

Optimum light transmittance for seed germination and early seedling recruitment of *Pinus koraiensis*: implications for natural regeneration

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Light transmittance regulated by canopy openness influences the microsite conditions for natural regeneration. The successful transition from seed germination to subsequent seedling recruitment (*i.e.*, early seedling survival and growth) determines the natural regeneration potential. However, there is little information on the effect of varying light transmittance on seed germination and seedling recruitment of *Pinus koraiensis* Siebold & Zucc. (Korean pine). We aimed to determine the optimum light requirements for this transition process in *P. koraiensis* to propose practical measures for improving its natural regeneration. The transition process was studied under five light transmittance regimes (100%, 60%, 30%, 15% and 5% of full light) over two consecutive years (2010 and 2011). The highest germination percentage in both years occurred at 30% light transmittance. Generally, mean germination time (MGT) declined with increased light transmittance. Seedling survival exhibited no significant differences between treatments for 1-year-old seedlings, but was higher at 30% than at 5% light transmittance for the 2-year-old seedlings. In contrast, seedling height, root collar diameter and total biomass were highest at 60%-100% light transmittance for both 1- and 2-year-old seedlings. Furthermore, the light transmittance also influenced the growth characteristics of *P. koraiensis* seedlings through regulating MGT. These results suggest that growth of *P. koraiensis* seedling requires a higher light transmittance (60-100%) than that required for seed germination, even though 30% light transmittance was favorable to the earlier emergence with larger specific leaf area. Silvicultural measures such as thinning are recommended to increase light irradiance in the forest understorey with the aim of improving the natural regeneration of *P. koraiensis*.

Keywords: Germination, Light Transmittance, Natural Regeneration, Seedling Emergence, Seedling Survival

Introduction

Korean pine (*Pinus koraiensis*) is mainly distributed in northeast Asia (Ma et al. 1992), where it covers about 500 000 km² of mixed broad-leaved Korean pine forests (MBKPF - Fig. 1). The MBKPF is one of the regional climax vegetation types in mountainous areas of northeast China, where *P. koraiensis* is mixed to a range of broad-leaved tree species such as *Juglans mandshurica*, *Fraxinus mandshurica* and *Quercus mongolica*. However, due to intense human disturbances and extreme natural disasters (Guo et al. 2010, Ren et al. 2012), most of

the original MBKPF (accounting for 52.5% of Northeast China) have been destroyed and replaced by secondary forests (Zhu 2002, Wu et al. 2004). The productivity and ecological functions of secondary forests are much lower than those of the original MBKPF. Specifically, secondary forests are characterized by the disappearance of basic structures and intrinsic functions, including the reduction in biodiversity, stability and ecosystem productivity (Zhu 2002). Increasing the proportion of *P. koraiensis* in the secondary forests, and further development of these MBKPF stands is consid-

ered important to promote these ecological and productive functions. However, development of a *P. koraiensis* component has been severely limited due to its failure in natural regeneration.

To achieve natural regeneration, it is vital to provide abundant seeds, ensure successful seed germination, seedling emergence and survival (Zhu et al. 2003). The successful transition from seed germination to subsequent seedling recruitment (*i.e.*, early seedling survival and growth) determines the regeneration potential for tree species (Hussain et al. 2001). Furthermore, these two key stages are the most sensitive to environmental conditions (*e.g.*, light, temperature and water conditions – Kyereh et al. 1999, Ruano et al. 2009) or biotic factors (*e.g.*, animal predation or browsing) in the regeneration processes. Due to the poor palatability of *P. koraiensis* seedlings, browsing pressure is not a factor limiting natural regeneration. However, the environmental requirements can change from the stages of seed germination to seedling recruitment (Figueroa & Lusk 2001). Therefore, determining the requirements for both these two key stages would be helpful to improve the natural

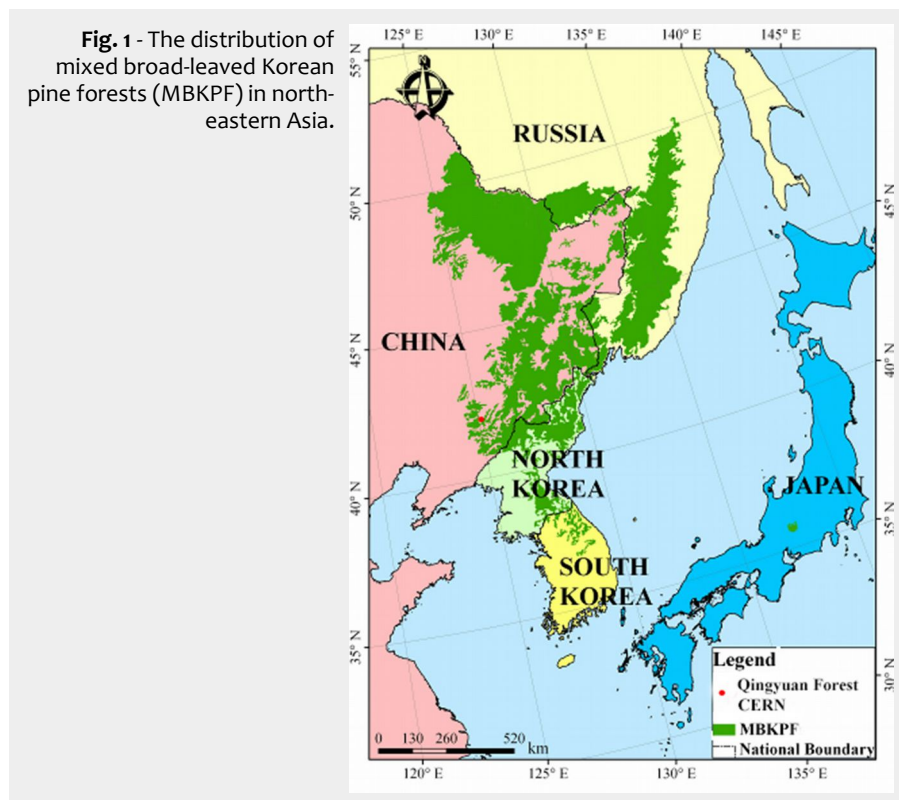
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Received: Jul 10, 2014 - Accepted: Feb 11, 2015

