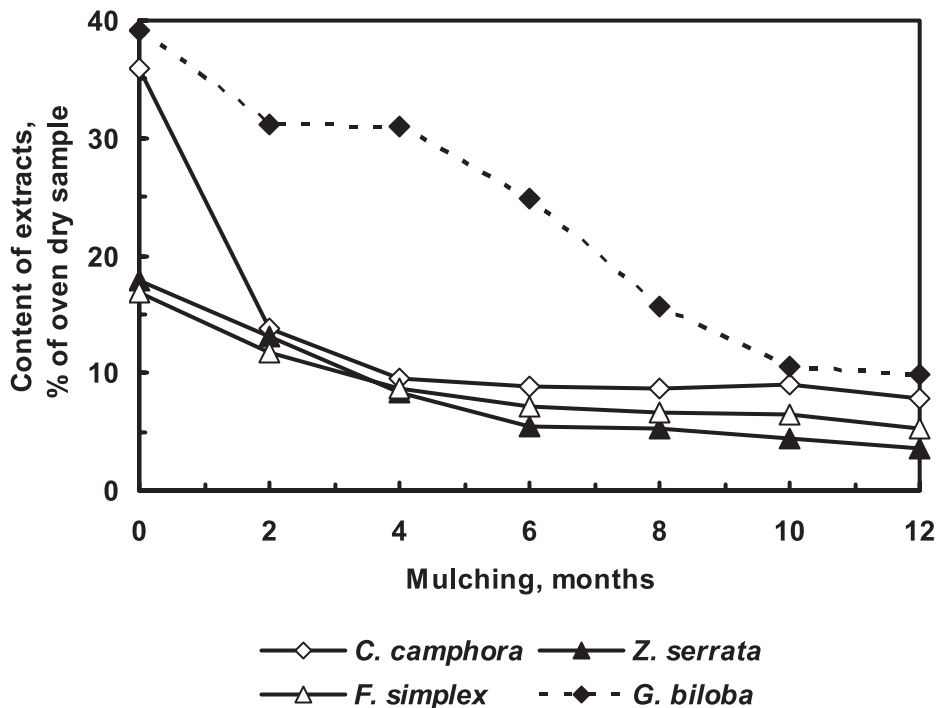


Fig. 2. Changes in extracts of leaf litter during mulching

early stage of decomposition (Fig. 2). The different decomposition pattern of *G. biloba* extracts during 1 year mulching would be due to its biologically active components, which resist biodegradation at the early stage decomposition. Extracts were lost 88.3%, 83.8%, 80.4% and 88.3% of their initial mass, respectively, indicating that leaf litter lost almost all of initial extracts during 1 year mulching.

### Solid residue

Soluble substances started to decompose first, followed by hemicellulose, cellulose, and lignin (BERG and STAAF, 1980). Neutral sugars content decreased (Fig. 3) and their total loss were 50.7%, 37.9%, 66.1% and 71.6% for *C. camphora*, *Z. serrata*, *F. simplex* and *G. biloba*, respectively. Glucose, which is about half mass of neutral sugars and mostly originates from cellulose, was lost 49.5%, 33.3%, 71.4% and 68.4% of their initial mass (Fig. 4). Cellulose exhibited higher decomposition rate than that of hemicellulose (SWIFT *et al.*, 1979). BERG *et al.* (1982) also reported that hemicellulose was degraded slower than cellulose. Those earlier findings are similar to the result obtained for *in vitro* enzymatic digestion of cell wall components of grasses (LAM *et al.*, 1993; HIGUCHI, 2002). In the present study, hemicellulose of *C. camphora*,

