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Spatial and Seasonal Variations in Leaf Mass per Area and Their Relationship to Leaf Nitrogen in a Secondary Northern Hardwood Forest in Japan

By

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Summary

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Photosynthetic capacity correlates closely with leaf mass per area (LMA) and nitrogen content in leaves. To extrapolate from leaf to canopy photosynthesis, vertical and seasonal variations in LMA and nitrogen content per leaf mass (*Nm*) were studied within the canopy of a secondary hardwood forest in northern Japan. The forest was regenerated in 1912 after forest fire and is now an uneven-aged mixed forest with a density of 672 trees per hectare. The canopy over-story is composed of *Betula platyphylla* var. *japonica*, *Quercus mongolica* var. *grosseserrata*, *Kalopanax septemlobus*, *Populus maximowiczii* and *Tilia japonica*, in order of decreasing predominance. We studied the early successional species *B. platyphylla* and the mid-successional species *Q. mongolica* and *K. septemlobus*. The vertical variation in LMA and *Nm* in these three species was measured by destructive sampling in the summer of 2001 and 2003. LMA correlated closely but non-linearly with the irradiance within the canopy; this relationship was different for each of the three species. In a subsequent experiment in 2002, seasonal changes in LMA and *Nm* were measured for leaves of the tree species at different heights, using a scaffold. In all three species, LMA and *Nm* were steady from mid-June to early September. Linear regression analysis indicated that the nitrogen content per leaf area (*Na*) of mature leaves correlates with LMA ($p < 0.001$), even when *Nm* is constant ($p > 0.05$) within the canopy. The mean value of *Nm* was significantly smaller in *B. platyphylla* than in the other two species ($p < 0.01$). We concluded that vertical variations in *Na* for mature leaves of each species within the canopy can be estimated by LMA partitioning by irradiance and a constant value of *Nm*.

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Introduction

The lowland forests of Hokkaido, the northernmost part of Japan, represent a boreal transition between northern hardwoods and coniferous forests. Much of this forest is being replaced with birch-dominated secondary forests accompanied by original hardwood species tolerant of shade. In modeling the canopy photosynthesis of such mixed forests, it is important to understand the light variations within the canopy as well as the light-related modifications in leaf morphology and photosynthesis of each species. The light-saturated photosynthetic rate (Pmax) of mature leaves correlates closely with leaf nitrogen content (N) and leaf mass per area (LMA) (ELLSWORTH & REICH 1993, REICH & al. 1995, KULL & NIINEMETS 1998, BOND & al. 1999). Mass-based N (*Nm*) positively correlates with mass-based Pmax. The correlation differs by species, and this inter-species difference in the correlation between mass-based N and Pmax depends on LMA. (REICH & al. 1998).

Within a species, area-based Pmax (Pmax/a) declines with canopy depth (HOLLINGER 1989). Pmax/a in mature leaves usually depends on the area-based nitrogen content (*Na*). LMA of mature leaves varies widely within a canopy and correlates positively with irradiance in the canopy (e.g., ANTEN & al. 1998). In contrast, *Nm* is relatively constant within the canopy (HOLLINGER 1996, NIINEMETS 1997, ROSATI & al. 2000). While other studies reported that *Nm* increases or decreases with irradiance in different species (e.g., KULL & NIINEMETS 1993).

Since the value of *Na* is the product of LMA and *Nm* (*Na*=LMA×*Nm*), spatial and seasonal variations in Pmax/a within and among species are characterized by a quantitative relationship between LMA and *Nm*. The present study aims to examine whether LMA and *Nm* in a forest canopy are species-dependent. We compared the three species that predominate at the study site: *B. platyphylla*, an early successional species, and *Q. mongolica* and *K. septemlobus*, mid-successional species. The comparison was in terms of (1) seasonal variations in LMA and *Nm* (to clarify the leaf maturation periods), (2) variation in LMA and *Nm* of mature leaves within a canopy, (3) variation in *Na* along the irradiance for each species.

