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Full Length Article

Growth and Nutrient Uptake in *Aralia elata* Seedlings Exposed to Exponential fertilization under Different Illumination Spectra

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Abstract

Natural non-wood forest product (NWFP) population tends to distribute in moist and highly-shaded undergrowth habitats, hence, their seedlings usually have a low growing rate. *Aralia elata* is one of the most highly-valued NWFP species in eastern Asia as sources of forest-derived food and traditional medicine. The objective of this study was to value the combined effects of artificial lighting spectra and exponential fertilization (EF) on seedling growth and nutritional responses of this species. One-year old *A. elata* seedlings were cultured with a 2×2 factorial design in an all-controlled environment to receive EF at 80 and 160 mg N seedling^{-1} [nitrogen (N)-phosphorus (P)-K, 12-9-12] under lighting spectra by light-emitting diodes (LEDs) (R/G/B, 30:69.3:0.7) and high-pressure sodium (HPS) lamps (R/G/B, 44.4:53.8:1.8) at the same light intensity of 60~64 μmol m^{-2} s^{-1}. After a growing season, seedlings had the greatest height (~60 cm) and root biomass (~700 mg) in the high-dose EF treatment under LED lighting. This combined treatment can also induce a steady-state nutrient uptake for N and P in *A. elata* seedlings compared to a lower dose of EF in the same LED spectrum. Higher foliar green-color degree suggested lower shoot N concentration but larger leaf area indicated a greater biomass accumulation. Therefore, cultivation of *A. elata* seedlings with EF at the rate of 160 mg N seedling^{-1} under a LED lighting spectrum is recommended. © 2020 Friends Science Publishers

Keywords: Ecosystem services; Forest food; Restoration; Nutritional symptom; Climate change; Anthropogenic activity

Introduction

Global consumption on non-wood forest products (NWFPs) is increasing due to the public's attitude to produce and use naturally produced food. NWFPs refer to goods from biological origin other than timber production from wooded lands (FAO 1999). Many NWFPs have high commercial importance and the gross number of commercial NWFP species was estimated to be 4000~6000 all over the world. Many NWFP resources are suffering over-exploitation but current knowledge on the regeneration of NWFP habit is neglected and less available compared to timber-species. The goal to harvest NWFPs has also been considered in forest management planning, which will be completely different from timber production. Trade-offs usually occurs between the yield of NWFP and production of tree-related products. For example, Gamfeldt *et al.* (2013) reported a trade-off between tree biomass accumulation and bilberry (*Vaccinium myrtillus* L.) production in boreal and temperate forests. Synergy models also indicated that NWFPs can have a negative correlation with cutting removals in boreal forests (Kurttila *et al.* 2018). Therefore, to obtain the optimum trade-off between NWFP yield and forest ecology, there are needs to be a new approach to the forest managing strategy compared to the traditional method with timber product yield as the unique goal.

Aralia etalta (Miq.) Seem (Araliaceae) is a traditional medicinal plant. The natural *A. elata* population mainly distributes in the Russian Far East, Northeast China, Japan, Korea, and north-eastern America (Sun *et al.* 2017). In Asia, *A. elata* is widely used as a mountain food and a traditional medicine. Pharmacological studies have revealed that extracts from leaves (Sun *et al.* 2017), root (Lee and Jeong 2009) and whole-plant body (Lee and Kang 2015) in natural *A. elata* individuals can be effective as an anti-arrythmia, antitumor, anti-inflammatory, and antioxidant. The increasing interest of NWFPs caused the intensive exploration of natural *A. elata* resources which even led to the destruction of the *A. elata* habitat. Recent investigation found that natural *A. elata* is also suffering the stress of highly-frequent foraging by red deer (*Cervus elaphus xanthopygus*) (Feng *et al.* 2018), which aggravates the depletion of resource of this species in addition to artificially over-exploitation in Northeast China. Therefore, it is necessary to restore the population of *A. elata* in forests. However, current studies on this species were mainly

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conducted on the pharmaceutical chemistry industry (Sun *et al.* 2017; Qi *et al.* 2018), resulting in limited useful knowledge on natural growth and restoration on this species.

To plant NWFP seedlings in land areas with a degrading plant population is an available approach to achieve artificial restoration. Several reports have indicated that seedling quality, which is established during the seedling culture process, is an important index for artificially cultured seedlings and can be used for predicting the further transplant performance (Grossnickle and MacDonald, 2018). A good-quality seedling should be hardened off after the nursery growth to improve morphological features to be able to compete for natural sources in the field. A high-quality larger seedling with improved height and root-collar diameter (RCD) is generally achieved because larger seedlings tend to utilize more light resource to promote photosynthesis (Pinchot *et al.* 2018). The natural attribute of *A. elata* determines its growing pattern which is a very short main stem but relatively long lateral branches (usually up to 30 cm) in the juvenile stage. This natural growth habit is the main cause of abiotic stress in *A. elata* seedlings (Feng *et al.* 2018). Therefore, most mature plants from naturally regenerated *A. elata* plants exhibit a long stem of at least 1.5 m at the cost of long-term growth for many years. To overcome this defect, the practical culture of *A. elata* seedlings usually requires up to 2–3 years with the purpose to obtain a seedling with a stem length of approximately 20 cm. The long-term seedling growth production cost of nursery space, labour, herbicide, and fertilizer.

