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Influence of Container Type and Growth Medium on Seedling Growth and Root Morphology of *Cyclocarya paliurus* during Nursery Culture

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Abstract: As a multiple function tree species, *Cyclocarya paliurus* (Batal) Iljinskaja is mainly planted and managed for timber production and medical use. To improve the seed use efficiency and outplanting performance of *C. paliurus*, the effects of container types and growth medium on the seedling growth and root morphology of *C. paliurus* were investigated by using a completely randomized block experimental design with a 4 × 3 factorial arrangement during nursery culture. Both container type and growth medium significantly affected the growth, biomass, and root morphological indexes of *C. paliurus* seedlings, but container size had a greater effect on the seedling quality of *C. paliurus* than the growth medium formula. The root-collar diameter and height of the seedlings were positively and significantly correlated with the biomass variables and root morphological variables, and could be considered essential attributes for evaluating seedling quality. Based on the results from this study, the management regime used here in *C. paliurus* seedling production is suggested to ensure good quality seedling delivery. Our study provides not only valuable insights into the container seedling culture of *C. paliurus*, it also enables nursery managers to optimize seedling production.

Keywords: *Cyclocarya paliurus*; container size; medium formula; morphological criterion; root-to-shoot biomass; seedling quality

1. Introduction

Seedling quality is critical to the ability of seedlings to survive extended environmental stresses and produce vigorous growth following outplanting. Rapid early growth of tree seedlings is closely related to the success of plantation establishment [1]. It has been reported that larger seedlings have better performance than small seedlings after planting under conditions of competition for resources with weeds [2–4], while decreasing survivals with increasing seedling sizes were observed on extremely harsh sites [5,6]. The morphological criteria for assessing seedling quality can be manipulated by nursery cultural practices [7], and the impact of nursery management practices on seedling quality has been studied on various plant species [8–11]. However, most of these studies were conducted in research station settings far from actual nursery production conditions [7].

Many advantages of container seedlings over bareroot stock have been reported [12,13], such as quick production, uniform size, extending the planting season, and performing well on adverse sites. Moreover, container planting offers potentially increased seed efficiency (the ratio of plantable seedlings to filled seeds), because growing conditions can be better controlled, which is very important for the tree seeds with low fertilization and germination. However, some disadvantages also exist in the production and use of container seedlings, such as nutritional imbalances and higher cost [14]. During the container seedling production, container size, growth medium, growing density, and design characteristics of the containers are important determinants of seedling quality [11,13], but are varied among the tree species [13]. Generally, the type and volume of the containers are some of the most important characteristics, because they have both major and direct impacts on seedling quality and production costs. Meanwhile, the optimum container size varies according to many different factors, including species, growing density, environmental conditions, and length of the growing season. Furthermore, one of the most serious problems in containers (especially for the seedlings with tap roots) is the tendency of root spiraling around the inside of the container when round, smooth-walled plastic containers were used [13], which can seriously reduce seedling quality and field performance after planting. A suitable growth medium for container seedlings is critical in order to provide support and nutrients, retain water, and allow oxygen diffusion to the roots. Although there is not an ideal growth medium for all growing plants, a growth medium should incorporate physical, chemical, and biological requirements for good plant growth together with the requirements of practical plant production [15]. Peat moss is used extensively as a substrate in container seedlings; however, in recent years, there has been increasing environmental and ecological concerns against the use of peat due to endangered bog ecosystems [16]. Moreover, due to the increased cost and decreased availability of peat moss in many countries, numerous organic materials (such as manure compost and rice hulls ash) have been sought and studied worldwide [14,17,18]. Recently, seedling growers have been looking for more local growth medium components to reduce the increased transportation costs of growing media [14,19].

