



Title	Growth and Photosynthetic Performance of Seedlings of Two Larch Species Grown in Shaded Conditions
Author(s)	Qu, Laiye; Ji, Donghun; Shi, Fuchen; Sasa, Kaichiro; Koike, Takayoshi
Citation	Eurasian Journal of Forest Research, 8(1), 43-51
Issue Date	2005-02
Doc URL	http://hdl.handle.net/2115/22189
Type	bulletin (article)
File Information	8(1)_P43-51.pdf



[Instructions for use](#)

Growth and Photosynthetic Performance of Seedlings of Two Larch Species Grown in Shaded Conditions

QU Laiye¹, JI Donghun², SHI Fuchen³, SASA Kaichiro⁴ and KOIKE Takayoshi^{4*}

¹ JSPS Fellow, Hokkaido University Forests, FSC, Sapporo 060-0809, Japan.

² Graduate School of Agriculture, Hokkaido University, Sapporo 060-8589, Japan.

³ College of Life Sciences, Nankai University, Tianjin 300071, P.R. China.

⁴ Hokkaido University Forests, FSC, Sapporo 060-0809, Japan.

Abstract

To study the regeneration pattern of the Japanese larch (*Larix kaempferi* Sarg.) and its hybrid larch (*Larix gmelinii* × *L. kaempferi*), we compared the growth and photosynthetic performance of two larch seedlings under differing light levels (8%, 16%, 32% shade conditions, with 100% light as control) and fertilizer regimes (5mg and 25mg per seedling). The total dry mass of the shaded seedlings was smaller than for the control seedlings after 12 weeks of the growing period in both larch species. The lowest needle area was observed in seedlings grown under 8% light level. The specific leaf area (SLA, needle area per unit dry mass) of seedlings grown at 100% light was significantly lower than in seedlings grown in 8% light. However, there was no significant difference in dry mass allocation for either larch species in the two nutrient treatments. Fertilizer regimes did not affect the needle area of either species. The total chlorophyll content per unit leaf area increased with increasing irradiance, due mainly to the reduction in SLA. Seedlings of both species grown at 100% light have a significantly higher light-saturated rate of net CO₂ assimilation (A_{max}) and higher V_{cmax} (maximum rate of carboxylation) than for the other light treatments given the same fertilizer level. The Japanese larch displayed greater changes in response to decreasing PPFD, as a result of its remarkable change in SLA. However, Japanese larch seedlings treated with high nutrient fertilizer were suppressed in shady conditions. These results suggest that the hybrid larch would be more dominant in slightly shaded conditions on infertile soil.

Keywords: Biomass allocation, hybrid larch, Japanese larch, nutrient, shade

Introduction

The Japanese larch (*Larix kaempferi* Sarg.) is most common in central Japan. It was introduced into Hokkaido Island for timber production. However, the Japanese larch has been suffering from the disease of shoot blight and grazing damage by wolves. The hybrid larch (*Larix gmelinii* × *L. kaempferi*) was developed to withstand such biological damage (Koike *et al.* 2000). The larch is a light-demanding deciduous conifer (Gower and Richards 1990, Koike *et al.* 2000). Irradiation is an essential ecological factor, on which all photoautotrophic plants depend (Lambers *et al.* 1998). Light energy directly drives many fundamental plant and biophysical processes, including photosynthesis, stomatal conductance, transpiration, and leaf temperature. Light energy also regulates many secondary plant features such as plant growth, seedling regeneration, the vertical structure and crown shape of forest stands, and the uptake and emission of trace gases involved in biogeochemical cycling and atmospheric chemistry.

With the development of leaves on canopy trees, the capability for seedling regeneration at the forest floor is reduced. To understand the regeneration of larch species on Hokkaido Island it is necessary to investigate capacity of the two species of larch

seedlings to grow in various light conditions now that many adult trees are reaching the age of reproduction. We focus on the natural regeneration of larch plantations because of its advantages. Natural regeneration is less expensive than planting, for example, since less labor and heavy equipments are needed. Furthermore, there is no problem in natural regeneration with the geographical origin of seed, so that the species and trees are well adapted to the site. Also, new seedlings have better root systems, or "natural" root morphology, than planted seedlings.

The photosynthetic apparatus of plants is remarkably adaptable to a wide range of light conditions. When a canopy gap forms, understory plants receive high irradiance. For the gap to close, the plants must adjust to the low availability of light. Survival in the shaded understory demands maximization of light capture for photosynthesis while minimizing the loss of energy and carbon in respiration. By contrast, leaves exposed to strong light must make efficient use of the available energy while avoiding loss of photosynthates by photoinhibition or other environmental stresses. The ability to balance these conflicting factors depends on other environmental factors such as nutrient availability and temperature, which often accompany changes in light availability.

