Initial carbon and nitrogen contents and sugar release characteristics in stream water during initial leaching of *Quercus glauca* leaves

Hirokazu Kitamura

Laboratory of Landscape Conservation, Minami-Kyushu University, Miyakonojo, Miyazaki 885-0035, Japan

Received October 13, 2011; Accepted January 26, 2012

To clarify the release characteristics of sugars in relation to initial carbon and nitrogen contents of evergreen *Quercus glauca* leaves during initial leaching period, which dominate and contribute considerable quantities of allochthonous material to streams in the region, well washed and dried new and old leaves of *Q. glauca* (sun leaves and shade leaves) were submerged in invertebrate-free stream water after measuring the carbon and nitrogen contents of each leaf. 1, 5, 10, 20 and 30 days after submersion, sugar concentrations of the water were measured. The mean initial carbon contents ranged from 45.75 to 46.86 (%), indicating higher values than the deciduous leaves in references. The mean initial nitrogen content ranged from 1.83 to 2.16 (%) and significant difference was observed between shade-exposed old leaves and leaves from the other groups (*P<0.0001*). The initial carbon and nitrogen contents of leaves did not influence the release rates of carbon and nitrogen from the leaves. Sucrose, glucose and fructose were detected. Sucrose accounted for more than 60% of the total sugars. The average concentration of total sugar in the water treatment containing new leaves without aeration reached a maximum concentration of 26 (mg/l) after 30 days of immersion. *Q. glauca* leaves have a longer sugar-leaching period and leached sugars less rapidly than the desiduous leaves. The release rate and subsequent decomposition into water and carbon dioxide were increased by abrasion of leaf surfaces. These differences between deciduous and evergreen leaves might arise due to differences in leaf composition and structure. Sugars are derived from soluble carbohydrates that are lost through the cuticle of evergreen leaves; however, the relatively thicker cuticle of the evergreen leaves than deciduous leaves reduces the rate of sugar release. The tough outer surfaces of evergreen leaves may also slow rate at which the mass of the leaf decreases, particularly until such time as the outer layer of the epidermis, which contains cutin, breaks down in water.

Kye wards: carbon and nitrogen contents, leaching, *Quercus glauca* leaves, stream water quality, sugar.

1. INTRODUCTION

Leaf litter in streams, also referred to as allochthonous material, is the primary source of energy for the secondary consumers in the streams. This organic matter is decomposed through a combination of physical, chemical and biological processes[1]. In leaves, the process of decomposition can be divided into leaching, conditioning, and fragmentation. Consequently, shortly after entering a stream, the soluble chemicals within the leaf are leached from the plant tissue, which is then colonized by microbes and fragmented by mechanical abrasion and invertebrate activity. The instream processing of this allochthonous material results in the production of dissolved organic matter (DOM)[19] and the release of inorganic nutrients into the water[2, 20]. Since considerable quantities of allochthonous material enter lotic ecosystems from riparian forests[4], substances derived from the decomposition of leaf litter may have an important effect on stream water quality.

Several studies have suggested that the leachates from the leaf litter of different tree species are markedly different, and that they have different effects on the chemical characteristics of stream water [8, 23]. Studies have also examined the physical degradation of leaf litter, including decreases in the mass of allochthonous material over time [5, 18] and the chemical changes associated with the degradation of deciduous leaf litter in streams [12, 13, 16]. The dependence of woodland stream ecosystems on allochthonous organic matter, primarily autumn-shed leaf litter, is well documented[21, 22, 29]. However, relatively few studies have examined the leachates of evergreen leaves or their effect on stream water quality during initial decomposition. Indeed, the nutrient contents of evergreen leaves and their contribution to the dissolved organic substances in lotic systems is still largely unknown.

In a previous paper, we clarified the release characteristics of total organic carbon (TC), total nitrogen (TN), and specific ions from evergreen Quercus glauca leaves

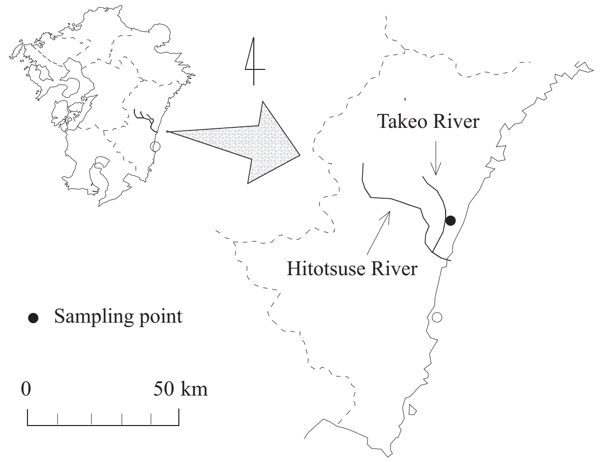


Fig. 1. Investigated site.

immersed in stream water for 30 days [17]. In this study, the release characteristics of sugars, which are an important component of leaf organic matter and are generally leached relatively rapidly 30), were clarified in *Quercus glauca* leaf litter. The duration of the study was the initial leaching period, over which sugar concentrations were measured in relation to initial carbon and nitrogen contents of the leaf. *Quercus glauca* is an evergreen species that is dominant in the riparian zone of temperate streams in southern Kyushu, Japan, and which contributes considerable quantities of allochthonous material to streams in the region.

2. METHODS

2-1. Materials

A detailed explanation of the methodology employed to prepare the leaf material for examination can be found elsewhere [17]. Briefly, leaf samples used for the experiments were collected from the riparian zone of the Takeo River in Saito City, Miyazaki Prefecture. The Takeo River, a tributary of the Hitotsuse River, ranges from 100 to 600 m in elevation and originates in the Southern Kyushu Mountains (Fig. 1). For the present study, all leaves were collected from a height of 2 to 4 m above the ground surface from a *Quercus glauca* tree that grew on the unfertilized alluvial plain of the river.