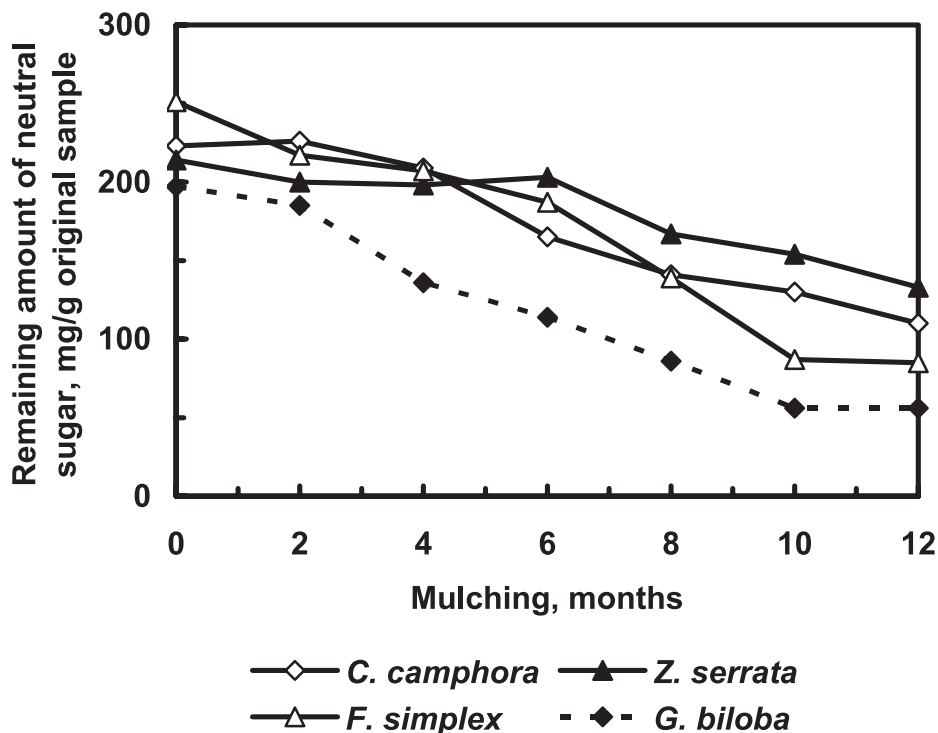


Fig. 3. Changes in remaining amount of neutral sugar of leaf litter during mulching