Plant growth depends on light quality because lights in different wavelengths have varied functions for photosynthesis. High-pressure sodium (HPS) lamps are widely used in agricultural plant factory practices and studies because its light can induce significant plant response compared to the natural sunlight (Taulavuori *et al.* 2018). The use of HPS lamps as a supplementary lighting source has been proven to promote growth of tree seedling crops (Wei *et al.* 2013; Zhu *et al.* 2016; Li *et al.* 2017; Zhao *et al.* 2019). Recently studies are accumulating to demonstrate that lights from light-emitting diodes (LEDs) can even induce the better growth of tree seedlings relative to HPS light (Apostol *et al.* 2015; Riikonen, 2016; Riikonen *et al.* 2016; Li *et al.* 2018; Zhao *et al.* 2019). However, the effect of the light spectra on stem-elongation and the plant growth in height appeared to be controversial. For example, Li *et al.* (2018) found greater height growth in *Dalbergia odorifera* seedlings under LED lighting than under HPS lamps, but Apostol *et al.* (2015) reported that the height growth of conifer seedlings under LED *vs.* HPS lights depends on species and seed source. The uncertainty of the effect of light spectrum on height growth of tree seedlings inspires the interest to test the lighting spectra effect on the growth in NWFP plants. The light intensity changed by regulating shading has been detected for the growth of *A. elata* seedlings at a hypothesized uniform spectrum (Gao *et al.* 2019). Hence, it is further of value to compare *A. elata* seedlings under contrasting spectra to determine the potential of main stem elongation.

Nutrient reserve (NR) within a seedling at the end of nursery culture is another important parameter to evaluate seedling quality (Grossnickle and MacDonald 2018). NR reflects the physiological attribute of seedlings and a higher NR can enhance transplanted seedling performance through promoted nutrient re-translocation (Pokharel and Chang, 2016). Exponential fertilization (EF) has been proven to promote seedlings by absorbing more nutrients than needed for growth generating inherent NR (Wang *et al.* 2017; Zhao *et al.* 2017). In the environment of continuous lighting, studies revealed that NR would decline because nutrient concentration was decreased by the increase of biomass accumulation (Wei *et al.* 2013; Wang *et al.* 2017; Li *et al.* 2018; Zhao *et al.* 2019). The decline of NR would cause insufficient nutrient re-translocation and impact performance of a transplanted seedling (Millard and Grelet 2010; Pokharel and Chang 2016; Ueda 2012). The usage of artificial lighting during *A. elata* seedlings culture should be incorporated with proper EF regime, which may compensate the NR decline effect during fast growing.

In this study, a bioassay was conducted under a highly controlled environment and *A. elata* seedlings were cultured by continuous lighting from HPS lamps and LED panels without any sunlight. Two doses of nutrient were delivered to seedlings through the regime of classical EF model. It was hypothesized that: (i) LED lighting can improve the morphological feature with greater height and more root biomass compared to the HPS, and (ii) a high dose EF can favour the growth and nutrient uptake in *A. elata* seedlings without any toxicity symptom.

Materials and Methods

Study site

This study was carried out in the Laboratory of Combined Manipulation of Illumination and Fertility on Plant Growth (Zhilunpudao Agric. S&T Ltd., Changchun, China) (43°58' N, 125°24' E). Seedlings were cultured in a specially manufactured aluminum-profile frame of 2.0 m × 1.2 m × 1.4 m (length × width × height). The inside space of the frame was divided into two halves by a black-out cloth to enable seedlings in both sides to receive LED or HPS lightings. Sunlight was thoroughly isolated from the frame by black-out curtains.

Plant materials

One-year old *A. elata* bare-root stocks were derived from a nursery at Tieli, Yichun, Heilongjiang, Northeast China. The average initial height and RCD of these seedlings were measured to be 4.0 cm and 0.5 cm, respectively. Most roots were approximately 30.0 cm in length and all roots were cut to retain a length of approximately 7.0 cm. To promote the main stem growth, all lateral braches' residuals were excised of from the stem but apical buds at the end of aerial tip of the stem were reserved. Finally, 192 uniform-sized bare-root stocks were used for the study.