Citation: Zhang M, Yan Q, Zhu J (2015). Optimum light transmittance for seed germination and early seedling recruitment of *Pinus koraiensis*: implications for natural regeneration. *iForest* 8: 853-859. - doi: 10.3832/ifor1397-008 [online 2015-05-22]

Communicated by: Gianfranco Minotta



regeneration of *P. koraiensis*.

In silviculture, adjusting the light transmittance to the forest understorey by changing the forest composition and canopy structure is crucial to improve tree regeneration (Pothier & Prévost 2002, Nocentini 2009). Canopy openness is a characteristic widely analyzed in many studies, since it relates with the light transmission through the canopy (Beaudet & Messier 2002, Zhu et al. 2003, Chianucci & Cutini 2012). In temperate deciduous forests, canopy openings caused by natural or human disturbances produce spatial heterogeneity of light regimes in forest understorey (Beaudet & Messier 2002). In addition, seasonal variation in leaf canopy also contributes greatly to the temporal variability of light conditions under temperate secondary forests (Rich et al. 1993). Consequently, the spatio-temporal variation of light in the understorey plays a critical role in determining patterns of tree regeneration. The opening of a canopy firstly changes the light regimes, and then brings about the variation in temperature, soil moisture, etc. The estimate of light transmittance in the understorey, therefore, represents a wide range of microsite conditions including light, temperature and soil moisture.

Many studies have been published on the response of seed germination or seedling survival/growth of *Pinus* spp. to different light conditions (Day et al. 2005). In particular, it has been reported that light requirements may differ between seed germination and the subsequent seedling growth. For example, Parker et al. (2006) concluded that both seedling germination

and percent emergence were significantly higher under low light (13% of full sunlight) than under moderate light (47% of full sunlight) for *Pinus strobus* L.; however, moderate light level favored seedling growth in this species (Parker et al. 2004). *P. pinea* maintained high germination percentages in dark conditions (Ganatsas & Tsakalimi 2007), while seedlings required fairly high light irradiance (Fady et al. 2004). *P. pinaster* exhibited the highest germination percentage and aboveground dry weight in 25% and 100% light intensity, respectively (Ruano et al. 2009). Pulinetz (1986) studied the development of *P. koraiensis* under different light intensities created by the shading of shrubs, and concluded that the light requirements varied greatly across the entire growth period. Zhang et al. (2012) found that 30% of the full sunlight was optimum for seed germination of *P. koraiensis* in both shade houses and field conditions. To our knowledge, 60% of full light was the most favorable environment for the growth of 5-year-old *P. koraiensis* seedlings (Zhu et al. 2014).

Despite the number of studies on the above topic, no reports based on continuous observations have been published with the aim of exploring possible differences in light requirements between seed germination and initial seedling recruitment in *P. koraiensis*. To quantify the light requirements of these two key stages, seeds of *P. koraiensis* were sowed at five light transmittances (100%, 60%, 30%, 15% and 5% of the full light), and emerged seedlings were monitored for two consecutive years. We hypothesized that the light requirements for seedling growth

were different from those for seed germination in *P. koraiensis*.

Materials and methods

Study site

The study was conducted at the Qingyuan Forest CERN (Chinese Ecosystem Research Network), Chinese Academy of Sciences (41° 51' 06" N, 124° 54' 33" E, 456-1116 m a.s.l.). The climate is a continental monsoon type with a windy spring, a warm, humid summer, and a dry, cold winter. Annual average precipitation in the area is 811 mm and mean annual air temperature is 4.7 °C with a maximum 36.5 °C in July and a minimum -37.6 °C in January. The frost-free period lasts 130 days and the growing season ranges from early April to late September (Zhu et al. 2008). The vegetation is representative of the natural secondary forest in this region, dominated by *Juglans mandshurica*, *Phellodendron amurense*, *Fraxinus mandshurica*, *Quercus mongolica*, and *Fraxinus rhynchophylla*.