Material and Methods

The study was conducted in the experimental forest of the Forestry and Forest Products Research Institute (42°59'N, 141°23'E; 180 m a.s.l), in Sapporo, Japan. Mean annual precipitation is 957 mm and mean annual temperature is 7.1°C. The distribution of species was assessed through an inventory of a 50 × 50-m plot. Predominant species are *Betula platyphylla* var. *japonica*, *Quercus mongolica* var. *grosseserrata*, *Kalopanax septemlobus*, *Populus maximowiczii* and *Tilia japonica*, in order of decreasing predominance. The first two species comprise more than 75% of the plot's basal area. In 2003 the tree density was 672 stems per hectare and the stand age about 91 years. Mean tree height was 18.3 m and mean stem diameter at 1.3 m was 23 cm. Three canopy species were used: *B. platyphylla*, an early successional species, and *Q. mongolica* and *K. septemlobus*, both middle successional species.

In the summer of 2001 and 2003, 8 sample trees of each species (24 trees total for each summer) were felled at 0.3 m above the ground. Each tree was cut at 1-m intervals from the felling point, and the canopy was cut into strata, each 1 m in length. Sample leaves (about 1000 cm^2) were taken from each stratum and leaf area was measured with a leaf area meter (Li-3000 area meter

Li-Cor Inc., Neb., U.S.A.). The leaves were oven-dried at 70°C, and the leaf mass per area (LMA; gm^{-2}) was determined for each height. Total nitrogen content of each sub-sample (about 200 cm^2) was determined using an NC-analyzer (Sumigraph NC-80, Shimazu, Kyoto, Japan). Nitrogen content was calculated on both a mass base (*Nm*; %) and an area base (*Na*; gm^{-2}, $Na=Nm\times LMA$).

We constructed a 26-m-high scaffold to access the leaves of the three species. Five to 10 leaves were collected at different heights in the canopy: 23 m and 18 m for *B. platyphylla*, and 23 m, 18 m and 11 m for *Q. mongolica* and *K. septemlobus*, from May to November 2002. LMA and nitrogen content of each leaf sample were measured.

Photosynthetic photon flux density (PPFD) within the canopy was measured with PPFD sensors (Li-190SA Li-Cor Inc., Neb., U.S.A.) at 1-m height intervals from 1 m to 26 m using the scaffold in August 2004. These measurements were carried out under overcast conditions. Relative PPFD (rPPFD; %) for each height was calculated as an average value of 200 replicates, with above and below canopy readings synchronized.

Results and Discussion

Seasonal changes in LMA and *Nm*

LMA and *Nm* of leaves in the upper canopy (23-m in height) showed seasonal variations (Fig. 1). The three species started to develop leaves from early May. LMA increased and *Nm* decreased until mid-June; both became relatively stable from mid-June to early September, when the leaves matured. LMA and *Nm* steadily decreased during October and all the leaves fell by mid-November.

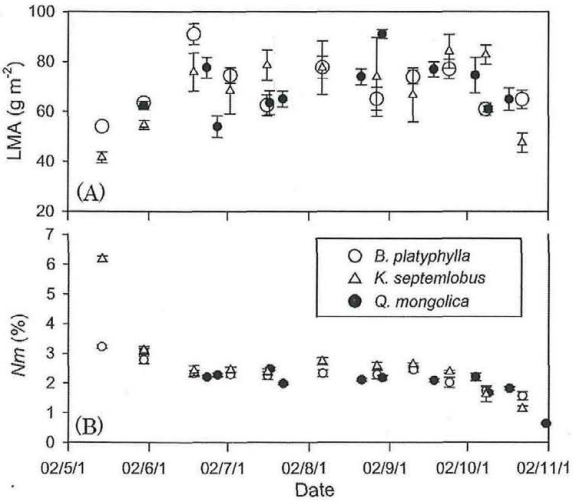

Fig. 1. Seasonal changes in LMA **(A)** and *Nm* **(B)** of upper-canopy leaves (23-m in height) for *B. platyphylla* (○), *K. septemlobus* (Δ) and *Q. mongolica* (●). Error bar means standard error of the mean.

Roughly the same seasonal changes in LMA and *Nm* were observed in lower-canopy leaves (data not shown), indicating no large difference in seasonal variations among the three species. In mature leaves of all species, variations in LMA were larger than in *Nm*. Based on these seasonal variations in LMA and *Nm*, the sample leaves obtained in destructive measurement during the summer (July to August) were regarded as mature.