Experimental design

The experiment was conducted by a factorial design with the lighting spectra and EF treatment as the two factors and within each factor two levels were incorporated. The photosynthetic photon flux rate (PPFD) was measured to be 60~64 μmol m^{-2} s^{-1} at 10 cm above the floor. As seedlings grew the PPFD at the apical bud would increase up to the maximum of 200 μmol m^{-2} s^{-1} due to the decline of distance between the seedling tip and lighting sources. Hence our seedlings would receive sufficient illumination intensity according to Apostol *et al.* (2015) and (Li *et al.* 2018). The higher dose of the total amount of N delivered to *A. elata* seedlings was set to be 160 mg seedling^{-1}. This dose fell in the optimum range of amounts (150~175 mg N seedling^{-1}) for raising containerized *Quercus ilex* seedlings suggested by Uscola *et al.* (2015). The lower dose in this study was chosen as the half of the high dose (80 mg seedling^{-1}) and a little lower than the suggested amount of N application to containerized *Podocarpus macrophyllus* seedlings (Wei *et al.* 2013).

Light spectra manipulation

The diodes emitting red (R) (600–700 nm), green (G) (500–600 nm), and blue (B) (400–500 nm) lights were welded to a panel (0.9 m×1.2 m, length × width) to emit a spectrum of 85:10:5 (R/G/B) in PPFD at the maximum electric current. This light spectrum was proven to be effective for tree seedling growth by trials of Apostol *et al.* (2015) and Li *et al.* (2018). In one half part of the frame, two HPS lamps (Zhilunpudao Agric. S&T Ltd., Changchun, China) 80 cm above the ground and the PPFD was measured to be 60~64 μmol m^{-2} s^{-1} 70 cm beneath the lamp. In the other half of the frame the illuminating intensities of the R and G+B lights from LEDs were controlled by two transformers in output-powers of 200-W and 150-W, respectively. To keep a similar PPFD between the two frame-halves the electric current for the R and G+B lights from LEDs was adjusted to be 10 and 20% of the ordinary level, respectively. As a result, the percent proportion of R/G/B spectra for HPS and LED lightings were measured to be 44.4:53.8:1.8 and 30.0:69.3:0.7, respectively 10 cm above the floor. The absolute spectral values in response to the continuous wavelength for the two light spectra are shown in Fig. 1.

Seedling culture and fertilizer regime

Containers were filled with commercial seedling culture substrate (Mushro-Dust^{TM}, Zhiluntuowei A&F S&T Ltd., Changchun, China), wherein peat, perlite, and spent

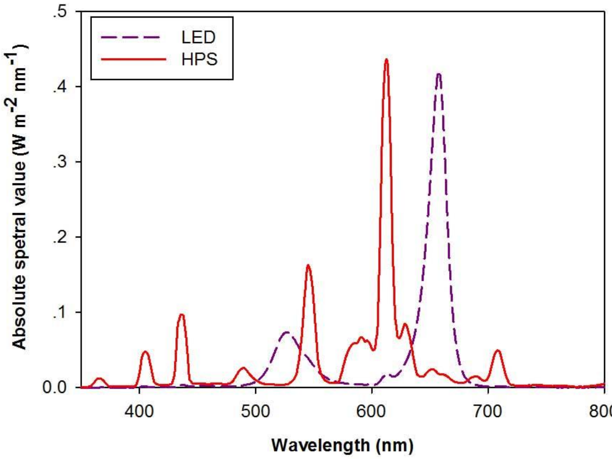

Fig. 1: The absolute spectral values of spectra from LED and HPS lightings for the culture of *A. elata* seedlings

mushroom residue were mixed to the volume ratio of 55:20:25 (v/v/v). The substrate was measured to have ammonium N of 120 mg kg^{-1}, nitrate N of 140 mg kg^{-1}, available P of 365 mg kg^{-1}, pH of 4.5, and EC of 1.0 dS m^{-1}. On April 16, 2018, seedlings were planted to 32-plug (4×8) containers in the size of 30 cm × 53 cm (width × length) by 2 plugs ×2 plugs spacing for one seedling. Therefore, 16 seedlings were planted in one container and totally 192 seedlings were planted in 12 containers for this study. Six containers were randomly chosen and placed in one frame half and the other six containers were placed in the other half. Either group of the six containers received one of the two lighting spectra. Three containers in one group received low dose EF and the other three containers in the same group as another three replicates for the high dose EF treatment. Since April 23, 2018, seedlings were fertilized by the solution of ammonium sulfate (NH_4SO_4) and mono-potassium (K) phosphate (KH_2PO_4) which contained N-P-K in the ratio of 12-9-12 over the 16 applications. Seedlings were fertilized once a week and the experiment was conducted in four months. The classical EF model was used in this study to calculate the weekly application dose (Wei *et al.* 2013; Li *et al.* 2017; 2018) with the given total amount of nutrient in preceding paragraphs. Temperature and relative humidity (RH) were monitored since the start of the experiment and weekly with every fertilizer applications. Throughout the experiment, the temperature ranged from 15.3°C to 36.1°C and RH ranged between 28 and 99%.