Plants can respond to their light environment at several integration levels. First, they can change the fraction of biomass invested in leaves, stems and roots. Second, they are able to modulate the leaf area per unit biomass invested in leaves (ie, the specific leaf area or SLA), by altering their anatomical structure. Third, they can change the relative investment of nitrogen among photosynthetic components (e.g. Evans and Poorter 2001). We are interested in how larch species adapt their regeneration to a shaded environment. Under different soil fertility conditions, which of the Japanese larch or hybrid larch will be dominant?

We examined the combined effect of light and nitrogen level on young seedlings of the two larch species. We discuss the role of the light environment in the growth of seedlings of the two larch species under different soil fertility conditions, to investigate the possible regeneration pattern of the two species.

Materials and methods

1. Experiment site and design

The experiment was conducted at the greenhouse belonging to the Faculty of Agriculture, Hokkaido University. The experiment involved four light regimes providing 8%, 16%, and 32% of full sunlight as well as an unshaded, full sunlight control (100% rPPFD). At each light treatment there were two levels of soil fertilization, 5mg and 25mg, for each of the two species (low and high fertilizer treatment, in the terminology of Qu *et al.* 2003).

2. Plant materials and establishment

Seeds of the Japanese larch and its hybrid (F1) were obtained from the Uryu Experimental Forest of Hokkaido University, which is located 230 km north of the city of Sapporo. The seeds were maintained at 4°C for 10-days (cold treatment) and were then individually germinated in pots with the soil media (clay-loam soil, peat moss and vermiculite, 2:2:1 by volume) for 30 days. During this 30-day culture period, the Japanese larch and hybrid larch seedlings received fertilizer via a mixed nutrient solution (N-P₂O₅-K₂O 15:15:15). JH (Japanese larch, high nutrient treatment) received 25mg N per seedling. JL (Japanese larch, low nutrient treatment) received 5mg N per seedling. Likewise, HH (hybrid larch, high nutrient treatment) received 25mg N per seedling, and HL (hybrid larch, low nutrient treatment) received 5mg N per seedling. Sixty pots were then placed outside on a table, either uncovered (100% relative photosynthetically active photon flux (rPPFD)), or covered with a black shade cloth (32% rPPFD, 16% rPPFD or 8% rPPFD, respectively). The maximum solar irradiance of the 100% rPPFD treatment on sunny days was about 1500 μmol m⁻²s⁻¹, similar to the light conditions in an open canopy gap (Matsuki *et al.* 2003). The 32%, 16% and 8% (about 480 μmol m⁻²s⁻¹, 240 μmol m⁻²s⁻¹ and 120 μmol m⁻²s⁻¹) treatments simulated the light conditions of large, middle and small canopy gaps.

3. Harvest and chemical analysis

The shade treatments were continued for 12 weeks to examine the growth and establishment processes of the

two larch species. 10 seedlings from each treatment of each species were harvested at four-week intervals. All seedlings harvested were separated into leaves, stems and roots. The root area and needle area were determined using fresh roots and needles by means of an Area Meas (Hongu, Akinori MYKA. Lab.1.01 Ver, 1995). Finally, the parts were dried to a constant mass at 60°C and weighed. The samples were milled and homogenized. The chlorophyll content was determined by the dimethylsulfoxide (DMSO) method described by Shinano *et al.* (1996). The N concentrations of the needle, stem and root were analyzed using a N-C analyzer (NC900, Shimadzu, Kyoto, Japan).

4. Leaf gas exchange

CO₂ assimilation rates were measured using a Li-Cor photosynthesis system (LI-6400, Lincoln, NE, USA). The light-saturated rate of CO₂ assimilation was measured at an ambient CO₂ concentration (360ppm). We preset the PPFD to 1500, 1000, 600, 300, 200, 100 and 0 μmol m⁻²s⁻¹ with a Li-Cor 6400-02B red/blue light source fitted to the leaf corvette. The CO₂ saturated photosynthetic rate was measured at a saturated PPFD of 1000 μmol m⁻²s⁻¹ (Farquhar and Sharkey 1982, Sharkey 1985). The CO₂ concentration was controlled by a Li-Cor LI-6400 CO₂ injection system. We preset the CO₂ concentrations to 360, 100, 0, 360, 600, 1000, and 1500ppm. All measurements were made at ambient conditions of air temperature and relative humidity. For each treatment, three seedlings were measured.

5. Statistical analysis

The mean values of dry mass of the organs, needle areas, nitrogen content, and SLA were analyzed using the nested procedure of the SAS software (SAS Institute, Inc., 1996). Analysis of variance (ANOVA) was used for balanced data, and the general linear model (GLM) for unbalanced data.

Results

1. Growth response

By 12 weeks, the number of needles of the Japanese larch and hybrid larch was clearly influenced by the amount of light (Fig. 1). The number fell with a decrease in irradiance for all seedlings tested. At each light level for each species, there was no significant difference between larches that received fertilizer and those that did not. The increase in needle area was delayed by shade treatment. Compared to the control conditions, seedlings grown under conditions of shade had smaller needle areas. The lowest foliar area was observed for seedlings grown under 8% light levels. Fertilizer regimes did not affect the needle area of either the larch species (Fig. 2). The specific leaf area (SLA, needle area per dry mass) of the seedlings grown in 100% light was significantly lower than that of seedlings grown in 8% light (Fig. 3).