As a highly valued and multiple-function tree species, *Cyclocarya paliurus* (Batal) Iljinskaja belongs to the Juglandaceae family, and is naturally distributed in the mountainous regions of subtropical China [20]. Many studies have demonstrated that *C. paliurus* not only has a great potential for fine timber production [21], the extracts of its leaves also possess a variety of bioactivities, such as hypoglycemic [22,23], anti-hyperlipidemia [24], anti-HIV-1 [25], antioxidant [23,26], and anticancer [27]. Therefore, a huge production of leaves is required for *C. paliurus* tea production and medical use. However, most of the *C. paliurus* resources are distributed in natural forests, and there are not enough *C. paliurus* plantations for leaf production [28]. Currently, *C. paliurus* can be only propagated from seeds, but the seeds have pronounced dormancy [20], and about 75% seeds are "empty" due to the asynchronous flowering period [29]. In order to improve the seed efficiency and outplanting performance of *C. paliurus*, producing container seedlings is one of the most important options to quickly increase plantation resources of *C. paliurus*, due to its flexibility of seedling production and field planting. However, the influence of nursery cultivation regimes on container seedling quality and the outplanting performance of *C. paliurus* has received almost no attention, and no study on seedling growth and root morphology of the species has been reported until now. The objectives of this study were to determine the effects of container type and growth medium on seedling growth and root morphology of *C. paliurus* during nursery culture and to select an optimum container type and growth medium for the seedling production of this species.

2. Materials and Methods

2.1. Plant Material and Experimental Layout

The seeds used in this study were collected in October 2013 from the dominant trees of the *C. paliurus* natural population located at the Tonggu site in Jiangxi Province of China (approximately 28°35′ N latitude, 114°22′ E longitude). To overcome the dormancy, the collected seeds were subjected to exogenous GA3 (gibberellin A3) and stratification treatments in early January 2014. After about 15 months of stratification treatment, the germinated seeds were sown into each container in 2015.

A randomized complete block design with a factorial treatment structure was conducted and carried out with three replications. Factors and levels were: container type and size with four levels

(C1: non-woven container with 760 mL; C2: non-woven container with 500 mL; C3: non-woven container with 280 mL; C4: black plastic container with 500 mL, all bought from Anqing Yike Seedling Company, Anqing, China), and growth medium with three levels (yellow subsoil: perlite: fermented chicken manure: peat by volume = 2:2:1:5 (F1); 2:2:2:4 (F2); 2:2:3:3 (F3)). Yellow subsoil was the soil under the depth of 20 cm collected from a field near the experimental site, which is the most readily available soil type in the region, with an organic content of 2.4 g kg^{-1}, available N content of 0.98 mg kg^{-1}, available P content of 10.1 mg kg^{-1}, available K content of 41.8 mg kg^{-1}, and pH value of 5.5. Fermented chicken manure (chicken manure fermented with agricultural residues) was bought directly from Nanjing Woyou Biological Fertilizer Company Limited (Nanjing, China), with an organic content of 45.0%, total N content of 3.5%, P content (P_2O_5) of 6.4%, K content (K_2O) of 1.6%, and pH value of 6.7. The levels of all of the heavy metals in the manure were within the permissible limits of Chinese agricultural standard for Microbial Organic Fertilizers (NY 884-2012). The peat was turfy peat with medium structure and the perlite was of coarse structure. After mixed according to the formula designed, the growth medium was sieved to 5 mm, and then filled in the containers by hand.

The experiment was conducted in an open-sided shed with shading nets and auto-spray irrigation systems at the Baima Research Base of Nanjing Forestry University (about 31°35′ N latitude, 119°10′ E longitude). After one month of growing, the container seedlings were moved up the overhead iron-wire grid in order to incorporate air root pruning, and the growing density was 100 container seedlings m^{-2}. There were 90 seedlings (30 seedlings for each replication) for each treatment, and a total of 1080 container seedlings were used in the experiment. During the experimental period, normal management practices were carried out, but no fertilization was applied.

2.2. Growth Medium Sampling and Measurement

After preparing the growth media, the different component materials were mixed by hand, according to the designed formula. Then, four samples of each growth medium were taken for the estimation of the physical and chemical properties prior to filling the containers. The pH values of the growing media were measured in a soil–water suspension (soil:water = 1:2.5) using an automatic titrator (SH/T0983, Changsha, China), and the bulk density of the growing media was determined by the cylindrical core method [30]. For the measurements of total N, P and K, the growth medium materials were digested using 5 mL concentrated H_2SO_4 and 1 mL concentrated $HClO_4$ at 120 °C for 30 min, and then at 360 °C until the digest was clear. After digestion, the N and P concentrations in the digest were determined using a flow-injection analyzer (BRAN+LUEBBE, Hamburg, Germany), whereas the K concentration in the digest was analyzed using a Hitachi108-80 atomic adsorption spectrometer (Hitachi Ltd., Tokyo, Japan). Organic matter in the growing media was measured by oxidation with potassium dichromate [31].