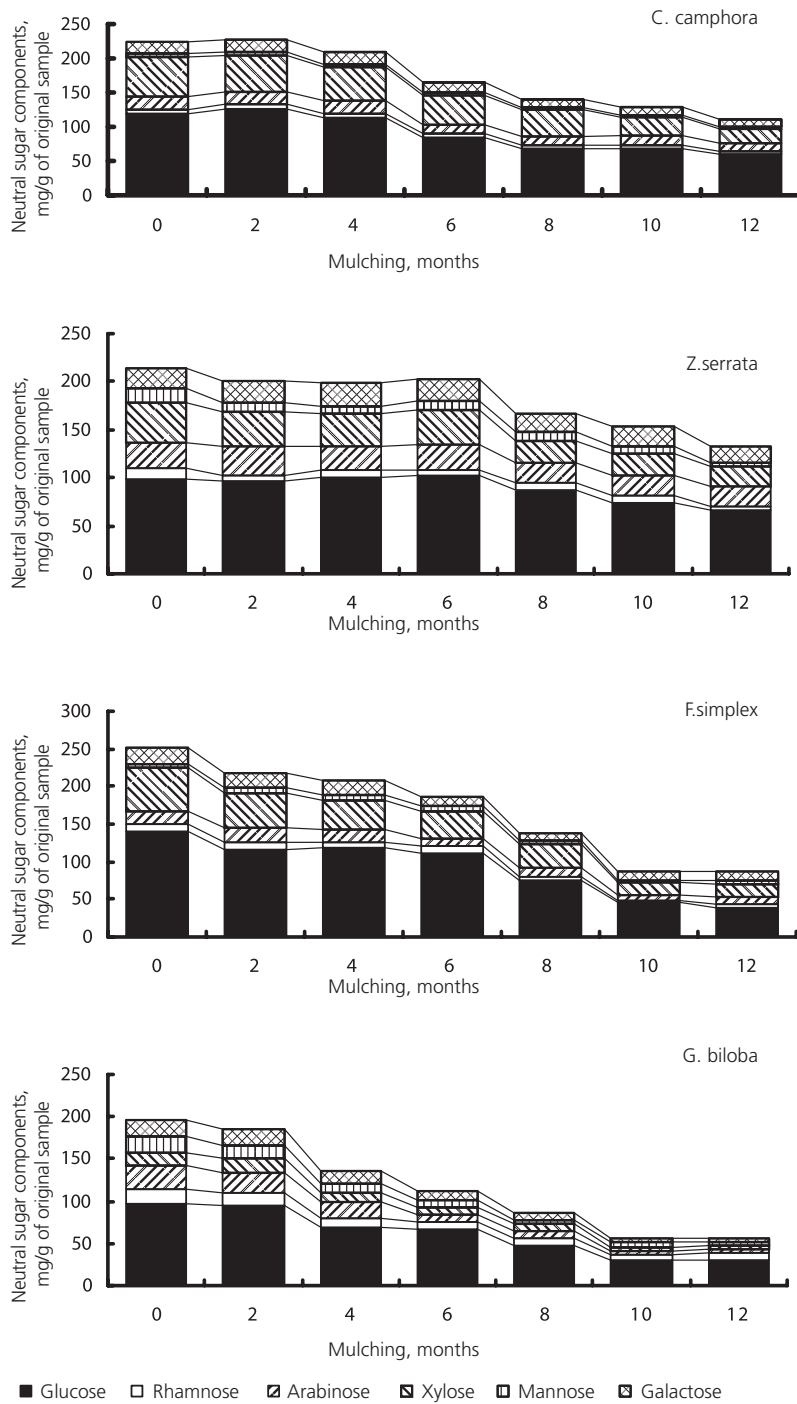


Fig. 4. Changes in neutral sugar components of leaf litter during mulching

*Z. serrata* and *G. biloba* leaf litter exhibited somewhat higher decomposition rate comparing to cellulose, while hemicellulose decomposition rate of *F. simplex* was lower than cellulose (Table 2). Uronic acids of *C. camphora*, *Z. serrata*, *F. simplex* and *G. biloba* were lost 76.8%, 23.9% and 90.9%, and 89.6%, respectively (Fig. 5).

Lignin content of leaf litter is an important biological factor affecting decomposition rate (MELILLO *et al.*, 1982). The relative content of Klason residue increased during mulching (Fig. 6), which agreed with the results reported in previous papers (BERG and WESSÉN, 1984; SALAMANCA *et al.*, 1998; SINGH *et al.*, 1999). Not only the relative content but also the absolute amount of Klason residue increased initially (Fig. 7). Lignin is a decay resistant biopolymer usually regarded as a rate regulating factor in leaf litter decomposition (BERG and STAAF, 1980). Hence, lignin content may be used to predict decomposition rate and weight loss of leaf litter (MEENTEMEYER, 1978; MCCLAUGHERTY and BERG, 1987; SALAMANCA *et al.*, 1998). The most widely used method for quantitative determination of leaf litter lignin has been the Klason procedure, which

Table 2. Loss of cellulose and hemicellulose during mulching for 12 months (% of original sample)

	<i>C. camphora</i>	<i>Z. serrata</i>	<i>F. simplex</i>	<i>G. biloba</i>
Cellulose	50	33	71	68
Hemicellulose	51	42	58	75