During the growth phase, carbohydrates are produced in the leaves of *Q. glauca* by photosynthesis; the amount of carbohydrates produced in the leaf, and by inference, the amount of carbohydrates that will subsequently be leached from the leaf, is thus a function of leaf age. In the present study, four groups of leaves were randomly collected. These groups, which corresponded with the groups used in a previous study [17], differed with respect to the amount of time that had passed from leaf emergence (leaf age) and the availability of sunlight: sun new-leaves (SUN), sun old-leaves (SUO), shade new-leaves (SHN) and shade old-leaves (SHO). Sun-exposed leaves (sun leaves) are leaves that have been exposed to direct sunlight for greater part of a day, and shade-exposed leaves (shade leaves) are leaves that have mainly grown in the shade. Leaves picked from the tip of branches of the tree were considered to be new leaves. The leaves had been growing since March 2010 and still appeared yellow-green in color due to the relatively short time that had elapsed since leaf expansion. Leaves that were picked from the older branches of the tree were also considered to be old leaves and were collected immediately before abscission, which means that they had been growing since the previous spring and could be distinguished from new leaves by their dark-green color. All the leaves collected were whole leaves and retained their original shape without any skeletonization. Any visually abnormal or injured leaves were not harvested at the time of sample collection on May 20, 2010. Leaf emergence in *Quercus glauca* at the study site started at the end of March and expansion of the leaves occurred from the beginning of April to the end of May every year. Stream water (100 L) was collected from the stream at the same site for all leaching experiments.

2-2. Measurement of initial carbon and nitrogen leaf contents

The harvested leaves described above were washed with distilled water to remove dust and aeroso [5], then air-dried for 1 week and oven-dried at 80°C for 12 h. Ten leaves of each group were then selected, and tissue samples measuring approximately 2 mg were removed from each leaf. Samples were always taken from an area 5 mm to the right of the midrib in the center of the leaf. Each fragment was analyzed using an ash-free CHNS/O Analyzer (2400 Series II, Perkin Elmer, Inc.,) to measure the initial carbon and nitrogen leaf contents.

2-3. Measurement of sugar concentrations leached from leaves

Sugar concentrations released from sun leaves, in which no significant differences in initial carbon and nitrogen contents of old and new leaves were observed, were analyzed. The harvested leaves described above were washed with distilled water to remove dust and aerosol, then air-dried for 1 week and oven-dried at 80°C for 12 h. Old and new leaves of sun leaves (i.e. SUN and SUO) were submerged in invertebrate-free stream water with or without aeration as described previously [17]. The four water treatments included old leaves with aeration, old leaves without aeration, new leaves with aeration, and new leaves without aeration. Water was aerated to simulate reaeration in natural streams [14]. In each treatment, 25 subsamples of five leaves were then randomly selected, weighed and submersed in 1 (l) of stream water in a beaker. All subsamples were then allowed to stand at ambient temperature (20-25°C) for 1, 5, 10, 20 and 30 days when five subsamples from each treatment were randomly selected for analysis. In addition, each water subsample was hen filtered through a 0.20 (μm) filter and sugar concentrations in the water were measured by high performance liquid chromatography and a Shodex column (NH2P-50 4D, Hitachi K.K.). (The rate of the decrease in leaf mass and the concentrations of TC and TN leached from leaves in each subsample during the initial leaching period have been published previously [17]).

3. RESULTS

3-1. Initial carbon and nitrogen contents of leaves

The initial carbon and nitrogen contents of the leaves in each group are shown in Fig.2. A relatively wide variety

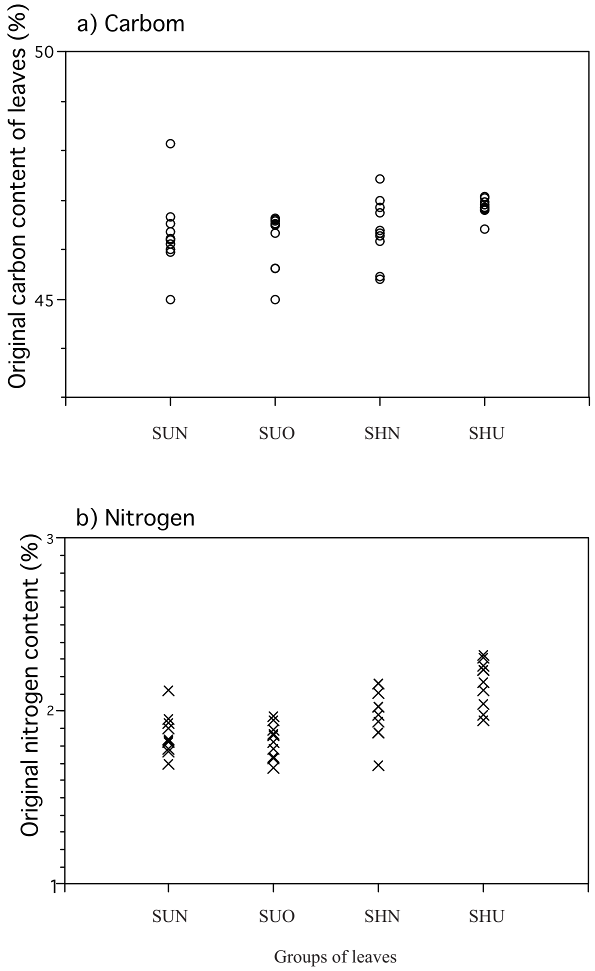


Fig. 2. Initial carbon (a) and nitrogen (b) contents in leaves from each leaf-type group.