The total dry mass of the shaded seedlings was smaller (P<0.05) than that of the control seedlings at 4, 8 and 12 weeks of growth for both larch species. There were significant differences for the Japanese larch and the hybrid larch at the second and third harvesting

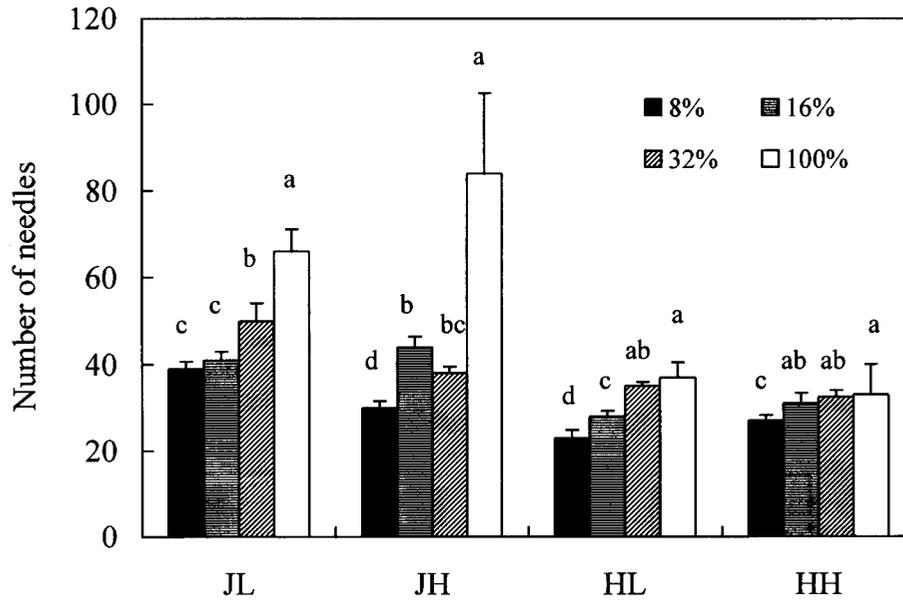


Fig. 1. Needle numbers of Japanese larch and hybrid larch seedlings grown under four light regimes (8%, 16%, 32% and 100%) in combination with two nutrient (5mg and 25mg) treatments. JL = Japanese larch seedling grown under low nutrient treatment (5mg); JH = Japanese larch seedlings grown under high nutrient treatment (25mg); HL = hybrid larch seedling grown under low nutrient treatment level (5mg); HH = hybrid larch seedlings grown under high nutrient treatment (25mg). Vertical bars represent the standard error of its mean ($n = 10$). Values with the same letter (a, b) are not significantly different among light conditions at $P < 0.05$.

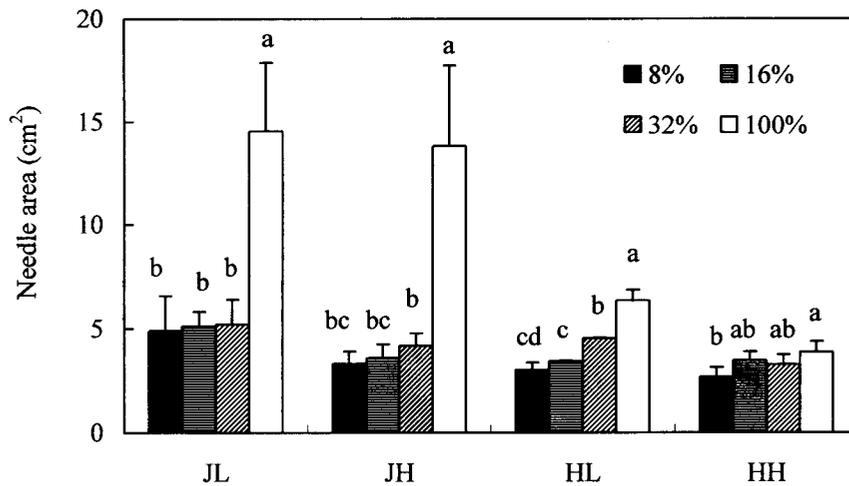


Fig. 2. Needle area (cm²) of Japanese larch and hybrid larch seedlings grown under four light regimes (8%, 16%, 32% and 100%) in combination with two nutrient levels (5mg and 25mg) treatments. Treatment codes are shown in Figure 1. Vertical bars represent the standard error of its mean ($n = 10$). Values with the same letter (a, b) are not significantly different among light conditions at $P < 0.05$.

times ($P<0.05$). The Japanese larch seedlings produced more dry matter than the hybrid larch up to 12 weeks. However, there was no significant difference in dry mass allocation between low and high nitrogen level treatments in either species (Tab. 1).

Figure 4 shows that the seedlings grown under 100% irradiance have a significantly lower shoot/root ratio than those under the other three irradiance conditions, given the same fertilizer regime. Fertilizer increased the shoot/root ratio for both larches in all four light regimes.