Seed collection and pretreatment

Fresh seeds of *P. koraiensis* were randomly collected from at least 3 (30 × 30 m) stands with reproductively mature trees at the Qingyuan Forest CERN in late September 2009. All seeds were air-dried from September to November. Then more than 1000 seeds were mixed with wet sand and stratified at low temperatures (between -5 and zero °C) from November 2009 to April 2010 to break the deep dormancy characteristic of this species. In May 2010, the seeds were soaked in 0.5% potassium permanganate (K₂MnO₄) for 30 minutes to sterilize the seed surfaces. Any seeds that sunk immediately were considered as "viable" (Tanaka-Oda et al. 2009), while floating seeds were considered as "non-viable" and then removed. Viable seeds of a similar size were used for the germination experiment in this study.

Experiment design

Four main seed plots were framed by wooden slats. Each plot was divided into three subplots (ca. 150 × 100 cm) with a 30 cm buffer area between the two adjacent subplots (Fig. 2). We assigned different light transmittance to each subplot by using different layers of neutral shading nets above the wooden slats. Each treatment was created by the use of independent shading nets, obtaining 3 replicates of 4 levels of light treatments (60%, 30%, 15% and 5% light transmittance), with full sunlight (100% light transmittance) as the control, which was also replicated three times. The relative light transmittance in each treatment was calculated by the specific photosynthetic photon flux density (PPFD) with respect to the full sunlight value (Fig. 3). For all the five treatments, the PPFD and topsoil (0-5 cm) temperature were measured simultaneously using quantum light sensors and temperature probes con-

nected to a data logger (Watchdog Model 200, Spectrum Technologies, Inc., Plainfield, IL, USA). Data were logged at 15-min intervals on three uninterrupted sunny days (from 07:00 to 18:00 – McLaren & McDonald 2003) monthly from May to September. Light quality was quantified by averaging the measures from 12:00 to 13:00 on one sunny day (Seiwa et al. 2009) using a different quantum sensor (Skye 110 Red/Far-Red light sensors, Skye instruments Ltd. Llandrindod Wells, Powys, UK). To improve ventilation in the shade houses, a distance of 2-3 centimeters was left between the netting edge and the ground. The relative humidity was monitored by HOBO Pro Model H08-032-08 (Onset Computer Corp., Bourne, MA, USA), and considered to be the same in the different shade houses because of the air circulation.

Fifty *P. koraiensis* seeds were sown at a spacing of 3-5 cm in each light treatment in May 2010. To prevent seed predation by mice or squirrels, all the seeds were sown in plastic pots (25 × 30 cm) which were protected by wire netting. The soil used in this experiment was taken from the top soil (0-5 cm) of the natural secondary forest at the Qingyuan Forest CERN. Before the experiment, soil was sieved to exclude stones, plant roots and *P. koraiensis* seeds, etc. No fertilizer was applied to the soil. The soil pH was 6.3 (Yang et al. 2013). The number of germinated seeds was recorded every 3-5 days from May to late September (the growth period) over two consecutive years (2010 and 2011), respectively. The germination experiment was terminated once no further germination occurred over a two week period (June 2011- September 2011). Germination date was recorded when the first needle sprout becoming visible (Xiong & Nilsson 1999, Zhu et al. 2003). Five to ten surviving seedlings in each light transmittance treatment were dug up randomly (every 5th seedling from a random start) in late September 2010 and 2011 respectively, to explore the survival rate, growth state, etc., for 1- and 2-year-old seedlings. Since we did not mark seedlings that emerged in 2010, we assumed that seedlings with relatively larger height and root collar diameter in 2011 could be classified as being two years old. After measuring the growth traits of harvested seedlings (including height, root collar diameter, number of leaves, root length and stem length), each seedling was separated into shoots and roots. These were dried at 90 °C for 30 min, then at 65 °C for 24 hours and finally the dry biomass were quantified.

Statistical analysis

Three parameters (germination percentage, coefficient of germination rate, mean germination time) were used to characterize seed germination. Seven additional parameters (survival rate, seedling height, root collar diameter, total biomass, specific leaf area, leaf area ratio, leaf mass fraction)

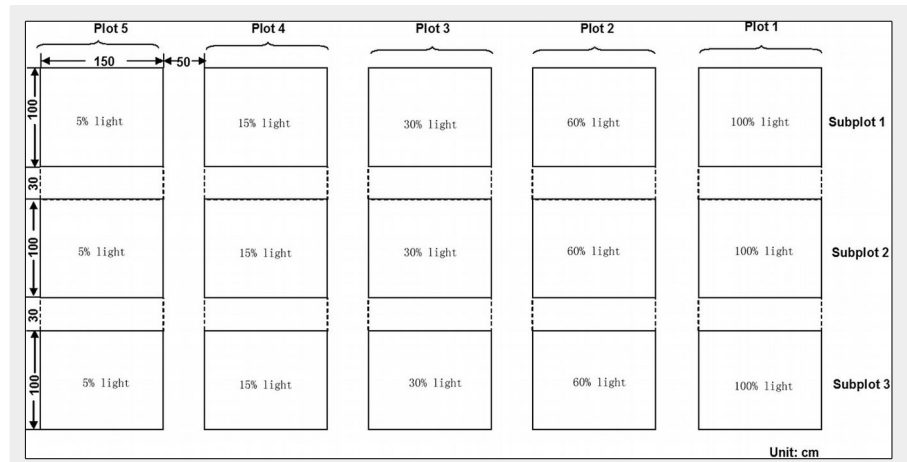


Fig. 2 - Outline of the experimental design.