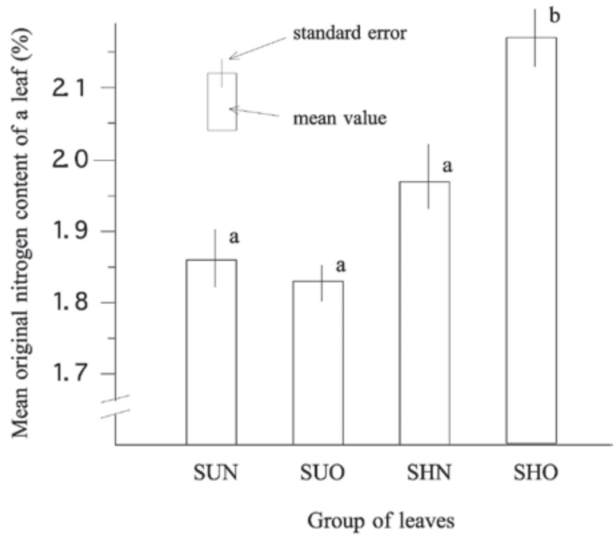


Fig. 3. Results of *Sheffe's* multipul range test. Letters above columns indicate statistically significant differences ($P < 0.05$).

Table 1. Result of one-way ANOVA

dependment variable	n	the mean square	*F*	*P*
carbon content	3	2.129	1.332	0.279
nitrogen content	3	0.235	13.899	<0.000

of compounds were observed. The mean initial carbon content and standard error in the SUN, SUO, SHN and SHO groups was 45.75±0.75, 46.19±0.18, 46.41±0.20 and 46.86 ± 0.06 (%), respectively. The mean initial nitrogen content and standard error in the SUN, SUO, SHN and SHO groups was 1.86±0.04, 1.83±0.03, 1.97±0.04 and 2.16±0.04 (%), respectively.

Differences among the groups of leaves were analyzed by one-way ANOVA (Table 1), which showed that there were no significant differences in initial carbon content among the groups. Conversely, the initial nitrogen contents of the leaves in the different groups were significantly different (P<0.0001). Since the results for nitrogen contents among groups were significantly different, differences among individual groups were estimated using *Scheffe's* multiple range test (Fig.3). For initial nitrogen content, significant differences were observed between the SHO group and the other three groups of leaves (P<0.05). No significant differences in the initial nitrogen content were observed in the other groups.

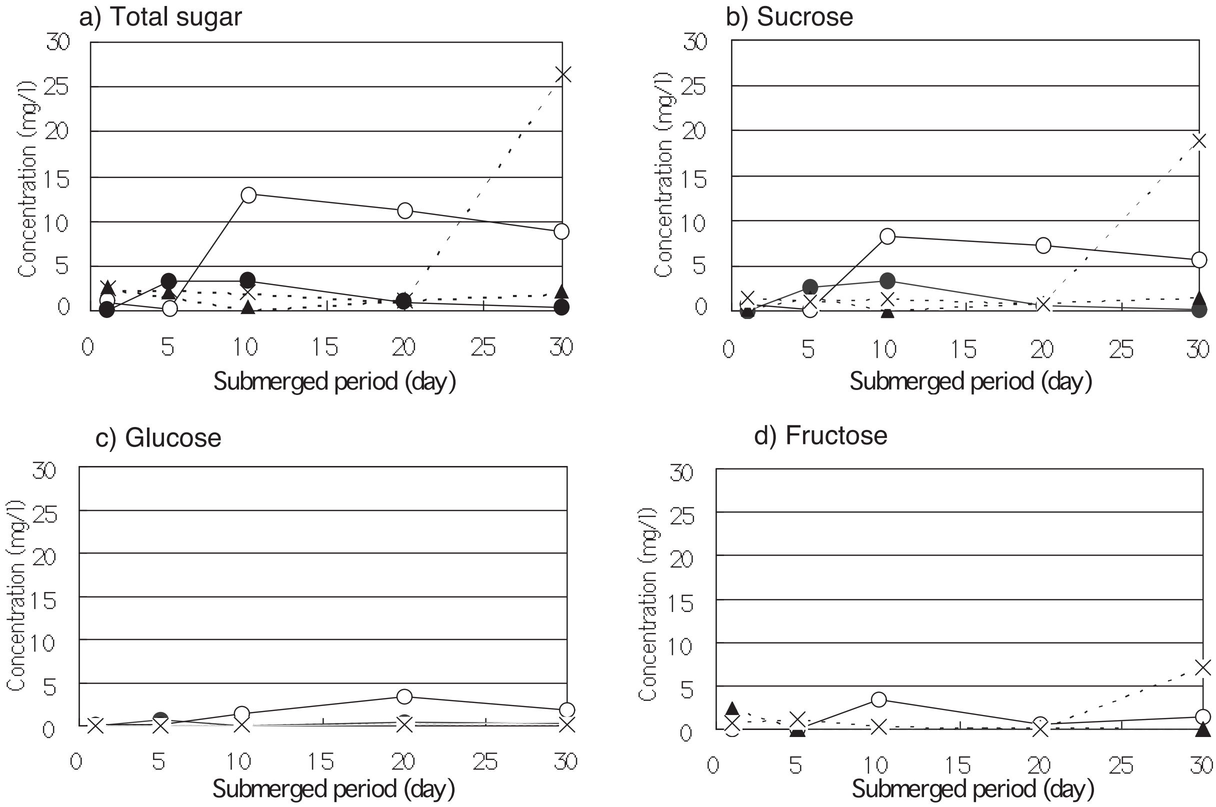
3-2. Sugar

Sugar concentrations released from sun leaves, in which no significant differences in initial carbon and nitrogen contents of old and new leaves were observed, were analyzed. No fragmentation of leaves was observed in any of the samples at the end of the experiment.

The mean sugar concentrations in the leaves subjected to each of the water treatments over the course of the experimental period are shown in Fig.4. To avoid errors in the estimated sugar concentrations of leachates obtained from leaves due to the differences in the mass of leaves submerged in the water, sugar concentrations in the figure are given per 1 (g) of leaf tissue.

In the present measurements, sucrose, glucose and fructose were detected. Initial values for sucrose, glucose and fructose in stream water were 2.5, 0.0, and 0.5 (mg/l), respectively. The total sugar concentration was obtained by adding the concentrations of sucrose, glucose and fructose.