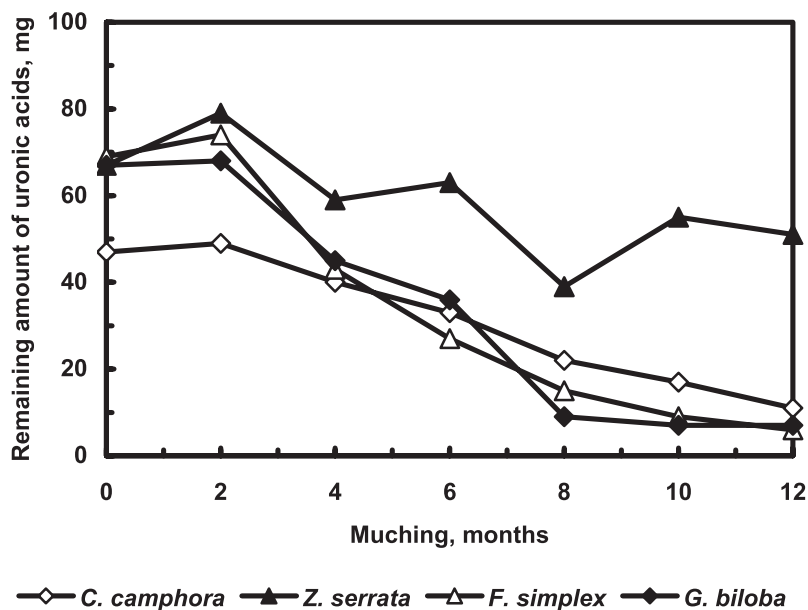


Fig. 5. Changes in uronic acid content of leaf litter during mulching



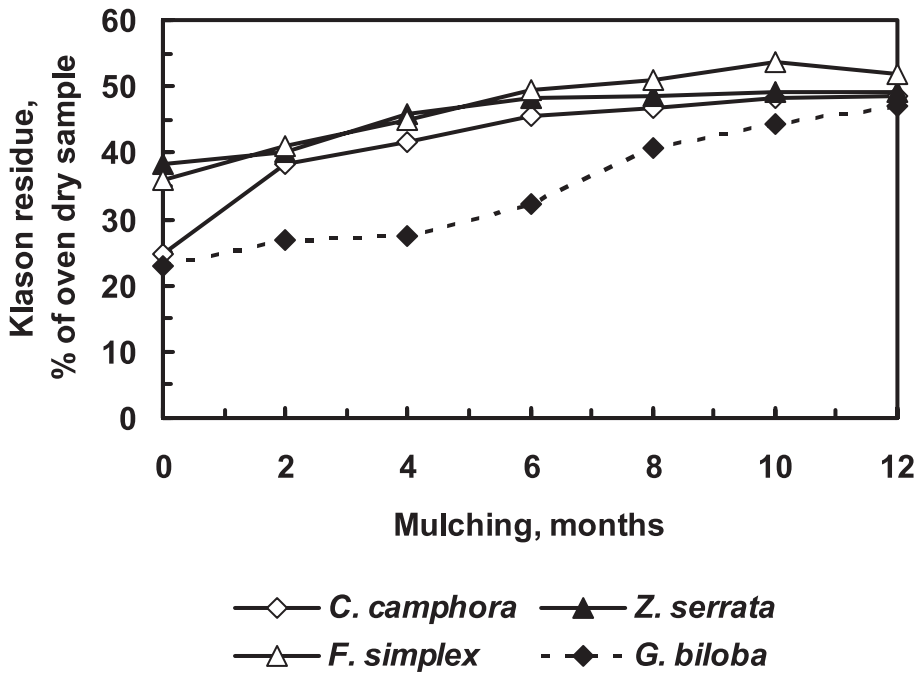


Fig. 6. Changes in relative content of Klason residue of leaf litter during mulching