were used to analyze the seedling survival and survival quality of *P. koraiensis*. The parameters not measured directly were calculated as described below. The germination percentage (*GP*) was calculated as (eqn. 1):

$$GP = \frac{S_{NM}}{S_{NO}} \cdot 100$$

where *S_{NM}* is the number of germinated seeds, and *S_{NO}* is the number of total experimental seeds (Moya et al. 2013). The mean germination time (*MGT*) was obtained as (eqn. 2):

$$MGT = \frac{\sum DN}{\sum N}$$

where *D* is the time in days from the sowing date, and *N* is the number of germinated seeds on a given day (Close & Wilson 2002). The survival rate (*SR*) was estimated as (eqn. 3):

$$SR = \frac{S_{NS}}{S_{NE}} \cdot 100$$

where *S_{NS}* is the number of surviving seedlings on a given day, and *S_{NE}* is the number of all emerging seedlings from the sowing day. In this study, survival rate was based on data obtained in late September

in 2010 and 2011, respectively.

Leaf area (*LA*) was calculated using the following equation (eqn. 4):

$$LA = \frac{\pi DL}{5 \sin(36^\circ)}$$

where *D* represents the width measured in the middle part of each needle by calipers, and *L* represents the needle length. Specific leaf area (*SLA*) was calculated as (eqn. 5):

$$SLA = \frac{LA}{LM}$$

where *LM* is the leaf biomass, and *LA* is the leaf area, while the leaf area ratio (*LAR*) was obtained as (eqn. 6):

$$LAR = \frac{LA}{TB}$$

where *LA* is the leaf area, and *TB* is the total biomass. The leaf mass fraction (*LMF*) was estimated as follows (eqn. 7):

$$LMF = \frac{LB}{TB} \cdot 100$$

where *LB* is the leaf biomass, and *TB* is the total biomass.

As suggested by Zuur et al. (2010), a transformation of the response variable

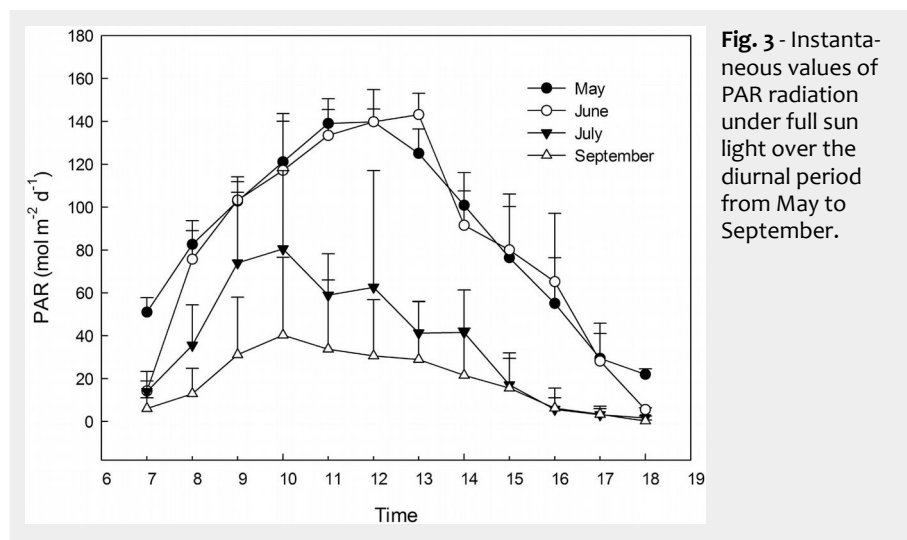
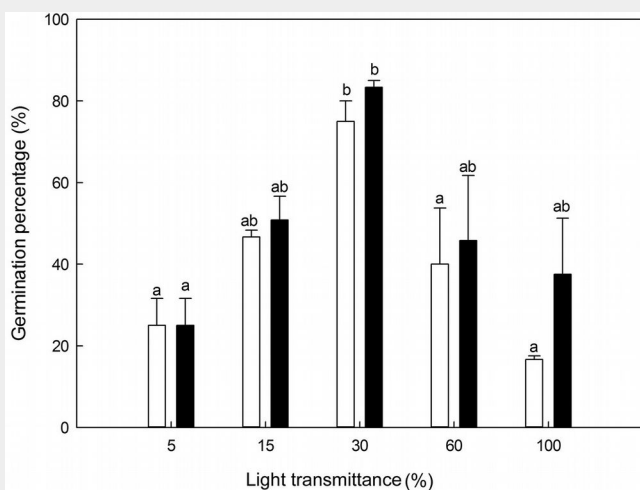
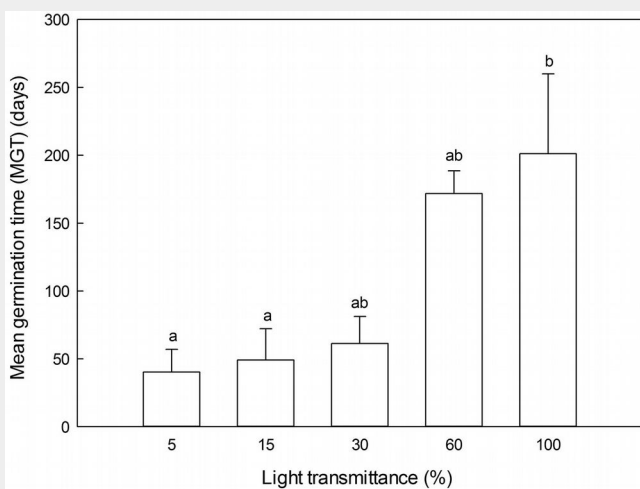
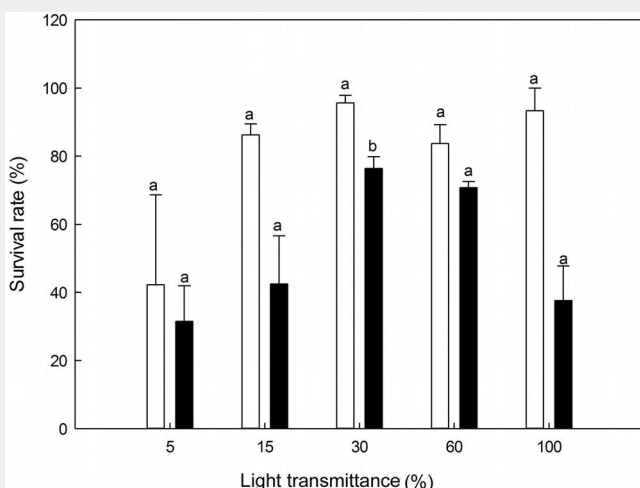