Seedling harvest and measurement

In late August of 2018, all seedlings were harvested and bulked as a measuring unit for 16 individuals per container. After measuring height and RCD, 12 seedlings per container were further divided into shoot and root parts by excising at the root-collar. Both parts were dried in an oven at 68.0°C for three days (72 h). Dried samples were measured for biomass, grounded manually, and nitrogen (N) and phosphorus (P) concentrations were determined using the Kjeldahl and ICP-OES determination methods, respectively (Wei *et al.* 2013; Li *et al.* 2017; Wang *et al.* 2017; Li *et al.* 2018). The other four seedlings were defoliated and fresh leaves were collected. Two leaves were randomly defoliated from each common twig at the middle of the whole stem length. Eight leaves were collected for one replicate unit. Leaves were scanned to generate a projected image at the resolution of 118.11 pixels cm^{-1} (HP Deskjet 1510 scanner, HP Inc., Palo Alto, CA, USA). The scanned image was opened in Photoshop (ver. 8.0, Adobe® Systems Incorporated Inc., San Jose, California, U.S.A.) and all colors in the background were removed (Fig. 2). The histogram information was read by navigator panel in Photoshop and the average value for all pixels through the green-color channel was recorded to evaluate the green color index (GCI) of the leaves. The histogram information was also used to determine the leaf area (LA) by the following calculation:

$$\text{LA} = \frac{Pixel_{all}}{Res.^2 \times Leaf_{number}} \qquad (1)$$

Where, $Pixel_{all}$ is the whole pixel value for the projected area of eight leaves, *Res.* is the resolution of the scanned image which is taken as 118.11 pixels cm^{-1} in this study, $Leaf_{number}$ is the number of leaves (n=8).

Statistical analysis

S.A.S. software (ver. 9.4 64-bit, S.A.S. Institute Inc., N.C., U.S.A.) was used to analyze the effects of spectra, fertilizer regime, and their interaction on measured parameters through the GLM procedure. Values indicating significant effects were indicated by analysis of variance (ANOVA) at the 0.05 level and compared by the Duncan test. Vector analysis was employed to diagnose the nutritional state of N and P in the shoots according to the methodology used by Li *et al.* (2018). Pearson correlation was employed to analyze the relationship between two leaf indices (green color index and leaf area) and other measured parameters.

Results

Seedling growth and biomass accumulation

The lighting spectra and fertilizer regime treatments had an interactive effect on height and root biomass in *A. elata* seedlings (Table 1). EF at the rate of 160 mg N $seedling^{-1}$ promoted height growth to be 58.0 cm under the LED spectrum, which was higher than in the same N rate of EF treatment under the HPS spectrum by 22% and higher than the EF treatment at 80 mg N $seedling^{-1}$ under the LED spectrum by 37%. Combined effects of EF at 160 mg N $seedling^{-1}$ under the LED spectrum also resulted in the greatest root biomass accumulation, which was higher than

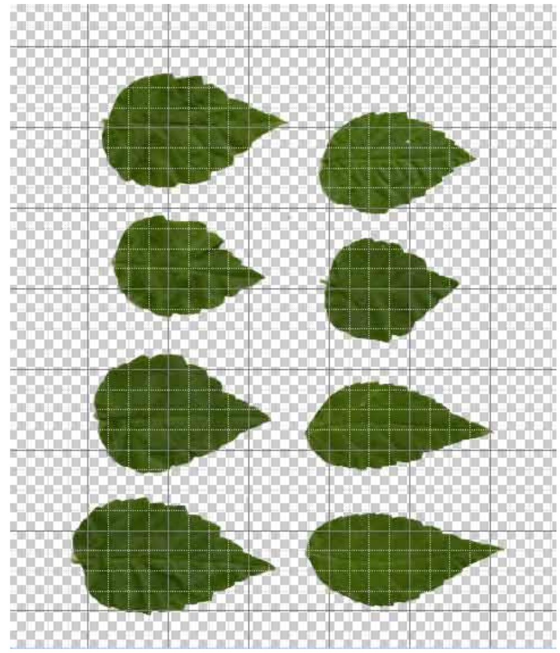

Fig. 2: The layout of analysis on scanned *A. elata* leaves for foliar green-color index and leaf-area through calculating the extracted pixel values from histogram in Photoshop software

in the other three treatments by 54–80%. The higher dose of EF treatment also resulted in higher RCD (high, 0.65 ± 0.05 cm; low, 0.54 ± 0.0 cm). However, RCD was promoted by the HPS spectrum (0.64 ± 0.07 cm) relative to the LED spectrum (0.55 ± 0.07 cm). Shoot biomass was greater in the higher rate of EF treatment (high, 4.09 ± 1.30 mg; low, 2.02 ± 0.57 mg), but root to shoot biomass ratio (R/S) showed the contrasting results (high, 0.14 ± 0.02; low, 0.23 ± 0.07).