For sucrose, glucose and fructose, only slight differences were apparent among treatments before day 5 and the concentrations of these sugars in each treatment was less than 4 (mg/l). However, marked differences appeared among leaf groups 10 days after immersion. After day 10, old leaves without aeration released considerably more total sugar than the leaves in the other water treatments; the mean concentration of total sugar in the old leaves without aeration was 13 (mg/l) after 10 days of immersion, which gradually decreased thereafter. Conversely, the average concentration of total sugar in the water treatment containing new leaves without aeration reached a maximum concentration of 26 (mg/l) after 30 days of immersion. In the leaves exposed to aeration, the mean concentration of total sugar reached a maximum after 5 to 10 days before gradually decreasing thereafter. Thus, sugar concentrations in water treatments without aeration



old leaves with aeration ●
old leaves without aeration ○
new leaves with aeratio ▲
new leaves without aeration ×

Fig. 4. Mean sugar concentrations in sample water treatment over time.

were considerably higher than they were in water treatments with aeration, and maximum sugar concentrations observed in water treatments without aeration after 10 days. In all treatments, the fluctuations observed in the sucrose concentration were similar to those observed for total sugar.

Glucose was seldom detected in the leachate during the experimental period, except in the treatment of old leaves without aeration when the leached glucose concentration ranged from 1.5 to 3.5 (mg/l) after 10 days. Similarly, fructose was only detected in the leachates from old leaves without aeration and new leaves without aeration, reaching maximum concentrations of 3 and 7 (mg/l), after 10 and 30 days after immersion, respectively.

The ratios of the mean concentrations of sucrose, glucose and fructose to the total sugar concentration on each sampling day are shown in Fig.5. As shown by the figure, sucrose accounts for more than 60% of the total sugars that are leached from leaves after 30 days in all treatments, except for the water treatment containing old leaves with aeration, which had the lowest sugar concentrations of all treatments.

4. DISCUSSION

4-1. Overall comparison of the decrease in leaf mass, and TC and TN release between deciduous and evergreen leaves

In deciduous leaves, most leaching occurs within a few days of immersion in water. For example, autumn shade leaves in water can lose up to 40 (%) of their dry mass over several days [11]; the ash-free dry mass of sugar maple, yellow birch, and beech was 85.4 (%) of the initial mass after 2 days of being submerged in the laboratory [25]. Webster *et al*. (1986) [32] demonstrated that up to 25 (%) of the initial dry mass of certain riparian, deciduous tree leaves (*e*.g., *Alnus sp*., *Salix* sp.) was lost by leaching in the first 24 h of immersion [30]. Contrary to the leaves of deciduous species, the evergreen leaves of *Q*. *glauca* examined, lost 4~7 (%) of their mass after 1 day of immersion in water, and *Q*. *glauca* leaves submerged in water without aeration lost less than 25 (%) of their initial mass after 30 in the present study [17]. These results of the previous study are similar to investigations conducted on *Q*. *alba* in which 5.16 (%) of the initial leaf mass was lost within 24 h [27]. The overall decrease in leaf mass of evergreen leaves at 30 days is thus less than a quarter that of deciduous leaves. In addition, the rate of the decrease in leaf mass rates is slower than that observed in deciduous leaves.

A carbon loss ratio, using total carbon (TC) or total organic carbon (TOC) and dissolved organic carbon (DOC), has often used to infer the amount of carbon leached from leaves. In the leaves of the deciduous *Salix gracilistyla*, considerable leaching of DOC from leaf litter occurred within 24 h after immersion in water; indeed, over a 14 day period, approximately 65 (%) of the total DOC in senescent leaves had been leached within 24 h of immer-