Variations in LMA and *Nm* of mature leaves within the canopy

In the destructive measurements, LMA decreased with decreasing rPPFD in the three species but there was a considerable dispersion in LMA for *K. septemlobus* (Fig. 2). LMA varied non-linearly with rPPFD, decreasing dramatically at below 10% rPPFD. This non-linearity is consistent with results of other studies (HOLLINGER 1996, BOND & al. 1999). To compare the response of LMA to rPPFD among species, we linearized the relationships by log-transformed rPPFD ($\ln_{rPPFD}$). Correlations between LMA and $\ln_{rPPFD}$ were significant ($p < 0.01$) and these parameters were also significant ($p < 0.01$) for all species (Table 1) though the species differed significantly in this correlation (ANCOVA $p < 0.01$).

Table 1. Regression statistics for three species between LMA and $\ln_{rPPFD}$, using the linear model $LMA=\beta+\alpha\cdot\ln_{rPPFD}$. Asterisk denotes significant at $p < 0.01$. Values not sharing same letter are significantly different at $p < 0.01$. SE means standard error of the mean.

Species	α (SE)	β (SE)	R^2
B. platyphylla	11.0 (0.8) *c	27.0 (2.7) *a	0.8*b
K. septemlobus	16.2 (1.1) *a	14.7 (2.6) *b	0.8*c
Q. mongolica	13.7 (0.5) *b	21.0 (1.3) *c	0.9*a

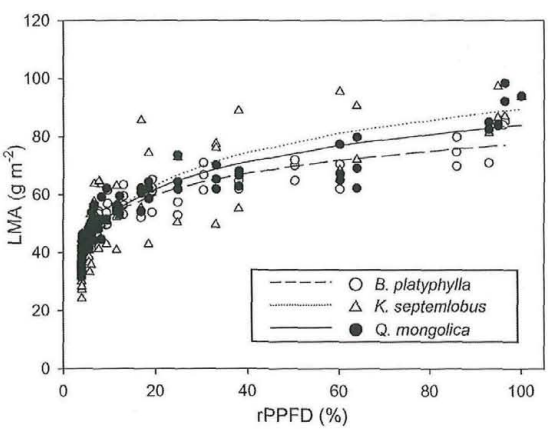

Fig. 2. The relationship between LMA and rPPFD for *B. platyphylla* (○), *K. septemlobus* (Δ) and *Q. mongolica* (●). Linearized regression equations are in Table 1.

There was no significant correlation between *Nm* and LMA ($p > 0.05$), even though *Na* correlated linearly with LMA ($p < 0.001$, and R^2 between 0.77 and 0.98) for each species (Fig. 3). The intercept of the relationship between *Na* and LMA was not significantly different from zero ($p > 0.01$) for each species. These results indicate that, because *Nm* is constant within the canopy, the variation in *Na* within the canopy is a direct consequence of the vertical partitioning of LMA in each species. However, the mean value of *Nm* for *B. platyphylla* (2.16 standard error; ±0.021) was significantly smaller than those for *Q. mongolica* (2.34 ±0.022) and *K. septemlobus* (2.38 ±0.016) (Scheffe Post Hoc test $p < 0.01$).

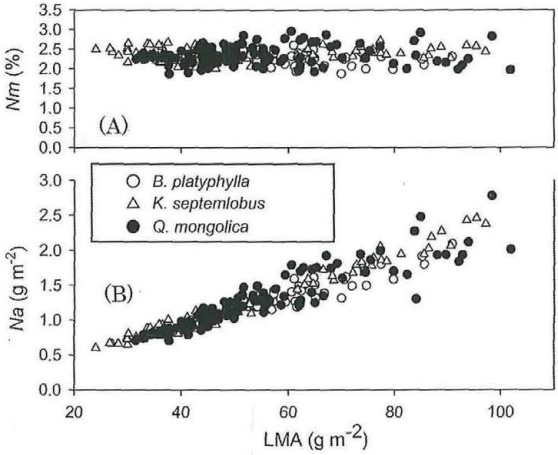

Fig. 3. **(A)** the relationship between LMA and *Nm*, and **(B)** the relationship between LMA and *Na*, both for *B. platyphylla* (○), *K. septemlobus* (Δ) and *Q. mongolica* (●). *Na* was significantly related to LMA for each species ($p < 0.001$).