N and P concentrations and contents

Shoot N concentration was highest in the EF treatment at 160 mg N $seedling^{-1}$ under the HPS spectrum, but lowest at 80 mg N $seedling^{-1}$ under the same spectrum (Fig. 3A). Treatments of lighting spectra and fertilizer regime had no effect on root N concentration (Table 2). Shoot P concentration was highest in the EF treatment at 80 mg N $seedling^{-1}$ under the HPS spectrum and lowest in the EF treatment at the same rate but under the LED spectrum (Fig. 3B). Root P concentration was lower in the EF treatment of 80 mg N $seedling^{-1}$ under the HPS spectrum or in the EF treatment of 160 mg N $seedling^{-1}$ under the LED spectrum than the other two treatments (Fig. 3B).

Shoot N content was higher in the EF treatment at 160 mg N $seedling^{-1}$ under HPS and LED spectra than in the other two treatments (Fig. 4A). Root N content was highest in the EF treatment at 160 mg N $seedling^{-1}$ under the LED spectrum. P content only responded to treatments in shoot

Table 1: Growth and biomass accumulation in *Aralia elata* (Miq.) Seem seedlings cultured by exponential fertilization (F) at rates of 160 (N160) and 80 mg N seedling^{-1} (N80) under lighting spectra (L) from high-pressure sodium (HPS) lamps and light emitting diode (LED) panels

Parameters	HPS		LED		*Pr > F*		
	N160	N80	N160	N80	L	F	L×F
Height (cm)	47.62 ± 2.72b[1]	49.78 ±5.60ab	58.24 ± 6.99a	42.36 ± 2.24b	0.5994	**0.0476**[2]	**0.0153**
RCD (cm)	0.68 ± 0.01	0.60 ±0.09	0.61 ± 0.05	0.49 ± 0.01	**0.0180**	**0.0102**	0.4982
Shoot biomass (g)	3.32 ± 0.77	2.48 ±0.15	4.87 ± 1.53	1.56 ± 0.54	0.5795	**0.0054**	0.0545
Root biomass (g)	0.45 ± 0.12b	0.44 ± 0.12b	0.70 ± 0.09a	0.39 ± 0.09b	0.1877	**0.0471**	**0.0409**
R/S[3]	0.13 ± 0.02	0.18 ± 0.04	0.15 ± 0.03	0.27 ± 0.09	0.1557	**0.0372**	0.2957

[1] Different letters in a horizontal row indicate significant difference according to Duncan test at 0.05 level; [2] Bold values indicate significant effect; [3] R/S, root to shoot biomass ratio

part, where the EF treatment under the LED lighting resulted in the highest P content (Fig. 4B).

Foliar characteristics

GCI was higher in the EF treatment at 80 mg N seedling^{-1} under LED lighting than in the EF treatment at 160 mg N seedling^{-1} under both spectra (Fig. 5A). However, LA was highest in the EF treatment at 160 mg N seedling^{-1} under LED spectrum (Fig. 5B). GCI had a negative relationship with RCD, shoot biomass, and shoot N concentration; while LA had a positive relationship with shoot and root biomass (Table 3).

Nutritional symptom diagnosis

Under the HPS spectrum, higher rate of EF countered the N limit by increasing both shoot biomass and N concentration (Fig. 6A). Under the LED spectrum, higher rate of EF can induce luxury N consumption due to increased N content without any change of N concentration. With HPS spectrum as the reference, LED spectrum induced excessive N toxicity at low rate of EF and inherent N dilution at high rate of EF (Fig. 6A).

Likely, seedlings under the LED spectrum had steady-state P uptake by higher rate of EF compared to those under the HPS spectrum (Fig. 6B). However, under the HPS spectrum higher rate of EF induced P dilution. Compared to the HPS spectrum, the LED spectrum induced excess P symptom and P dilution in the EF treatment at low and high rates, respectively (Fig. 6B).