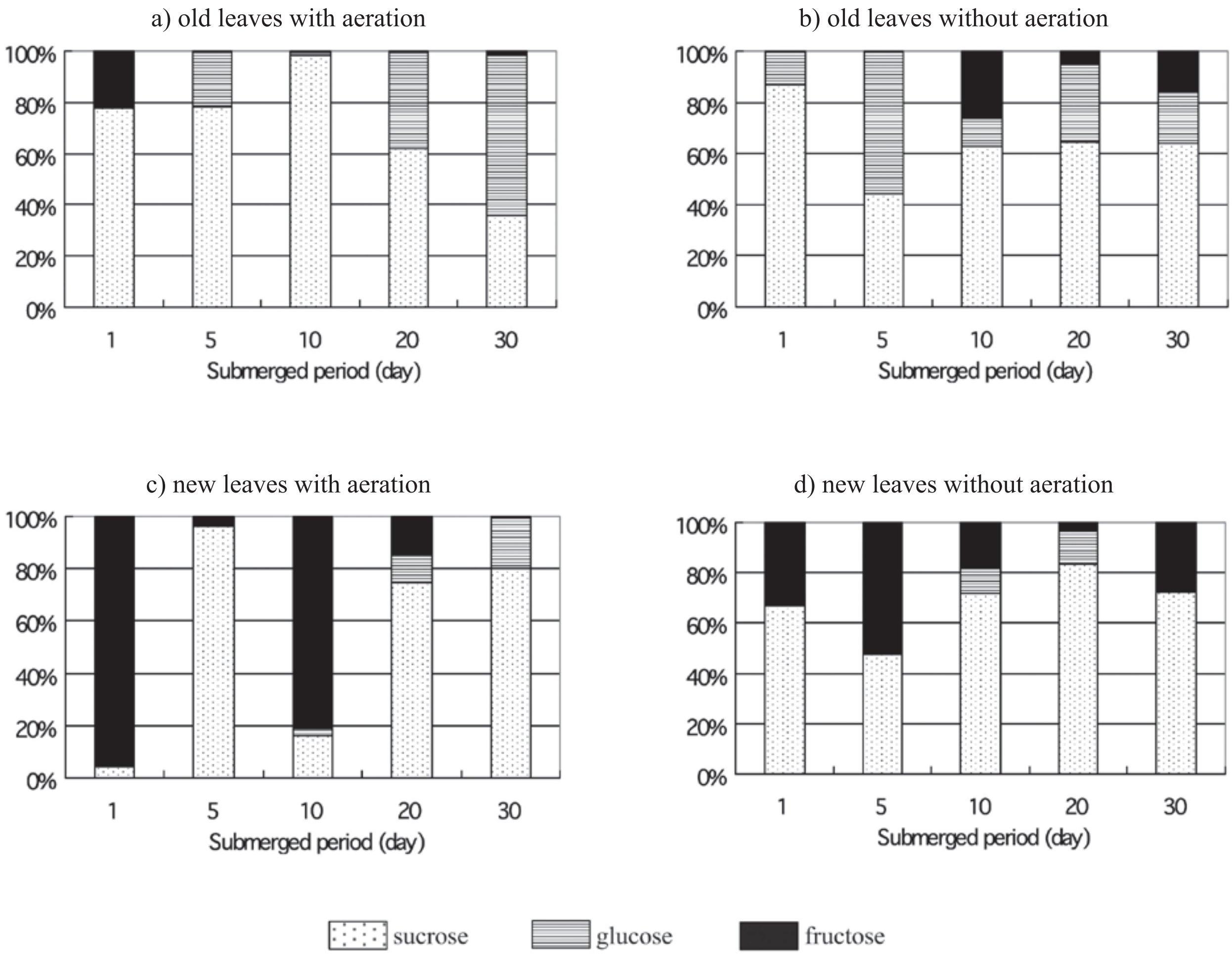


Fig. 5. Each sugar concentration as a percentage of total sugar concentration over time.

sion[28]. In the previous study, TC concentrations from *Q. glauca* leaves in different water treatments increased for 5 to 10 days after immersion[17]. Taken together, the findings of these previous studies indicated that the rate of DOC leaching from leaves of *S. gracilistyla* seemed to be similar to the rate of TC leaching from the leaves of *Q. glauca*. However, the total concentration of TC leached from *Q. glauca* leaves was higher than the DOC conce trations from *S. gracilistyla* leaves.

These differences in the characteristics of carbon release between *S. gracilistyla* and *Q. glauca* leaves may be due to differences in the cuticular surfaces of these deciduous and evergreen leaves. In submerged leaves, the outer layer of the epidermis, which is covered by cutin, degrades and leaches water-soluble materials as it becomes more permeable. Given the anatomical and morphological differences between evergreen leaves and deciduous leaves, leaching from these leaves is also expected to differ between species.

According to the results of the previous study[17], as with TC concentrations, TN concentrations leached from *Q. glauca* leaves increased for the first 5 to 10. Thereafter, TN concentrations in the leachate of the non-aerated treatments generally remained constant, while those in the aerated treatments continued to rise after the 10th day. The TN concentration in the leachate from old leaves after 10 days was slightly higher than that leached from new leaves. The increases in the TN concentration in leachate of each water treatment can be attributed to the increase in the nitrogen content of the decomposing leaves resulting from the increase in microbial activity[3, 7].

4-2. Initial carbon and nitrogen contents of *Q. glauca* leaves

Table 2 shows the values for the overall comparison of carbon and nitrogen contents in tree leaves reported previously. In sun leaves of *Q. glauca*, the mean initial carbon contents range from 45.75 to 46.86 (%) and no significant differences were observed among the four leaf groups (SUN, SUO, SHN and SHO). The mean initial nitrogen content in the leaves of the four groups ranged from 1.83 to 2.16 (%) and significant difference was observed between SHO leaves and leaves from the other groups. Compared to the previous studies listed in Table 2, the values obtained for initial carbon content in *Q. glauca* in this study are relatively high. These relatively high carbon values in *Q. glauca* could potentially be attributed to cutin covering the surface of the evergreen leaves. However, these values are expected to differ between species, particularly with respect to the timing of leaf emergence,

Table 2. General comparison of carbon and nitrogen contents in leaves in the references

Materials	Species	Contents (%)	References
Carbon content	*Alnus glutinosa*	42	6)
	Popla gr.*nigra*	39	6)
	Salix alba	42	6)
	Salix gracilistyla	47	30)
Nitrogen content	*Alnus glutinosa*	2.2	6)
	Popla gr.*nigra*	0.9	6)
	Salix alba	1.6	6)
	Salix sp.	0.9	7)
	Pinus contorta Louden	0.7	7)
	Picea glauca	0.5	7)
	Acer macrophyllum	0.6	10)
	Alnus rubra	2.2	10)
	Tsuga heterophylla	0.6	10)
	Black lcust	2.1	24)
	Betula lenta L.	0.7	24)

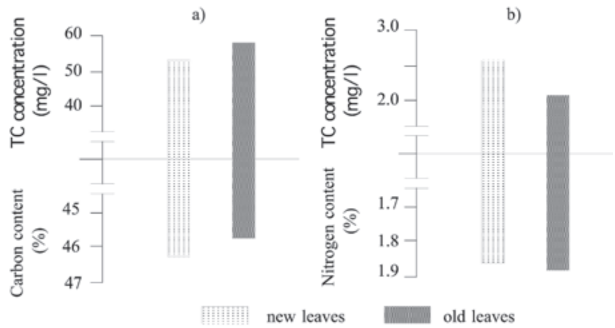


Fig. 6. Comparison of initial carbon content against TC (a) and initial nitrogen content against TN (b).

when the leaves were harvested, and the availability of sunlight. For example, the organic carbon content of the macrophytic tissue remained relatively constant throughout the growing season, declining near the end of the experiment in August[3]. It is thus possible that, during the growth period, the carbohydrates produced in leaves by photosynthesis are locked within the cuticles and epicticular waxes, increasing the carbon content of the leaves.

In addition, in the present study, significant differences were observed in the initial nitrogen content of sun leaves and shade leaves (P<0.0001). Because the initial carbon content of leaves depends on weather conditions (te perature and precipitation) of the spot in which plants have been growing and physiological processes of the tree as well as sunlight availability[27, 28], further measurements and analysis on initial nitrogen contents of leaves under several conditions must be required. The finding that the initial nitrogen content is greater in shade leaves than it was in sun leaves could possibly be, in part, attributed to the immobilization of nitrogen in microorganisms on the surface of the shade leaves. However, further research is required to clarify this issue.