2. Partitioning of N

There was a significant difference in N content between the two fertilizer regimes for the Japanese

larch and hybrid larch seedlings ($P<0.05$). The high fertilizer treatment gave a higher content of N in the needle, stem and root organs than the low fertilizer treatment. There was no combination effect between the fertilizer and the amount of light. The N content of the Japanese larch and hybrid larch seedlings were clearly influenced by the shading.

3. Chlorophyll analysis

Figure 2 summarizes the chlorophyll analysis completed at the 12-week mark. The total chlorophyll content per unit leaf area increased with increasing irradiance, with the largest values of 0.33-0.38 for 100% light and the smallest values of 0.17-0.25 for 8% light (Fig. 5). The chlorophyll values for the 32% and

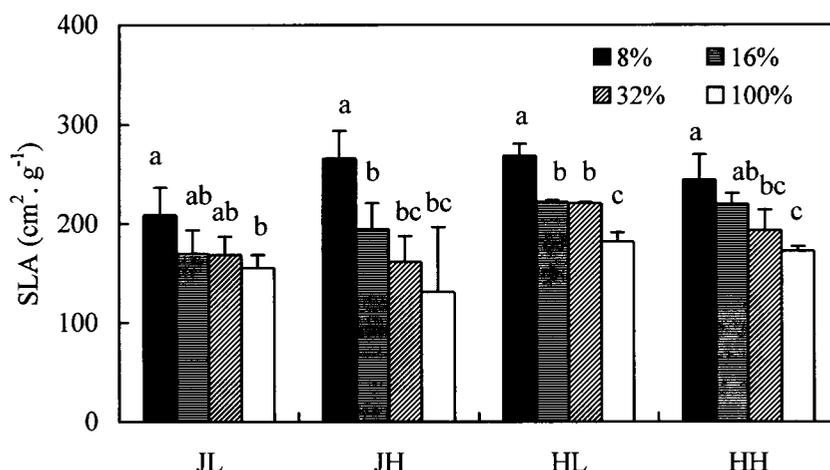


Fig. 3. Specific leaf area (SLA)(cm² g⁻¹) of Japanese larch and hybrid larch seedlings grown under four light regimes (8%, 16%, 32% and 100%) in combination with two nutrient levels (5mg and 25mg) treatments. Treatment codes are shown in Figure 1. Vertical bars represent the standard error of its mean (n = 10). Values with the same letter (a, b) are not significantly different among light conditions at $P<0.05$.

Table 1. Effects of light and nutrient treatments on the dry mass allocation of the seedlings (mg) for the Japanese larch and hybrid larch during the growth period (12 weeks). Values with the same letter (a, b) within row are not significantly different from each other at $P<0.05$. All values are means of ten replicates.

Nitrogen treatments		5mg				25mg			
Light regimes		8%	16%	32%	100%	8%	16%	32%	100%
Japanese larch	4 week	14.8±1.7bc	17.0±0.5ab	17.0±3.7ab	19.1±2.3a	12.7±0.8b	13.1±1.1b	13.2±0.5b	21.1±3.2a
	8 week	43.7±5.4bc	47.4±2.5bc	57.0±2.8b	108.3±10.1a	19.1±2.4d	32.5±4.7bc	43.0±3.1b	95.5±9.3a
	12 week	60.7±5.6bc	69.4±6.1bc	83.8±8.3b	150.2±20.3a	25.8±1.5c	54.7±6.2b	59.3±2.7b	241.2±24.7a
Hybrid larch	4 week	13.7±3.7b	12.6±3.2bc	12.9±0.9bc	18.6±3.7a	12.5±1.1b	11.3±1.4b	9.5±3.2bc	22.1±0.8a
	8 week	28.1±1.7bc	27.4±1.5bc	38.4±5.5b	46.1±6.3a	25.4±2.2bc	23.6±2.7c	29.4±2.0b	50.4±1.8a
	12 week	33.1±4.4c	32.5±6.7c	62.0±5.7ab	77.4±7.9a	35.4±3.5bc	32.6±6.0c	43.0±4.6b	83.6±8.5a