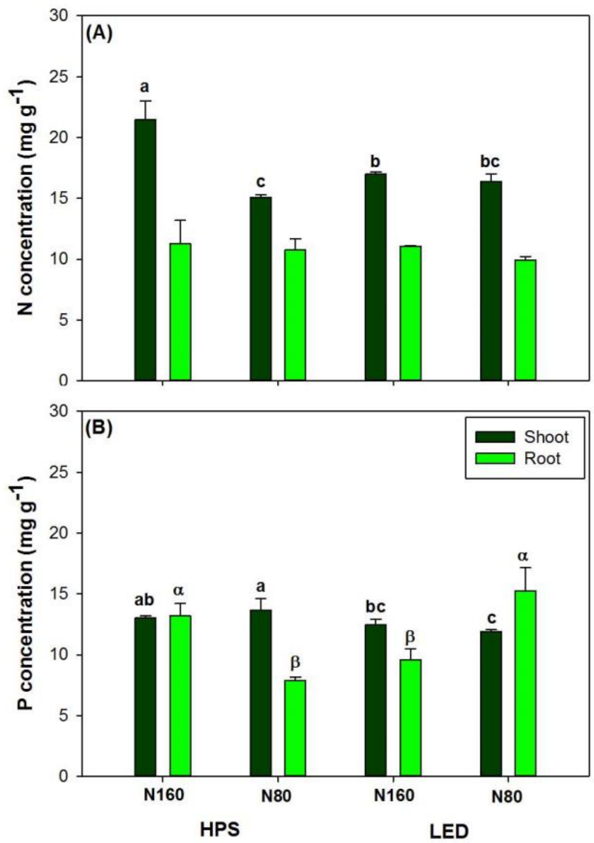

Fig. 3: Nitrogen (N) and phosphorus (P) concentrations in shoot and root parts of *A. elata* seedlings cultured in exponential fertilization treatments at rates of 80 (N80) and 160 mg N seedling^{-1} (N160) under HPS and LED lighting spectra. Different letters indicate significant difference at 0.05 level according to Duncan test. Lower-case letters of a, b, and c are marked for shoot part and roman-letters of αand β are marked for root part

Discussion

The EF treatment at the rate of 160 mg N seedling^{-1} under the LED lighting spectrum resulted in the highest seedling-height growth and root biomass accumulation. LED spectrum was also reported to promote height growth in *Dalbergia odorifera* seedlings compared to the HPS spectrum (Li *et al.* 2018) in spite of the contrasting results regarded in *Picea abies* and *Pinus sylvestris* seedlings (Riikonen 2016; Riikonen *et al.* 2016). The increment of height growth in *A. elata* seedlings under the LED lighting compared to the HPS lighting only occurred when seedlings were exposed to the high dose of EF treatment. The nutritional condition was also critical for the difference of root biomass between the two lighting spectra. Similar to height growth results, our findings on root biomass in tree seedlings under different lighting spectra were highly variable (Apostol *et al.* 2015; Riikonen 2016; Riikonen *et al.* 2016; Li *et al.* 2018). The RCD response in previous studies, however, agrees to our study in that quite rare change can be found among treatments (Riikonen 2016; Riikonen *et al.* 2016). Apostol *et al.* (2015) found that RCD growth in

Table 2: *P* values from analysis of variance (ANOVA) on effects of light spectra (L), exponential fertilization (F), and their interaction (L×F) on nitrogen (N) and phosphorus (P) concentration and content in *Aralia elata* (Miq.) Seem seedlings

Nutrient element	Organ	L	F	L×F
N concentration	Shoot	**0.0155**[1]	**0.0001**	**0.0005**
	Root	0.4462	0.2506	0.6506
P concentration	Shoot	**0.0076**	0.9231	**0.0271**
	Root	**0.0284**	0.7944	**<0.0001**
N content	Shoot	0.9356	**0.0010**	**0.0102**
	Root	0.2006	**0.0171**	**0.0151**
P content	Shoot	0.8697	**0.0084**	**0.0220**
	Root	0.1012	0.1291	0.3606

[1]Bold values indicate significant effect

Table 3: Pearson correlation between foliar indices (Green color index and leaf area) and parameters in *Aralia elata* (Miq.) Seem seedlings

Nutrient element		Green color index	Leaf area
Height	*R*	-0.3154	0.4706
	P	0.3180	0.1225
RCD[1]	*R*	**-0.7696**[2]	0.0633
	P	**0.0034**	0.8438
Shoot biomass	*R*	**-0.5884**	**0.5775**
	P	**0.0442**	**0.0493**
Root biomass	*R*	-0.3532	**0.6706**
	P	0.2601	**0.0170**
Shoot N concentration	*R*	**-0.6533**	-0.0367
	P	**0.0212**	0.9098
Root N concentration	*R*	-0.03797	-0.0895
	P	0.9067	0.7821
Shoot P concentration	*R*	-0.2466	-0.2773
	P	0.4397	0.3828
Root P concentration	*R*	0.1504	-0.1599
	P	0.6409	0.6197

[1]RCD, root-collar diameter; [2] Bold values indicate significant effect

coniferous tree seedlings was slightly modified by lighting spectra for some species and seed sources but either HPS or LED treatment can promote RCD growth. Li *et al.* (2018) reported that RCD of *D. odorifera* seedlings did not respond to the spectra treatment unless combined with the addition of chitosan oligosaccharide solution. Therefore, it can conclude that the response of RCD to lighting spectra depended on the interactive effects with other manipulations. The screening for species, seed source, and addition of polymer compound all can be alternative approaches to interact with spectra in affecting RCD growth, but the involvement of fertilizer regime needs to be confirmed by more studies.