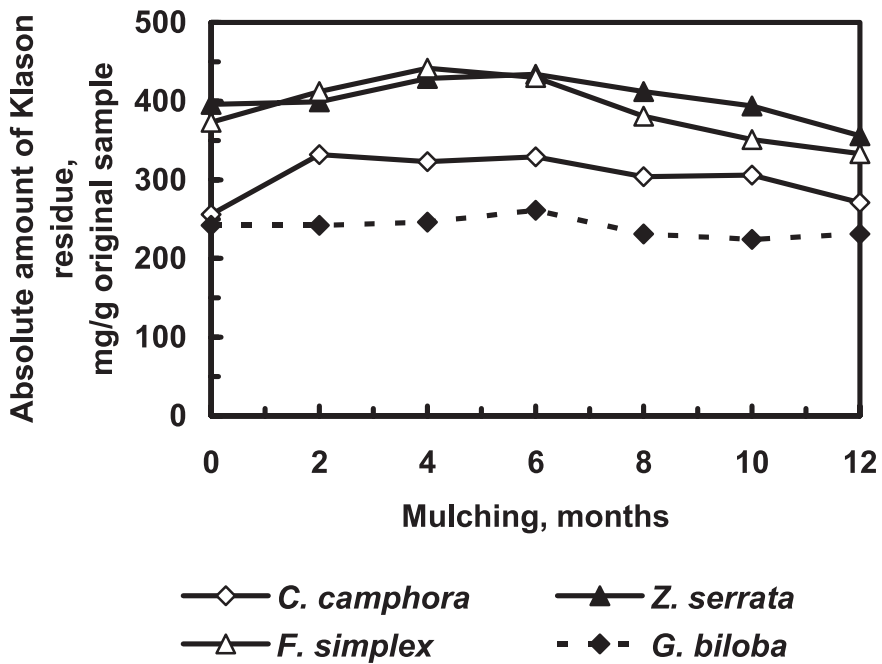


Fig. 7. Changes in absolute amount of Klason residue of leaf litter during mulching

Table 3. Methoxyl content of Klason residue

	<i>C. camphora</i>	<i>Z. serrata</i>	<i>F. simplex</i>	<i>G. biloba</i>
Methoxy content (mmol/kg)	1,217	806	1,174	519
Klason residue <sup>a</sup> (% SR)	38.1	46.7	42.7	37.1
Assumed lignin content <sup>b</sup> (%)	9.3	7.5	10.0	3.9

<sup>a</sup> SR: solid residue (extract free sample).

<sup>b</sup> Calculated based on methoxyl content of Klason residue according to the formula given in experimental section.

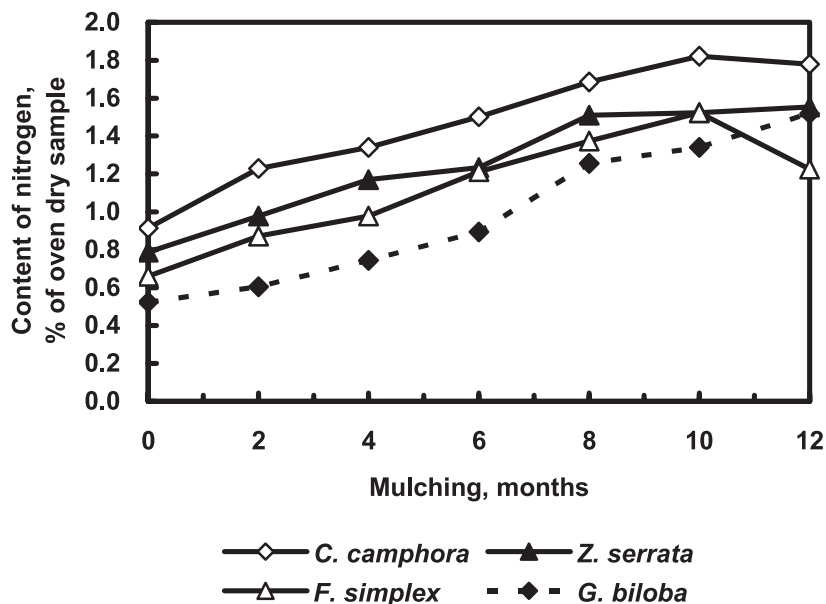


Fig. 8. Changes in relative content of nitrogen of leaf litter during mulching

affords an insoluble residue from hydrolysis with sulfuric acid. BERG (1986) explained Klason procedure is correct only for newly shed litter material because the analytical method also registers secondary compounds and humic substances formed during the decomposition process. However, even when applied to newly shed leaf litter, the value ranged from 22.6% (*G. biloba*) to 38.3% (*Z. serrata*) (Table 1). Klason procedure is the standard method for lignin determination of wood samples, while the method usually gives an overestimated value for the lignin of herbaceous plant materials (IYAMA and WALLIS, 1990). The Klason residue is representative undegradable material of the solid residue, and not necessarily lignin. The methoxyl group is a marker functional group of lignin and the accurate content of lignin can be estimated from the methoxyl content of the Klason residue (JIN *et al.*, 2003). In Table 3, rough estimation of real

lignin content is given as assumed lignin content. Those values were calculated based on the assumption that one lignin aromatic carries one methoxyl group. Although the detailed analysis on the structure of Klason residue is still under way, it is clear that lignin content of leaf litter is much smaller than the value of Klason residue. Even though part of Klason residue of leaves is not lignin, it is clear that the residue obtained by Klason procedure is critical factor in leaf litter decomposition.