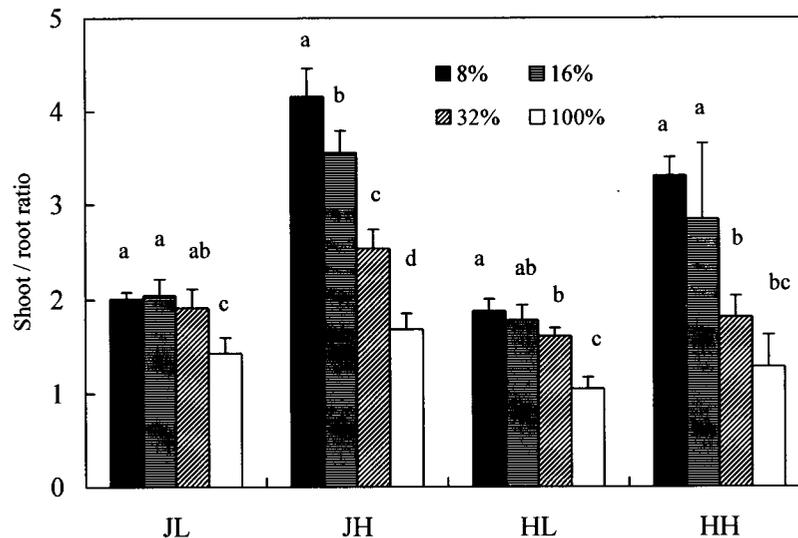


Fig. 4. Shoot / root ratio of Japanese larch and hybrid larch seedlings grown under four light regimes (8%, 16%, 32% and 100%) in combination with two nutrient (5mg and 25mg) treatments. Treatment codes are shown in Figure 1. Vertical bars represent the standard error of its mean ($n = 10$). Values with the same letter (a, b) are not significantly different among light conditions at $P < 0.05$.

the 16% light treatments lie between these extremes. Similarly, the chlorophyll a/b ratios increased with increasing irradiance, from 8% to 100%, for both low and high nutrient levels (Fig. 5).

4. Leaf CO_2 assimilation

By the 12th week, the steady-state rate of CO_2 assimilation had increased to an almost steady value with increasing irradiance. Rates of CO_2 assimilation saturated at a lower PPFD in the shaded seedlings than in the 100% light seedlings (Fig. 6). Seedlings grown in 100% light have a significantly higher light-saturated rate of net CO_2 assimilation (A_{max}) than those at other light treatments at the same level of fertilizer, for both larch species. The maximum rate of carboxylation (V_{cmax}) displays the same pattern as A_{max} . The value of V_{cmax} was higher for the 100% light treatment than in the shaded treatments. However, the A_{max} value of the seedlings treated with high nutrient levels was suppressed in shady conditions. This tendency was more pronounced in Japanese larch seedlings (Tab. 2).

Discussion

We found that CO_2 assimilation in the shaded seedlings tends to saturate at a lower PPFD with lower A_{max} than for the unshaded seedlings (Fig. 6). This indicates that the larch seedlings had a reduced photosynthetic capacity under shady conditions. The reductions in growth and dry mass production were coincident with a reduction in CO_2 assimilation rates in the shaded seedlings (Tab. 1 and Fig. 6). The trend from a high CO_2 assimilation rate in control plants and a low CO_2 assimilation rate in shaded plants is similar to that observed in previous studies (Matsuki *et al.* 2003, Senevirathna *et al.* 2003).

Nitrogen does generally affect photosynthetic capacity (Evens 1989). However, the allocation pattern of leaf nitrogen of larch seedlings was strongly affected by the light environment. In the low nutrient regime, the Japanese larch tended to have a higher

photosynthetic rate than the hybrid larch (though not significantly so). However, in the high nutrient regime the photosynthetic rate of the Japanese larch was clearly suppressed in the shaded treatments. The same effect did not occur in the hybrid larch, however. The larch seedlings which received high nutrient supply had a higher (though not significantly) needle nitrogen content. This high nutrient supply had reduced root growth compared to a low nutrient supply (Fig. 4). It seems that larch seedlings can take up more nitrogen with increasing nutrient availability, to the extent of excess nutrient consumption by larch seedlings (Qu *et al.* 2003). Moreover, excess nitrogen may sometimes become harmful to the growth of seedlings (Bazzaz 1996).

Plants usually need energy that directly drives many fundamental plant and biophysical processes. This high nutrient supply appears to have had a negative effect on the growth of the Japanese larch in the shaded treatments. Plants generally need energy to assimilate nitrate from nitrogen (NO_3^- -N). Therefore, for the larches to assimilate NO_3^- -N, they need energy to first reduce the nitrate nitrogen to the ammonium form of nitrogen (NH_4^+ -N). However, the energy production of plants was suppressed under the shaded conditions; Japanese larch seedlings in the shade could not use NO_3^- -N. The hybrid larch may therefore have a higher capacity to produce more energy used to assimilate NO_3^- -N. The hybrid larch appears to adapt to this high nutrient condition better than the Japanese larch under low light conditions.

Shading increased the shoot/root ratio, which implies an increased biomass fraction to the shoot and a reduced allocation to the roots (Fig. 4). Such a change in allocation maintains a constant transpiration rate per unit root mass (Sims and Pearcy 1994), or may supply the greater demand for nutrients required for faster growth in strong light.