Fig. 3 - Instantaneous values of PAR radiation under full sun light over the diurnal period from May to September.

Tab. 1 - Environmental conditions in the shade houses. Different letters indicate significant differences in environment conditions among light treatments.

| Shade house/site | PAR (mol m ⁻² d ⁻¹) | Red/Far Red ratio | Topsoil temperature (°C) | Air humidity (%) |
|------------------|--------------------------------------------|---------------------------|---------------------------|------------------------|
| 100% | 60.60 ± 10.66 | 1.11 ± 0.003 ^a | 24.75 ± 0.53 ^b | 71 ± 3.8 ^a |
| 60% | 39.70 ± 6.76 | 1.10 ± 0.005 ^a | 23.17 ± 0.49 ^b | 62 ± 9.3 ^a |
| 30% | 20.15 ± 3.11 | 1.10 ± 0.002 ^a | 20.75 ± 0.51 ^a | 69 ± 10.5 ^a |
| 15% | 11.15 ± 2.60 | 1.09 ± 0.002 ^a | 20.33 ± 0.53 ^a | 80 ± 4.8 ^a |
| 5% | 5.83 ± 1.39 | 1.08 ± 0.003 ^a | 19.92 ± 0.24 ^a | 79 ± 11.0 ^a |

Fig. 4 - Germination percentages of *Pinus koraiensis* under different light transmittances. Different letters indicate significant differences in germination percentage along the light transmittances ($P \leq 0.05$). The black and white column represents the cumulative germination percentage in the first year (2010) and the second year (2011), respectively.**Fig. 5** - Mean germination time (MGT) of *Pinus koraiensis* under different light transmittances for 2-year-old seedlings. Different letters indicate significant differences in MGT along the light transmittances ($P \leq 0.05$).**Fig. 6** - Survival rates of *Pinus koraiensis* seedlings under different light transmittances. Different letters indicate significant differences in survival rate along the light transmittances ($P \leq 0.05$). The white and black column represents the survival rate for 1- and 2-year-old seedlings, respectively.

should be applied when departure from normal distribution or heterogeneity of variance occur. Using normal probability plots, percentage data (i.e., germination percentage, survival rate) was tested for normality, and germination data was transformed as arcsine-square-root to meet the assumptions of ANOVA (Ren & Tao 2004, Seiwa et al. 2009). Post-hoc Tukey's test was applied using the SPSS® v. 16 software package (SPSS Inc., Chicago, IL, USA) to ascertain the significance of differences in seed germination for the two years and seedling survival/growth across the different light transmittance regimes for 1-year-old seedlings. The non-parametric Mann-Whitney test was applied to *SLA* data to overcome their departure from normality. To exclude the confounding effect of morphology in the first year, a covariance analysis (ANCOVA – South et al. 1989) with light as the fixed factor and morphology values of 1-year-old seedlings as the covariate factor was conducted for 2-year-old seedlings. A backwards multiple regression was carried out to quantify the contributions of light transmittance and *MGT* to 1- and 2-year-old seedling growth characteristics, including seedling height, root collar diameter (*RCD*), total biomass, specific leaf area (*SLA*), leaf area ratio (*LAR*), and leaf mass fraction (*LMF*). *SLA*, *LAR* and *LMF* were plotted against the light transmittance, respectively. Differences obtained at a level of $P \leq 0.05$ were considered significant.

Results

Internal environmental conditions in shade houses

In all the five light transmittance regimes, the topsoil temperature varied significantly (Tab. 1). However, the topsoil temperature exhibited no significant differences from 100% to 60% light transmittance, and from 30% to 5% light transmittance. The Red/Far Red ratio and air humidity did not vary between the five treatments (Tab. 1).