Figure 6 shows the relationship between mean TC and TN concentrations leached from sun leaves of *Q. glauca* in water without aeration during the initial leaching period for 15 days in a previous study[17]. In the same figure, the mean initial carbon and nitrogen contents in the *Q. glauca* leaves harvested in the present study are shown; peak TC and TN concentrations were observed in the initial leaching period, i.e. 5 to 10 days after immergence.

In the present study, TC and TN contents were not determined from the same leaves that were used to determine the initial carbon and nitrogen contents. This is because removing a portion of the leaf for sampling to measure carbon and nitrogen would affect both the quantity of leachate and the rate of leaching from the leaf in subsequent experiments. For example, the amount of water-soluble organic substances leached from needle leaf litter during one day increases from about 1 (%) to 10-12 (%) if leaves are cut beforehand[26]. Similarly, the initial carbon and nitrogen contents in the present study were not measured using the same leaves that were used for the TC and TN leaching experiments. Nonetheless, based on the data shown in Fig.6, it dose not seem that the initial carbon and nitrogen contents of leaves do not influence the release rates of carbon and nitrogen from the leaves. However, future studies should verify the leaching rates and the contents of carbon and nitrogen of the leaves indicated by the present study through further statistically significant measures considering the timing of leaf emergence, when the leaves were harvested (growing season and non-growing season). In addition, it is necessary to clear the chemical contents of soils in which plants have been growing, because materials absorbed from their roots tend to be accumulated in the plants, leaching when submerged.

4-3. Sugar-leaching characteristics of Q. glauca leaves

Although leachates from leaves contain a variety of chemicals, sugars are one of the major abundant forms of DOM leached from leaves. In *S. gracilistyla* leaves, the total amount of sugar leached within 24 h after leaf imersion was accounted for approximately 40 to 70 (%) of the total amount of sugar that leached from the leaves after 14 days. In addition, peak levels of sugar in the leachate were observed after 5 to 10 days[28]. Similarly, 70 to 80 (%) of the sugars in leaves of white oak and hickory leaves are leached during the first two weeks of immersion[6]. Compared to these rapid sugar-leaching rates in deciduous leaves, the rates from the evergreen leaves of *Q. glauca* were markedly slower and relatively little sugar leached within the first few days after immersion. Peak sugar concentrations in water containing either old or new leaves were observed after 10 and 30 days, respectively (Fig.4). These findings show that *Q. glauca* leaves have a longer sugar-leaching period than *S. gracilistyla*. In addition, *Q. glauca* leaves leached sugars less rapidly than *S. gracilistyla* leaves.

In *S. gracilistyla* leaves, carbohydrates produced by photosynthesis are considered to be locked in the cuticles and epicuticular waxes of the leaves during the growth period. However, in the senescence period, carbohydrates in leaves are broken down into simple sugars, such as sucrose, and the biomembranes become permeable to solublecarbohydrates[28]. Contrary to this finding, the present results show that, in new and old leaves of *Q. glauca*

Table 3. Ratios of total sugar concentrations and TC over time for different water treatment groups

Submerged period (day)	total sugar concentrations (ppm)					TC concentrations (ppm)					total sugar / TC (%)				
	1	5	10	20	30	1	5	10	20	30	1	5	10	20	30
old leaves with aeration	0.03	3.36	3.37	0.97	0.37	19.29	49.05	50.06	65.98	71.23	0.1	6.8	6.7	1.5	0.5
old leaves without aeration	0.89	0.24	13.12	11.22	8.79	15.85	52.75	45.19	52.50	51.62	5.6	0.5	29.0	21.4	17.0
new leaves with aeration	2.44	1.50	0.07	1.14	1.81	24.06	51.58	55.51	67.77	75.17	10.1	2.9	0.1	1.7	2.4
new leaves without aeration	2.20	2.21	1.75	0.94	26.07	21.06	57.12	54.55	55.49	56.56	10.4	3.9	3.2	1.7	46.1

harvested during the growing period, more than 60 (%) of total sugar leached from the leaves by the 30 day after immersion in water was sucrose (Fig.5).

During the experimental period, the TC in each treatment increased to 50 (mg/l) within the first 5 days, as carbon was leached from the leaves of all samples [17]. After 5 days, TC concentrations remained constant in non-aerated water and increased after day 10 in aerated water, regardless of whether the leaves were new or old. However, considerable differences in the amount of TC that had leached into the water were observed among treatments by the end of the experiment (old leaves with aeration, 71.2 (mg/l); old leaves without aeration, (51.6 mg/l); new leaves with aeration, 75.2 (mg/l); new leaves without aeration, 56.6 (mg/l)). The total sugar concentration/TC ratios that were measured over the course of the experiments are shown in Table3. The table shows that after 30 days, the total sugar/TC ratios in the leachate ranged from 0.5 and 2.4 (%), and 17 and 46 (%), with and without aeration, respectively. These findings indicate that the ratio of total sugars to TC in *Q. glauca* leaves fluctuates depending on whether or not the water is aerated.

The major agents for removal of sugars from the water may be water temperature, microbial activity in the water, and oxygenation. In the present experiments, only very low concentrations of sugars were detected in each sample after 10 days of immersion under aerated conditions. The reason for this may be because the sucrose that leached from *Q. glauca* leaves may have been oxidized and converted into water and carbon dioxide (see reaction (**1**) below). Consequently, the aerated condition may not prevent sugars from leaching from the leaves, but rather because the sucrose in the leachate was oxidized by aeration. The reactions showing how water-soluble sucrose (disaccharide) was broken down into glucose and fructose (simple sugars) by hydrolysis is shown in equations (**2**) and (**3**) below. However, detailed analysis is required in order to better understand the release of sugar from *Q. glauca* leaves.