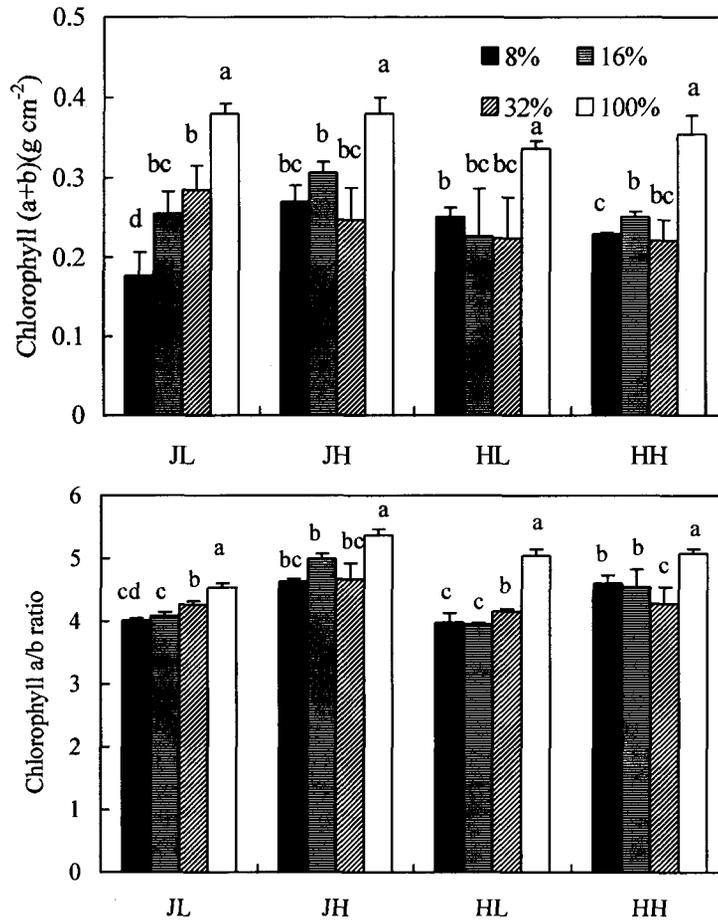


Fig. 5. Chlorophyll (a+b) (g cm⁻²) and chlorophyll a/b ratio of Japanese larch and hybrid larch seedlings grown under four light regimes (8%, 16%, 32% and 100%) in combination with two nutrient (5mg and 25mg) treatments. Treatment codes are shown in Figure 1. Vertical bars represent the standard error of its mean (n = 10). Values with the same letter (a, b) are not significantly different among light conditions at P<0.05.

Table 2. The Mean Values of maximal rate of net photosynthesis at ambient CO₂ (A_{max}, μmol m⁻²s⁻¹) and Maximum rate of carboxylation (V_{cmax}, μmol m⁻²s⁻¹) under various light conditions (8%, 16%, 32% and 100%) and various fertilization levels (5mg and 25mg) of the Japanese and hybrid larch. Values with the same letter (a, b) are not significantly different from each other at P<0.05. All values are means of three replicates.

		Japanese larch		Hybrid larch	
		Nitrogen		Nitrogen	
		5mg	25mg	5mg	25mg
A _{max}	8%	5.23b	1.37b	2.37c	1.05c
	16%	5.44b	1.36b	3.80bc	2.59bc
	32%	5.72b	1.87b	4.10b	3.19b
	100%	6.76a	5.72a	6.44a	4.98a
V _{cmax}	8%	31.8b	21.7b	36.8b	25.1c
	16%	34.2b	20.8b	24.8bc	30.2bc
	32%	39.1b	29.0b	41.8b	38.7b
	100%	63.2a	45.6a	50.8a	42.8a

SLA was an important growth parameter measuring response to shade. The increase in SLA with shade indicates that thinner leaves with lower mass developed under the shaded conditions (Fig.3). This is an adaptation of larch seedlings to shade. In general, thinner leaves can use limited light more efficiently and gain the maximum amount of carbon under the shaded conditions (Evans and Poorter 2001, Terashima and Hikosaka 1995, Senevirathna *et al.* 2003). At the leaf level, a given amount of biomass can be spread over a small or large area. Plants grown in strong light generally have thick leaves with a low SLA, due in part to multi-layers of palisade or longer palisade cells (Hanson 1917). This increases the number of chloroplasts and the amount of photosynthetic enzymes, and thereby enhances the photosynthetic capacity per unit leaf area.

The total chlorophyll content per unit leaf area and the chlorophyll (Chl.) a/b ratio decreased in response to shade (Fig. 5). Terashima and Hikosaka (1995) found that the Chl a/b ratio can be a useful indicator of N partitioning within a leaf, sine it should be positively

correlated with the ratio of PSII cores to the light harvesting chlorophyll-protein complex (LHCII). LHCII contains the majority of Chl b, and consequently has a lower Chl a/b ratio (1.3-1.4) than the other Chl binding proteins associated with PSII (Evans 1989; Green and Durnford 1996). It follows that Chl a/b ratios should increase with increasing irradiance at a given level of N availability (Hikosaka and Terashima 1995). The total leaf chlorophyll per unit leaf area decreased with shade, probably because the thinner leaves produce fewer cells per unit leaf area under shaded conditions (Senevirathna *et al.* 2003).