Effects of light transmittance on seed germination

The germination percentage of *P. koraiensis* was significantly affected by the light transmittance in the two years (ANOVA: $F=9.64$, $P < 0.01$ in 2010; $F=3.40$, $P=0.05$ in 2011 - Fig. 4). The germination percentage was slightly higher in the second year than in the first year under all the light transmittance regimes. In both years, the germination percentage was the highest at 30% light transmittance, while the lowest was found at 100% (the first year) and 5% light transmittance (the second year), respectively.

MGT was the highest in full sunlight and declined with decreasing light transmittance, with no significant differences among the four lower light transmittance regimes (Fig. 5).

Effects of light transmittance on early seedling recruitment

No significant differences in survival were found among the five light transmittance treatments for 1-year-old seedlings (ANOVA: $F=2.91$, $P>0.05$ - Fig. 6). However, 30% light transmittance showed significantly higher survival rates along the light levels for 2-year-old seedlings (ANCOVA: $F=4.87$, $P=0.03$ - Fig. 6). Overall, the survival rates of all the treatments were seemingly lower for 2-year-old seedlings than for 1-year-old seedlings. Compared to 1-year-old seedlings, 2-year-old seedlings exhibited significantly lower survival rates ($P<0.05$) at 30% and 15% light transmittance (Fig. 6).

Seedling height of *P. koraiensis* increased with age (ANOVA: $F=10.63$, $P<0.001$ for 1-year-old seedlings; ANCOVA: $F=10.82$, $P<0.001$ for 2-year-old seedlings - Fig. 7). It was significantly higher in 100% light transmittance regime for 1-year-old seedlings and in 60-100% light transmittance regimes for 2-year-old seedlings (Fig. 7). Generally, the root collar diameter of *P. koraiensis* seedlings was significantly larger at 100%, 60% and 30% light transmittance, though decreased slightly at 15% and 5% light transmittance with seedling age (ANOVA: $F=4.27$, $P<0.01$ for 1-year-old seedlings; ANCOVA: $F=11.28$, $P<0.001$ for 2-year-old seedlings - Fig. 7). The largest root collar diameter was always recorded in the 100% light transmittance treatment (Fig. 7).

There were significant differences in biomass between 100% light transmittance and the other treatments for 1-year-old *P. koraiensis* seedlings (ANOVA: $F=14.03$, $P<0.01$). Light irradiance (univariate analysis: $F=7.38$, $P=0.03$) and the interaction between light and biomass in the first year (univariate analysis: $F=6.91$, $P=0.03$) jointly affected the biomass accumulation pattern in the second year. For 2-year-old seedlings, biomass at the 60% and 100% light transmittance was significantly higher as compared with the other three light regimes (Fig. 8).

For 1-year-old seedlings, a significant logistic relationship was determined between the *SLA* and the light transmittance ($SLA_1 = 91.14 - 6.15 \cdot \ln(Light)$, $R^2=0.854$, $P<0.05$). A significant linear correlation was found between *LAR* and light transmittance for the 1-year-old seedlings ($LAR_1 = 2.76 - 0.01 \cdot Light$, $R^2 = 0.904$, $P<0.05$), and between *LMF* and light transmittance for both ages of seedlings ($LMF_1 = 0.51 - 0.0008 \cdot Light$, $R^2 = 0.749$, $P<0.10$; $LMF_2 = 0.499 - 0.001 \cdot Light$, $R^2 = 0.916$, $P<0.05$).

Contributions of light transmittance and germination parameters on seedling performance

Except for the *SLA*, all the characteristics of 1- and 2-year-old seedlings can be explained by light transmittance, *MGT*, or a combination of them (Tab. 2). For 1-year-old plants, seedling height, root collar diameter, *LAR* and *LMF* were all significantly