$$C_{12}H_{22}O_{11} + 12O_2 \rightarrow 11H_2O + 12CO_2 \quad (\mathbf{1})$$

$$C_{12}H_{22}O_{11} + H_2O \rightarrow C_6H_{12}O_6 + C_6H_{12}O_6 \quad (\mathbf{2})$$

$$C_6H_{12}O_6 + 6O_2 \rightarrow 6H_2O + 6CO_2 \quad (\mathbf{3})$$

As mentioned above, the rate of sugar release from *Q. glauca* during the first 5 days was relatively slow compared to rates of leaching in *S. gracilistyla* leaves. The time taken for total sugar concentrations to peak after starting the experiment was approximately 5 days in leaves of *S. gracilistyla*, and from 10 to 30 days in *Q. glauca* [30]. In the deciduous leaves of the vine maple and alder, a rapid decrease in leaf mass occurs primarily by decomposition of the most labile constituents, including soluble carbohydrates (such as sugars), soluble protein, hemicellulose, and additional soluble organic matter [10], while the decrease in leaf mass proceeds more slowly in evergreen leaves. Differences in the release patterns of total sugar between deciduous and evergreen leaves might arise due to differences in leaf composition and structure. Sugars are derived from soluble carbohydrates that are lost through the cuticle of evergreen leaves; however, the relative thickness of the evergreen leaf cuticle reduces the rate of sugar release. The tough outer surfaces of evergreen leaves (e.g., cuticle) may also slow rate at which the mass of the leaf decreases [17], particularly until such time as the outer layer of the epidermis, which contains cutin, breaks down in water. The release rate of total sugars from leaf litter submerged in water and subsequent decomposition into water and carbon dioxide may be increased by abrasion of leaf surfaces.

5. CONCLUSIONS

The initial carbon content of evergreen *Q. glauca* leaves appears to be relatively higher than that of deciduous leaves because of the tough outer surfaces of evergreen leaves (e.g., cuticle). After leaves of *Q. glauca* enter streams in runoff, water soluble carbohydrates such as sugars, which consist of cutin, are leached over a relatively longer period than in deciduous leaves. Thus, the sugars in the tougher evergreen *Q. glauca* leaves take longer to leach than they do from the softer deciduous leaves. Similarly, the decrease in the mass of submerged *Q. glauca* leaves is slower than it is in the softer evergreen leaves of deciduous trees.

In *Q. glauca*, the amounts and rates of carbon and nitrogen components leached from the leaves are not influenced by the initial carbon and nitrogen contents of the leaves. This implies that more components are leached from leaves after leaf immersion during the initial leaching period of two weeks regardless of the initial carbon and nitrogen contents of the leaves. In this study, no significant difference was observed in the initial carbon content between new and old *Q. glauca* leaves, or in sun and shade leaves.

Although numerous studies have been conducted on submerged leaves in deciduous species to estimate the nitrogen dynamics in the leaves during leaching and decomposing period [10, 21, 29], few of these studies have considered the possible role of sunlight exposure of the leaves and how this impacts leaching. However, in *Q. glauca* leaves, the initial nitrogen content of the leaves appears to be influenced by the extent to which the leaves were exposed to sunlight. Further experiments considering the potential contribution of abiotic factors at the growing sites should therefore be undertaken.

DOM is leached from any canopy leaves and leaf litter on the forest floor, as well as from leaves that have been transported to streams in runoff; all of the these sources of DOM affect stream water quality. It is therefore that further experiments be undertaken to more accurately clarify the release and uptake of nutrients in leaching and decomposing process of leaves.

ACKNOWLEDGMENTS

This research was supported in part by Grant-in-Aid for Scientific Research(C)(215801860001) from the Ministry of Education, Sports Culture, Science and Technology.

REFERENCES

1) **Allan, J.D.** (1995) Stream ecology. Chapman and Hall, New York.

2) **Body, C.E.** (1970) Losses of mineral nutrients during decomposition of Typha latifolia, *Arch.Hydrobiol* **66-4**: 511-517.

3) **Brian, H.H.** (1979) Uptake and release of nutrients by aquatic macrophytes. *Aquatic Botany* **7**: 87-93.

4) **Cummins, K.W., Wilzbach, A.M., Gates, D.M., Perry, J.B. and Taliaferro, W.B.** (1989) Shredders and riparian vegetation. *Bioscience* **39-1**: 24-30 .

5) **Elizabeth, E.L. and Robert, A.B.** (1987) The global water cycle. Prentice-Hall, Inc., New Jersey.

6) **Eric, C.** (1987) Changes in the chemical composition of alder, poplar and willow leaves during decomposition in a rive. *Hydrobiologia* **148**: 35-44.

7) **Hodkinson, I.D.** (1975) Dry weight loss and chemical changes in vascular plant litter of terrestrial origin, occurring in a beaver pond ecosystem. *Journrl of ecology* **63**(**1**): 131-142.

8) **Hynes, H.B.** (1960) The biology of pollited waters. Liverpool Univ. press: 1-202.

9) **James, R.S., Frank, J.T. and Nancy, S.T.** (1975) The processing of conifer and hardwood leaves in two coniferous forest stream: Ⅰ. Weight loss and associated invertebrates. *Verh. Internt. Verein. Limnol.* **19**: 1617-1627.

10) **James, R.S., Frank, J.T. and Nancy, S.T.** (1975) The processing of conifer and hardwood leaves in two coniferous forest stream: Ⅱ. Biochemical and nutrient changes. *Verh. Internt. Verein. Limnol.* **19**: 1628-1639.

11) **Kaushik, N.K. and Hynes, H.B.N.** (1971) The fate of the dead leaves that fall into streams. *Arch. Hydrobiol.*: 465-515.

12) **Keith, V.S. and Washington, D.C.** (1964) Effect of leaves on water Quality in the Cacapon river. Art.161 in U.S. GEOL. Survey Prof. 475-D: 181-185.

13) **King, J.M., Day, J.A. and Davies, B.R.** (1987) Leafpack dynamics in a southern African mountain stream. *Freashwater Biology* **18**: 325-340.

14) **Kitamura, H.** (1995) Supply and transfer of oxygen in natural stream water body by reaeration, *Journal of the Japan Society of Erosion Control Engineering* **48-4**, 11-19, (in Japanese with English abstract).