Relative content of nitrogen increased in all leaf litter (Fig. 8) Those nitrogen contents correspond to 5.6%, 5.0%, 4.4% and 3.1% as protein content ( $N\% \times 6.25$ ) for *C. camphora*, *Z. serrata*, *F. simplex* and *G. biloba*, respectively, increased to 12.0%, 10.0%, 8.1% and 10.6% during one year mulching. The percentage increases in the nitrogen content were largely attributable to more rapid losses of non-nitrogenous leaf constituents while the weight of nitrogen present in the leaves remained relatively constant.

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### Summary

Leaf litter decomposition and changes in the chemical components of four urban tree species (*Cinnamomum camphora* Sieb., *Zelkova serrata* Makino, *Firmiana simplex* W. F. Wight, *Ginkgo biloba* Linn) during one-year mulching were analyzed. The mass loss of the leaf litters ranged from 30.5% (*Z. serrata*) to 52.7% (*G. biloba*) after one year. Annual decomposition rates were 0.63, 0.36, 0.48 and 0.75 for the leaf litters of *C. camphora*, *Z. serrata*, *F. simplex* and *G. biloba*, respectively. Both water and 80% aqueous ethanol extracts decreased significantly during the one year of mulching. The amount of total neutral sugars decreased and their total losses were 50.7, 37.9, 66.1 and 71.6% based on their initial amount. Glucose accounted for about half the mass of the total neutral sugar. The relative content of Klason residue increased during mulching, suggesting that Klason residue resists decomposition. Even though the main part of Klason residue of leaves is not lignin, it should be emphasized that the residue obtained by the Klason procedure is a critical factor in leaf litter decomposition. The nitrogen content increased during the one year of mulching and the high content of nitrogen content would also contribute to the high content of Klason residue in leaf litter.

**Key words:** Urban tree, Leaf litter, Mulching, Klason lignin, Cell wall composition

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## マルチングにおける都市緑化樹木落葉の細胞壁化学成分の経時的变化

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### 要 旨

都市の代表的な都市緑化樹木であるクスノキ (*Cinnamomum camphora* Sieb.), ケヤキ (*Zelkova serrata* Makino), アオギリ (*Firmiana simplex* W. F. Wight), イチョウ (*Ginkgo biloba* Linn) を対象として, それらの落葉のマルチングにおける分解過程を明らかにするため, 落葉をリターバッグに封入し, 圃場に敷きならした。2ヶ月ごとに回収し, 重量, 細胞壁化学成分の測定を行った。全体の重量の減少は1年の試験期間中に最低のケヤキで30.5%, 最高のイチョウで52.7%であった。分解速度の指標となるオルソン指数関係式による分解係数Kはクスノキ, ケヤキ, アオギリ, イチョウ (以下, この記載順による) でそれぞれ0.63, 0.36, 0.48, 0.75であった。落葉およびそのマルチング処理試料の成分分析は, 主に抽出区分 (80% 含水エタノールおよび水による逐次抽出) を除いたあとの抽出残渣について行った。マルチングによって抽出区分の割合は速やかに減少したが, 抽出残渣の重量減少は緩慢であった。細胞壁多糖類の主体である中性糖の含有率はすべての樹種で当初 20% 前後であったのが, 1年のマルチング後に各樹種でそれぞれ, 50.7%, 37.9%, 66.1%, 71.6% 減少した。一般的にリグニン量の指標とされている Klason 残渣の, 未抽出の試料中の含有率は当初 22.6% (イチョウ) から 38.3% (ケヤキ) の範囲であったが, マルチングとともにその相対含有率は継続的に増加した。このように, マルチングによる落葉の重量減少に寄与する成分は, 抽出区分と細胞壁構成多糖が主体であって, Klason 残渣を構成する成分は落葉中の難分解物であることが確認された。窒素の含有量は木材に比べ高く, マルチング過程で増加した。高い窒素含有量は落葉の Klason 残渣含有量が高くなる一つの要因と考えられる。

キーワード: 都市緑化樹木・落葉・マルチング・細胞壁成分・Klason リグニン