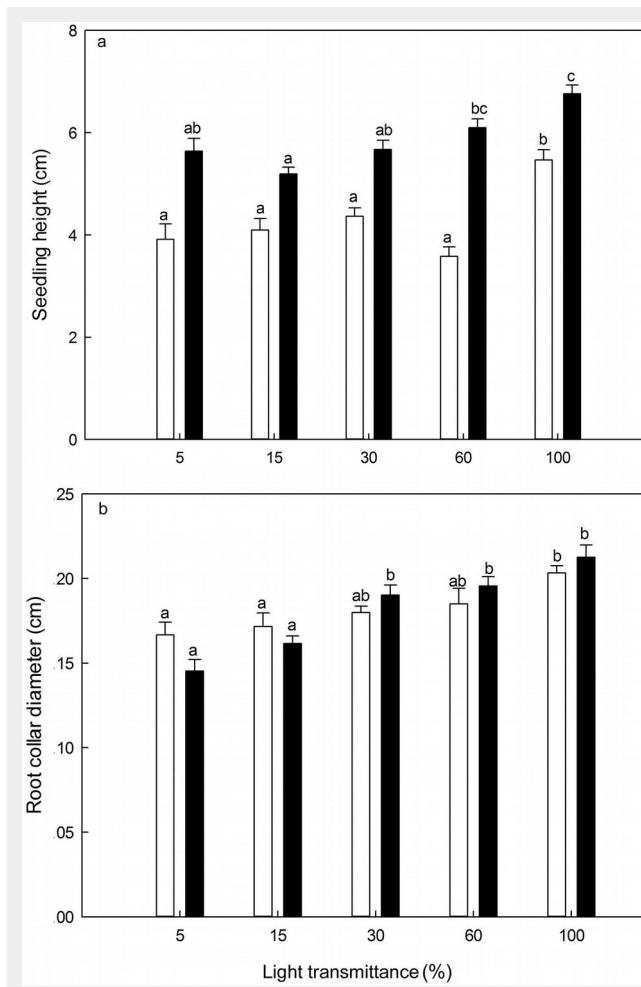


Fig. 7 - Seedling height (a) and root collar diameter (b) of *Pinus koraiensis* under different light transmittance regimes. Different letters indicate significant differences in seedling height and root collar diameter along the light transmittances ($P\leq 0.05$). The white and black column represents seedling height and root collar diameter for 1- and 2-year-old seedlings, respectively.

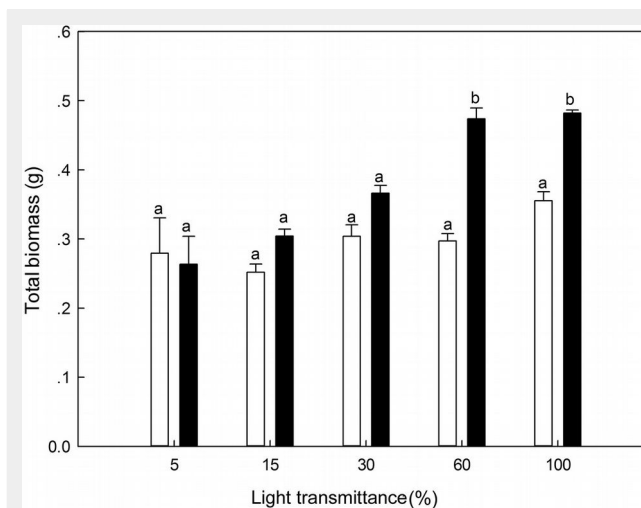


Fig. 8 - Total biomass of *Pinus koraiensis* under different light transmittance regimes. Different letters indicate significant differences in total biomass along the light transmittances ($P\leq 0.05$). The white and black column represents the total biomass for 1- and 2-year-old seedlings, respectively.

Tab. 2 - Effects of light transmittance and mean germination time (*MGT*) on 1- and 2-year-old seedling growth characteristics. The standardized coefficients of the backwards multiple regression are shown for variables which contributed significantly to the model. Values outside and inside the brackets represented the data obtained for 1- and 2-year-old seedlings, respectively. (-): no observation; (*): $P<0.05$; (+): $P<0.10$.

| Variables | Light transmittance | <i>MGT</i> | R^2 |
|-------------------------------------|---------------------|----------------|--------------|
| Seedling height (<i>SH</i>) | 3.08*(-) | -2.52*(0.93*) | 0.92*(0.87*) |
| Root collar diameter (<i>RCD</i>) | 1.51*(0.92*) | -0.54*(-) | 0.99*(0.84*) |
| Total biomass (<i>TB</i>) | 0.88*(-) | -(0.96*) | 0.78*(0.91*) |
| Specific leaf area (<i>SLA</i>) | - (-) | - (-) | - (-) |
| Leaf area ratio (<i>LAR</i>) | -1.99*(1.59*) | 1.09*(-2.42*) | 0.99*(0.99*) |
| Leaf mass fraction (<i>LMF</i>) | -2.50*(-) | -1.70*(-0.96*) | 0.97*(0.92*) |

related to the combination of light transmittance and *MGT*, while total biomass was only related to light transmittance. For the 2-year-old seedlings, height, total biomass and *LMF* were significantly related to *MGT*, and *RCD* was significantly related to the light transmittance, while *LAR* was related to the combination of light transmittance and *MGT*. For *LAR* and *LMF*, light transmittance and *MGT* could explain more than 90% of their variances for 1- and 2-year-old seedlings.

Discussion

Light requirements for seed germination of *P. koraiensis*

The germination percentage in both years was the highest at 30% light transmittance. The results showed the same response to light transmittance in both years, demonstrating that light levels play a critical role in seed germination. According to our field observations, 30% light transmittance occurred in broadleaved secondary forest stands in the early spring prior to leaf expansion (see Fig. 1 in Zhang et al. 2012). Thus, early spring seem a favorable period for seed germination of *P. koraiensis*. This result clearly confirms the findings of our previous field experiment (Zhang et al. 2012), in which 80% of *P. koraiensis* seeds germinated prior to leaf expansion.

The *MGTs* of *P. koraiensis* were also affected by light transmittance. According to previous reports (Pons 1992, Silvertown & Doust 1993), it is advantageous for plants to initiate germination when conditions are favorable for seedling survival. However, the optimum conditions for seedling growth might not correspond to those for seed germination and seedling survival. Indeed, our results indicated that the full sunlight favors the seedling growth, whereas it prolongs the mean germination time and slows down the germination rate. The different germination percentages in two light regimes with similar temperature and moisture conditions (Tab. 1, Fig. 4), 30% and 5% light transmittance, emphasized that there were significant effects of light on seed germination of *P. koraiensis*. Furthermore, compared with full sunlight conditions, shading could play a critical role in increasing germination percentage, and shortening the germination time. In summary, 30% light transmittance was the optimum light condition for seed germination in *P. koraiensis*, with the highest germination percentage and a relatively fast germination rate (lower values of *MGT*).