In conclusion, the Japanese larch showed greater changes in response to decreasing PPFD, due to a remarkable change in SLA. This tendency was more pronounced given a high nitrogen supply. However, Japanese larch seedlings could not utilize administered high levels of nitrogen under shaded conditions. Based on these findings, the hybrid larch is expected to be more dominant in slightly shaded conditions on infertile soil.

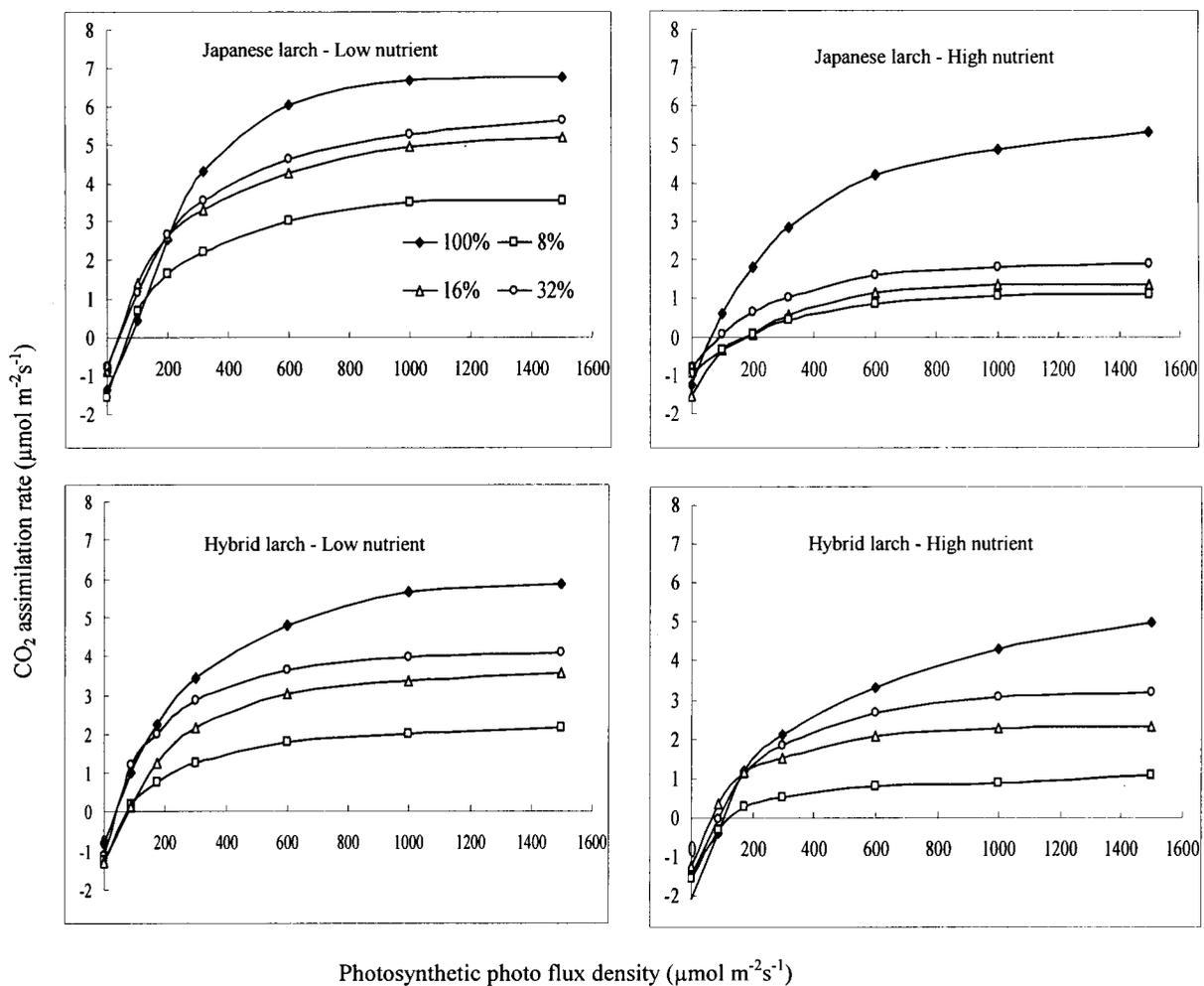


Fig. 6. An example of the relationship between leaf CO₂ assimilation rate and photosynthetic photo flux density (PPFD) in Japanese larch and hybrid larch grown under four light regimes (8%, 16%, 32% and 100%) in combination with two nutrient (5mg and 25mg) treatments.