The seedling shoot part has also been employed to diagnose the whole-plant nutritional status for *Picea mariana* (Salifu and Timmer 2003), *Quercus rubra* (Birge *et al.* 2006; Salifu and Jacobs 2006), *Q. alba* (Birge *et al.* 2006). *A. elata* seedlings exposed to EF treatment at 160 mg N seedling^{-1} under LED lighting did not have different shoot N concentration compared to that under lower dose of EF treatment under the same lighting spectrum. However, shoot N concentration in both treatments was lower than in seedlings receiving high-N EF treatment under the HPS

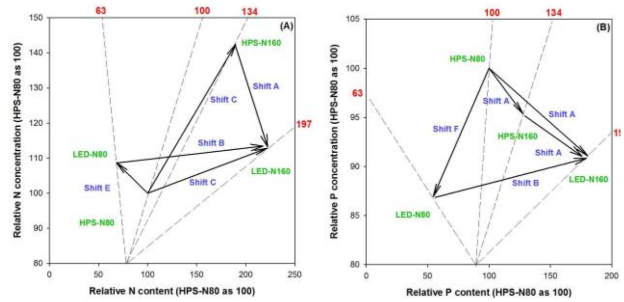

Fig. 6: Vector diagnosis of nutritional state for N (A) and P (P) in the shoot part of *A. elata* seedlings cultured in exponential fertilization treatments at rates of 80 (N80) and 160 mg N seedling^{-1} (N160) under HPS and LED lighting spectra. Vector shifts indicate the possible symptom of relative nutrient state adapted from Salifu and Timmer (2003). Shift A, nutrient dilution; shift B, steady-state uptake; shift C, nutrient deficiency; shift E and F, excess nutrient supply to toxicity

lighting. The uptake and allocation of P showed similar trend as N among treatments. Actually, under the LED lighting spectrum the higher dose of EF had induced steady-state uptakes of N and P in seedlings compared to the lower dose EF treatment. This was mainly caused by steady accumulation of shoot biomass accordingly with stem elongation but the meanwhile change of nutrient concentration was not significant. The high EF-dose of 160 mg N seedling^{-1} in present study was adapted from Uscola *et al.* (2015), wherein the dose range of 150~200 mg N seedling^{-1} induced luxury consumption for N and P in *Q. ilex* seedlings. However, with the high dose of EF treatment the LED lighting induced a dilution symptom in seedlings for N and P compared to the HPS lighting by accumulating biomass without sufficient nutrient uptake to shoot (Fig. 6A). Previous studies also found that lighting environment can modify nutritional state of tree seedlings through adjusting biomass accumulation and nutrition concentration (Wei *et al.* 2013; Li *et al.* 2017). Although the LED lighting can promote seedling height growth and biomass accumulation compared to the HPS lighting, the dose of 160 mg N seedling^{-1} delivered by EF was more proper for seedlings under HPS lighting and insufficient for seedlings under LED lighting.

Although, root N concentration did not respond to any of the treatments, root N content changed and reached the highest value in the EF treatment at 160 mg N seedling^{-1} under LED lighting. This corresponded to the root biomass trend among treatments because nutrition content is the product of concentration and biomass. Root P concentration showed some contrasting results. The higher rate of EF treatment can promote P concentration in roots under the HPS lighting only, but under the LED lighting root P concentration was decreased. These results can be supported by the nutritional symptom diagnosis, which indicated that higher dose of EF treatment induced steady-state P uptake to shoots under LED lighting but P dilution in shoot under

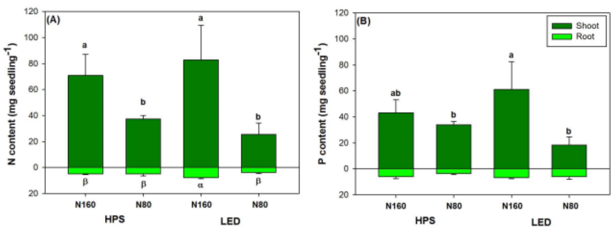

Fig. 4: Nitrogen (N) and phosphorus (P) contents in shoot and root parts of *A. elata* seedlings cultured in exponential fertilization treatments at rates of 80 (N80) and 160 mg N seedling^{-1} (N160) under HPS and LED lighting spectra. Different letters indicate significant difference at 0.05 level according to Duncan test. Lower-case letters of a, b, and c are marked for shoot part and roman-letters of α and β are marked for root part