Light requirements for seedling survival and growth of *P. koraiensis*

In this study, no significant differences in survival rate were found for the 1-year-old seedlings (Fig. 6), suggesting that *P. koraiensis* seedlings had a wide plasticity at the earliest stage, allowing their acclimation to different light transmittance regi-

mes. However, 30% light transmittance had the highest survival rate for the 2-year-old seedlings, indicating similar light requirements for both seedling survival and seed germination of *P. koraiensis*. Carbohydrates are critical for maintaining the metabolism and provide a buffer for *P. koraiensis* seedlings under stressful conditions (Zhang et al. 2013). Thus, the overall lower survival rates for 2-year-old seedlings than for 1-year-old seedlings might reflect a lag effect of *P. koraiensis* seedlings to different light irradiance. Morphologically, 60%-100% light transmittance was the optimum level for seedling height, root collar diameter and total biomass of *P. koraiensis* seedlings (Fig. 7, Fig. 8). This is supported by the finding that the light-saturated photosynthetic rates in 60% and 100% treatments were higher than those in other light treatments for 5-year-old seedlings (Zhu et al. 2014). Furthermore, as a response to shadier conditions, the largest *SLA*, *LAR* and *LMF* were found in 5% light transmittance to maximize the light capture.

There was a discrepancy between the best light conditions for seed germination/seedling survival (30% light transmittance) and those for seedling growth (60%-100% light transmittance). This is due to the trade-off between carbon allocation for growth and for storage and defense (Poorter & Kitajima 2007). In the lowest light transmittance regime (5%), more carbon was allocated to seedling defense and storage resulting in the overall highest survival rate. However, at the 60% or 100% light transmittance levels, more carbon was allocated to seedling growth than to defense or storage.

Implications on light requirements for seed germination and early seedling growth of *P. koraiensis*

In this study, the interaction of light and *MGT* influence the growth of 1- and 2-year-old seedlings, as revealed by the regression analysis carried out (Tab. 2). Seedlings emerging from different light transmittance regimes may be influenced by both the abiotic (e.g., light) and biotic (e.g., *MGT* in the present study) factors (Noe & Zedler 2001). *MGT* was reported to have a negative relationship with seedling size, i.e., later germinating seeds produce smaller seedlings (Demir et al. 2008). In this study, we also found negative relationships between *MGT* and the height and root collar diameter of 1-year-old seedlings.

Overall, seed germination/seedling survival rate and seedling growth showed different response functions to light transmittance. Seed germination in response to light had a peak value at 30% light transmittance, while seedling height, root collar diameter and total biomass were greater with increased light. This implied that the light requirements of *P. koraiensis* were higher for seedling growth than for seed germination. Such uncoupled light requirements have also been reported for other

tree species which can germinate under closed canopy, but need higher light at the seedling stage (Kyereh et al. 1999, Fady et al. 2004, Parker et al. 2004, 2006, Ganatsas & Tsakalidimi 2007). Moreover, the germination of most seeds of *P. koraiensis* occurs in early spring before leaf expansion in the secondary forest (Zhang et al. 2012). The subsequent reduction in light transmittance after leaf expansion would be disadvantageous to seedling growth of *P. koraiensis*. The distinct light requirements for seed germination and seedling growth of *P. koraiensis* would be a major factor hampering its natural regeneration, strongly supporting our hypothesis in the present study. Therefore, to improve the natural regeneration of *P. koraiensis* in the secondary forest understory, disturbances (e.g., thinning) should be applied to allow more light reaching the understory layer.

Conclusions

Light requirements increased from 30% to 60-100% light transmittance during the transition from seed germination to seedling recruitment in *P. koraiensis*. The germination characteristics (i.e., *MGT*) showed a coupling effect with light transmittance on the subsequent pine seedling growth. However, the highest *MGT* in *P. koraiensis* was observed at 30% light transmittance, which is not the best light regime for seedling growth. There might be a hindrance for the natural regeneration process of *P. koraiensis* resulting from the decreased light transmittance after leaf expansion in the growing season, contrasting with the elevated light requirement for growth. Therefore, disturbances such as gap openings or thinnings are strongly recommended as an operational measure to enlarge canopy openness in temperate secondary forests and improve the natural regeneration of *P. koraiensis*.

Acknowledgements

Min Zhang contributed to the experimental design, carried out the experiment, and analyzed the data; she also shared in writing the manuscript. Qiaoling Yan contributed to the design of the experiment and helped to revise the manuscript. Jiaojun Zhu contributed to the design of the experiment, helped to write the manuscript, supervised the work and coordinated the research project.

We would like to thank Dehui Zeng, Kai Yang, Lizhong Yu, Miao Wang, Jinxin Zhang and Zhanqing Hao for their constructive suggestions about this study. Thanks are also due to Qun Gang, Wenwu Ji and De'an Zheng for their field work.

This research was supported by the National Natural Science Foundation of China (contract no. 31330016 and 30830085).

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