15) **Kitamura, H.** (2006) Estimation of cation and anion leaching characteristics and dissolved BOD-adsorption capacity of homemade charcoal produced by community participation. *Bulletin of Minamikyushu University* **36A**: 7-19.

16) **Kitamura, H.** (2007) An extreme decrease in dissolved oxygen concentrations resulting from litter decomposition in a mountain stream in the Southerm Kyusyu Mountain. *Bulletin of Minamikyushu University* **37A**: 57-68 (2007).

17) **Kitamura, H.** (2010) A case study on Leaching characteristics of *Quercus glauca* leaves in Southern Kyusyu stream water. *Bulletin of Minamikyushu University* **41A**: 43-52.

18) **Likense, G.E., Bormann F.H, Johnson M.N., and Pierce R.S.** (1967) The Calcium, magnesium, potassium, and sodium budgets for a small forested ecosystem. *Ecology* **48-5**: 772-785.

19) **Lousier, J.D. and Parkinson, D.** (1975) Litter decomposition in a cool temperate deciduous forest. *Can. J. Bot.* **54**: 419-436.

20) **Lousier, J.D. and Parkinson, D.** (1978) Chemical element dynamics in decomposing leaf littetr. *Can. J. Bot.* **56**: 2795- 2812.

21) **Mark, M.B.** (1977) Decomposition and nutrient exchange of litter in an alluvial swamp forest. *Ecology* **58**: 601- 609.

22) **Maurice, A.L. and Hynes, B.N.** (1976) The fate of "dissolved" organic carbon derived from autumn-shed maple leaves (Acer saccharum) in a temperate hard-water stream. *Limnology and oceanography* **21-3**: 436-443.

23) **McDowell, W.H. and Fisher, S.G.** (1976) Autumnal processing of dissolved organic matter in a samll woodland stream ecosystem. *Ecology* **57**: 561-569.

24) **Meyer, J. and Johnson, C.** (1983) The influence of elevated nitrate concentration on rate of leaf decomposition in a stream. *Freshwater Biology* **13**: 177-183.

25) **Meyer, J.L.** (1980) Dynamics of phosphorus and organic matter during leaf decomposition in a forest stream. *IKOS*. **34**: 44-53.

26) **Nykvist, N.** (1963) Leaching and decomposition of water-soluble organoic substances from differnt type of leaf and needle litter. *Studia Forestalia Suecica* **3**: 3-29.

27) **Omori, M.** (2009) Photosynthesis and respiration, Asakura Publishing Co., Ltd, Tokyo, 145.

28) **Photosynthesis Education Team** (2007) Science of Photosynthesis, University of Tokyo Press,Tokyo, 291.

29) **Robert, C.P. and Kenneth, W.C.** (1974) Leaf processing in a woodland stream. *Freshwater Biology* **4**: 343-368.

30) **Sasaki, A., Shikenya, S., Takeda, K. and Nakatsubo, T.** (2007) Dissolved organic matter originating from the riparian shrub Salix gracillistyla. *T. For Res.* **12**: 68-74.

31) **Suberkropp, K., Godshalk, G.L., and Klug, M.J.** (1976) Change in the chemical composition of leaves during processing in a woodland stream. *Ecology* **57**: 720-727.

32) **Webster, J.R. and Benfield, E.F.** (1986) Vascular plant breakdown in freshwater ecosystems. *Ann. Rev. Ecol.* **17**: 567-594.

北村泰一
南九州大学環境園芸学部環境園芸学科
緑地保全学研究室

要　約

河畔域から渓流に供給されるリーフリターは膨大な量にのぼると推測され，その溶出成分は渓流水質の形成に重要な役割を果たすものと思われるが，落葉広葉樹リーフリターの分解に伴う重量損失とリター構成成分の変化については知られ

ているが，常緑樹リーフリターの溶出特性については不明な点が多い．そこで本研究は，南九州河畔域に広く分布する常緑広葉樹のアラカシリーフリターの溶出初期における重量損失と炭素・窒素放出特性を考察した前年度研究に引き続き，溶出初期におけるアラカシリーフリターの炭素・窒素含有量と糖類溶出特性を明らかにすることを目的とした．

2010年5月，一ツ瀬川支流竹尾川下流（宮崎県西都市）の河畔域に生育するアラカシの当年葉（2010年春以降に形成された葉）と一年葉の陰葉と陽葉を採取し，十分な洗浄・乾燥と重量測定の後，各グループの葉10 枚を無作為に選定し，葉央部の主脈脇から2mg の破片に含まれる炭素および窒素の含有量を測定した．この測定と並行して1,000（CC）の現地渓流を満たした50 個のビーカーに5 枚ずつ投入してサブサンプルとし，温度調整をしない実験室に置いた．このうち25 個のビーカーには市販ポンプにより酸素を供給し続けた．葉を投入してから1，5，10，20，30 日後に，各実験区から無作為にサブサンプルを5 個づつ回収し，葉の乾燥重量を計測し投入水に含まれる糖類濃度を測定した．

炭素含有量は45.75 ～ 46.86（%），窒素含有量は1.83 ～ 2.16（%）で，これらは落葉樹リーフリターでの既往報告値よりも高い数値であった．炭素含有量は葉のグループ間で有意な違いはなかったが，窒素含有量は一年生陰葉と他のグループとの間に有意な違いが認められた．葉の炭素および窒素含有量の多寡は，葉からの炭素および窒素溶出量に影響しないことが推測された．糖類については，スクロース，グルコース，フルクトースが検出され，全糖類の60％ はスクロースであった．全糖類濃度は投入から30日後に最大値26（mg/l）を示し，アラカシリーフリターからの糖類溶出期間は，落葉樹の既往報告結果と比較して長いことが判明した．また，糖類の溶出とその後の分解は酸素供給により促進された．

アラカシリーフリターと落葉樹リターとの違いは，葉の構造と組成の違い，とりわけアラカシリーフリターの表面を厚く覆うクチン質に起因する可能性があると推論した．