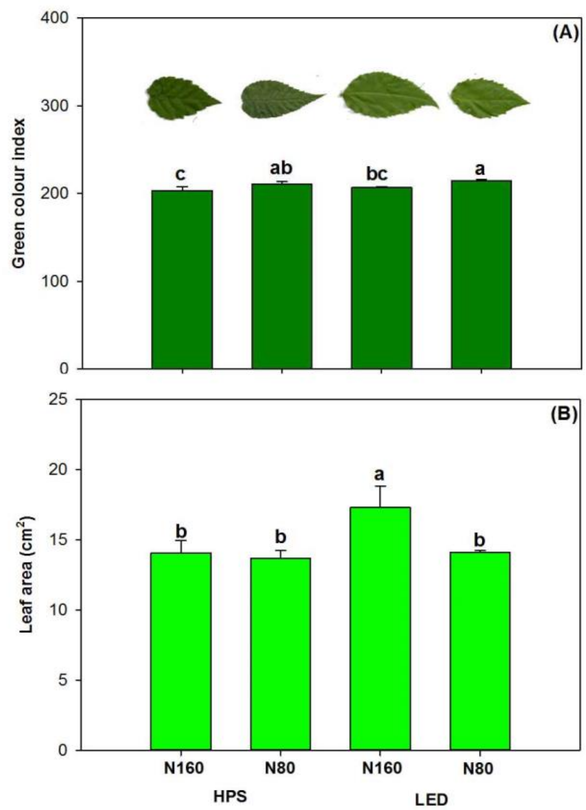

Fig. 5: Foliar indices of green-color index (**A**) and leaf area (**B**) in *A. elata* seedlings cultured in exponential fertilization treatments at rates of 80 (N80) and 160 mg N seedling^{-1} (N160) under HPS and LED lighting spectra. Different letters indicate significant difference at 0.05 level according to Duncan test

HPS lighting. Therefore, when facing higher nutrient availability LED spectrum tended to induce more P allocation to the shoot part but the HPS spectrum appeared to be insufficient for this inducing-effect. On the other hand, at the low P fertility of 80 mg N seedling^{-1}, higher P concentration was found in roots of seedlings under the LED lighting than the HPS lighting, while shoot P was indicated to be over-supplied under LED spectrum compared to the HPS spectrum. This was because LED spectrum tended to detain P in root at low fertility hence resulted in insufficient P allocated to the shoot part to be involved in the biomass production. In contrast, when P fertility was enhanced by the 160 mg N seedling^{-1} treatment, the LED spectrum had the condition to promote biomass production hence diluted root P concentration and induced shoot P state. Because quite rare studies have reported the interactive effects of EF and lighting spectra on P uptake and allocation (Li *et al.* 2018), it is hard to compare our results about nutritional response with others. Further work is needed to test this combined effect on more tree species with more lighting and fertility manipulations.

Therefore, it is reasonable to accept first hypothesis but partly accept the second one. Higher dose of nutrient supply through EF can improve growth and nutrient uptake under the LED lighting, while in the HPS spectrum seedling growth was not changed.

Because many NWFP plants grow on the understory floor and distribute in moist and highly-shaded environment, to predict their nutrient state is of practical meaning for the efficiently developing the target species from the natural habitat. The foliar indices of area and green-color degree are two direct parameters that can be obtained easily and have several physiological meanings. Rabara *et al.* (2017) was the first to reveal the green-color degree in leaves of artichoke seedlings exposed to a range of artificial lighting spectra. However, authors therein did not further analyze the nutrient uptake and allocation and the relationship between green-color and nutritional state. Zhu *et al.* (2019) studied the relationship between foliar GCI and shoot N concentration and found a significant negative correlation in pepper (*Capsicum annum* L.) seedlings. Xu *et al.* (2019) reported that leaf area had positive relationship with nutrient status instead of GCI. In the present study, it was found a negative relationship between GCI and shoot N concentration. These results together suggest that the darker of the green color in leaves the lower N concentration can be found therein. In addition, it was found negative relationships with GCI and RCD and shoot biomass. The responses of these two indices resulted from the corresponding decline of N indicated by higher GCI. On the other hand, the largest leaves with highest leaf area were found in seedlings exposed to the EF treatment at 160 mg N seedling^{-1}. The leaf area in present study was largest in seedlings exposed to the EF treatment at 160 mg N seedling^{-1} under the LED lighting, wherein seedlings had highest shoot height and root biomass. Foliar area was positively to biomass accumulation in both shoot and root parts. These results together suggest that the leaf area increased accordingly with biomass accumulation and shoot growth. For broad-leaved species, the absorption of direct light scales with leaf area (Niinemets 1999).

Conclusion

To accelerate the use of *A. elata* as a NWFP species for regeneration in the understory community, its seedlings were suggested to raise by exponential fertilization (EF) at the rate of 160 mg N seedling^{-1} (N-P-K, 12-9-12) under LED lighting spectrum (percent R/G/B, 30:69.3:0.7). This treatment can not only result in the greatest growth with shoot height of ~60 cm and root biomass of 700 mg but also induced higher N and P contents in the shoot part. This treatment can also induce a steady-state nutrient uptake for N and P in *A. elata* seedlings compared to lower dose of EF in the same LED spectrum. Foliar indices can be used to predict seedling growth and nutritional state. Higher foliar green-color degree suggested less shoot N concentration but larger leaf area indicated greater biomass accumulation. Future work is suggested to test the transplanted performance of *A. elata* seedlings to the montane field so as to determine whether the cultural tendency in response to treatments would be maintained among treatments. In addition, more NWFP species should be tested by the methodology of the current study but with more manipulations on fertilizer-application and artificial-lighting regimes.

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