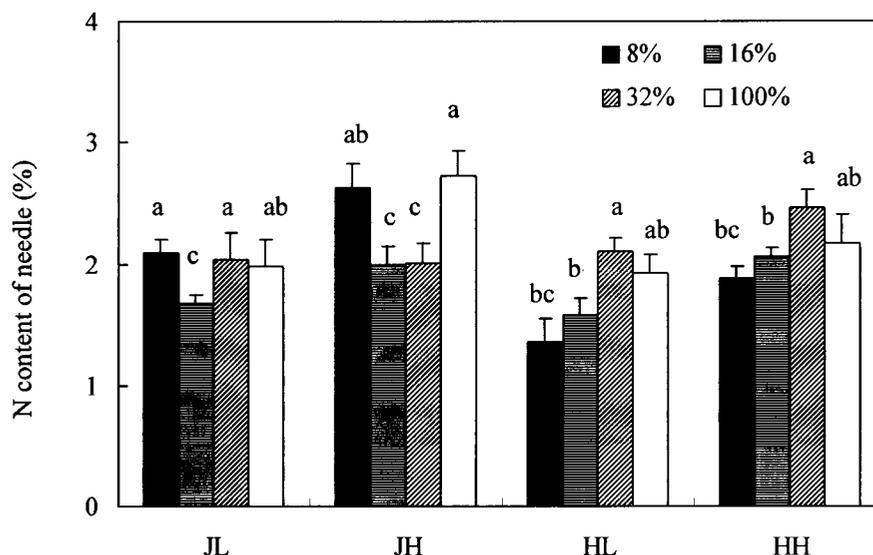


Fig. 7. N content (%) in needle of seedlings of Japanese larch and hybrid larch in four light regimes (8%, 16%, 32% and 100%) in combination with two nutrient treatments (5mg and 25mg). Treatment codes are shown in Figure 1. Vertical bars represent the standard error of its mean (n = 10). Values with the same letter (a, b) are not significantly different among light conditions at $P < 0.05$.

Acknowledgment

We are grateful to Dr. Y. Tamai for support of the greenhouse cultivation of seedlings.

References

- Bazzaz, F.A. (1996) Plant in changing environments. Chambridge University Press. Chambridge. Pp320.
- Evans, J.R. (1989) Partitioning of nitrogen between and within leaves grown under different irradiances. Australian J. Plant Physiol. 16:533-548.
- Evans, J.R. and Poorter, H. (2001) Photosynthetic acclimation of plants to growth irradiance: the relative importance of specific leaf area and nitrogen partitioning in maximizing carbon gain. Plant Cell Environ. 24:755-767.
- Farquhar, G.D. and Sharkey, T.D. (1982) Stomatal conductance and photosynthesis. Ann. Rev. Plant Physiol. 33:317-345.
- Gower, S.T. and Richards, J.H. (1990) Larches: deciduous conifers in an evergreen world. BioScience 40: 818-826.
- Green B.R. and Durnford, D.G. (1996) The chlorophyll-carotenoid protein of oxygenic photosynthesis. Annu. Rev. Plant physiol. Plant Mol. Biol. 47: 685-714.
- Hanson, H.C. (1917) Leaf-structure as related to environment. Amer. J. Bot. 4:533-560.
- Hikosaka, K. and Terashima, I. (1995) A model of the acclimation of photosynthesis in the leaves of C_3 plants to sun and shade with respect to nitrogen use. Plant Cell Environ. 18: 605-618.
- Koike, T., Yazaki, K., Funada, R., Maruyama, Y., Mori, S. and Sasa, K. (2000) Forest health and vitality in northern Japan – A history of larch plantation. Res. Note, Fac. Forestry, Univ. of Joensuu, 92: 49-60.
- Lambers, H., Chapin III, F.S. and Pons, T.L. (1998) Plant physiological ecology. Springer-Verlag, New York, USA, p540.
- Matsuki, S., Ogawa, K., Tanaka, A. and Hara, T. (2003) Morphological and photosynthetic response of *Quercus crispula* seedlings to high-light conditions. Tree Physiol. 23:769-775.
- Qu, L., Quoreishi, A.M. and Koike, T. (2003) Root growth characteristics, biomass and nutrient dynamics of seedlings of two larch species raised under different fertilization regimes. Plant Soil 255:293-302.
- SAS Institute Inc. (1996) SAS/STAT user's guide. SAS Institute INC. Cary, N.C.
- Senevirathna A.M.W.K., Stirling, C.M. and Rodrigo, V.H.L. (2003) Growth, photosynthetic performance and shade adaption of rubber (*Hevea brasiliensis*) grown in natural shade. Tree Physiol. 23:705-712.
- Sharkey, T.D. (1985) Photosynthesis in intact leaves of C_3 plants: physics, physiology and rate limitations. Bot. Rev. 51:53-105.
- Shinano, T., Lei, T.T., Kawamukai, T., Inoue, M.T., Koike, T. and Tadano, T. (1996) Dimethylsulfoxide method for the extraction of chlorophylls a and b from the leaves of wheat, field bean, dwarf bamboo, and oak. Photosynthetica 32:409-415.
- Sims D.A. and Pearcy, R.W. (1994) Scaling sun and shade photosynthetic acclimation of *Alocasia macrorrhiza* to whole-plant performance – I. Carbon balance and allocation at different daily

photon flux densities. *Plant Cell Environ.* 17:
881-887.
Terashima I. and Hikosaka, K. (1995) Comparative

ecophysiology of leaf and canopy photosynthesis.
Plant Cell Environ. 18:1111-1128.