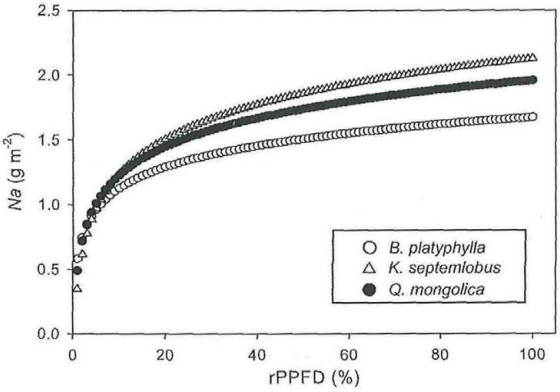

Fig. 4. The relationship between calculated *Na* and rPPFD for *B. platyphylla* (○), *K. septemlobus* (Δ) and *Q. mongolica* (●). The calculation method is shown in the text.

Relationships between *Na* and rPPFD were obtained using the LMA-rPPFD relationships and mean value of *Nm* of each species. Except in very dark positions, *Na* followed the order (highest to lowest) of *K. septemlobus, Q. mongolica,* and *B. platyphylla* (Fig. 4). The non-linear relationship between *Na* and rPPFD in each species indicates that a canopy photosynthetic model should consider the effect of carbon cost (e.g., leaf construction respiration) on *Na* distribution (HOLLINGER 1996, BOND & al. 1999), because the optimal distribution of *Na* should be directly proportional to rPPFD when rPPFD is a predominant factor controlling *Na* (ANTEN & al. 1995).

We conclude that both LMA and *Nm* cause the difference in vertical variation of *Na* between light-demanding *B. platyphylla* and the two shade-tolerant species. The variation of LMA within the canopy results in the difference in *Na* partitioning between *K. septemlobus* and *Q. mongolica.*

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References

ANTEN N.P.R., SCHIEVING F. & WERGER M.J.A. 1995. Patterns of light and nitrogen distribution in relation to whole canopy carbon gain in C_3 and C_4 mono- and dicotyledonous species. - Oecologia 101: 504-513.

— , MIYAZAWA K., HIKOSAKA K., HAGASHIMA H. & HIROSE T. 1998. Leaf nitrogen distribution in relation to leaf age and photon flux density in dominant and subordinate plants in dense stands of a dicotyledonous herb. - Oecologia 113: 314-324.

BOND B.J., FARNSWORTH B.K., COULOMBE R.A. & WINNER W.E. 1999. Foliage physiology and biochemistry in response to light gradients in conifers with varying shade tolerance. - Oecologia 120: 183-192.

ELLSWORTH D.S. & REICH P.B. 1993. Canopy structure and vertical patterns of photosynthesis and related leaf traits in a deciduous forest. - Oecologia 96: 169-178.

HOLLINGER D.Y. 1989. Canopy organization and foliage photosynthesis capacity in a broad-leaved evergreen montane forest. - Funct. Ecol. 3: 53-62.

— 1996. Optimality and nitrogen allocation in a tree canopy. - Tree Physiol. 16: 627-634.

KULL O. & NIINEMETS U. 1993. Variations in leaf morphometry and nitrogen concentration in *Betula pendula* Roth., *Corylus avellana* L. and *Lonicera xylosteum* L. - Tree Physiol. 12: 311-318.

— & NIINEMETS U. 1998. Distribution of leaf photosynthetic properties in tree canopies: comparison of species with different shade tolerance. - Funct. Ecol. 12: 472-479.

NIINEMETS U. 1997. Distribution patterns of foliar and nitrogen as affected by tree dimensions and relative conditions in the canopy of *Picea abies*. - Trees 11: 144-154.

REICH P.B., KLOEPPEL B.D., ELLSWORTH D.S. & WALTERS M.B. 1995. Different photosynthesis-nitrogen relations in deciduous hardwood and evergreen coniferous tree species. - Oecologia 104: 24-30.

— , ELLSWORTH D.S. & WALTERS M.B. 1998. Leaf structure (specific leaf area) modulates photosynthesis-nitrogen relations: evidence from within and across species and functional groups. – Funct. Ecol. 12: 948-958.

ROSATI A., DAY K.R. & DEJONG T.M. 2000. Distribution of leaf mass per unit area and leaf nitrogen concentration determine partitioning of leaf nitrogen within tree canopies. - Tree Physiol. 20: 271-276.