2.3. Plant Sampling and Measurement

The morphological characteristics of seedling height and root collar diameter were measured on every seedling grown for the experimental phase of this study at the interval of 30 days (n = 1080). Additionally, based on the mean values of seedling height and root-collar diameter that were measured in each treatment, root dry mass, shoot dry mass, and root morphological indexes were measured on a subset of seedlings (n = 36) on 16 October 2015 using destructive sampling techniques. After carefully washing root systems free of all media, biomass was measured after the separated roots and shoots were dried at 60 °C for 72 h. All of the root morphological indexes (total root length, root surface area, root volume, and mean root diameter) were determined and analyzed by WinRHIZO.PRO systems (Version 2007, Regent Instruments Inc., Quebec, QC, Canada). Using the values measured, the ratios of root-to-shoot biomass and height-to-diameter were calculated for all destructively sampled seedlings.

2.4. Statistical Analysis

All of the statistics were performed using the SPSS statistical software package version 15 (SPSS Inc., 2005, Chicago, IL, USA), and the results are reported as mean ± standard deviation. Distribution was tested for normality by Kolmogorov–Smirnov criterion, and the homogeneity of variances was tested by Levene's test. In order to determine the main effect, as well as identify whether there were significant interactions between container type and growth medium for the measured indexes, a two-way analysis of variance (ANOVA) was adopted. When the interaction was not significant, the one-way ANOVA was conducted for each treatment, and followed by least significant difference tests (LSD) at $p < 0.05$. However, if the interaction was significant, the one-way ANOVA was only conducted to determine whether the integrated effect of container types and growing media significantly affected seedling growth and root morphology at $p < 0.05$. Associations between seedling morphological variables were determined by using Pearson's correlation analysis, while a regression analysis was conducted between seedling growth and biomass.

3. Results

3.1. Properties of the Growing Media

There were significant differences in the physical and chemical properties among the three growth media, except for bulk density (Table 1). The pH was between 6.11 and 6.66, while the highest appeared in the F3 formula. However, the bulk density ranged from 0.73 to 0.85 g cm^{-3}, and the highest bulk density (0.85 g cm^{-3}) was observed in the F1 formula. Generally, when the proportion of the fermented chicken manure increased and peat proportion decreased, the total nutrient contents of N, P, and K as well as pH values significantly increased, but organic matter content was lowered. When compared with the F1 formula, the total contents of N, P, and K increased by 44.0%, 183.8%, and 15.8% in the F2 respectively, while enhanced by 76.0%, 273.7%, and 24.2% in the F3 formula.

Table 1. The physical and chemical properties of three growth media.

Growth Medium	pH Value	Bulk Density (g cm^{-3})	Organic Matter (g kg^{-1})	Total Nitrogen (g kg^{-1})	Total Phosphorus (g kg^{-1})	Total Potassium (g kg^{-1})
F1	6.11 ± 0.04 b	0.85 ± 0.04 a	75.58 ± 0.45 a	0.50 ± 0.60 c	0.99 ± 1.91 c	8.25 ± 0.27 c
F2	6.44 ± 0.02 a	0.77 ± 0.07 a	73.3 ± 0.30 b	0.72 ± 1.52 b	2.81 ± 4.10 b	9.55 ± 0.15 b
F3	6.66 ± 0.13 a	0.73 ± 0.09 a	71.39 ± 0.41 c	0.88 ± 2.21 a	3.70 ± 4.66 a	10.25 ± 0.1 a

Within the same column, the means followed by different letters are statistically different ($p < 0.05$, least significant difference (LSD) test).

3.2. Seedling Growth and Biomass

Two-way analysis of variance showed that both growth media and container types significantly affected seedling growth and seedling biomass, but no significant effect was observed for the ratio of root-to-shoot biomass (Table 2). Based on the F-values in Table 2, container size had larger effects on seedling growth and biomass production of *C. paliurus* than the growth medium. Meanwhile, a significant interaction between growth media and container types was detected for seedling height, root-collar diameter, height-to-diameter ratio, and root-to-shoot biomass ratio, whereas no significant interaction was observed for the shoot, root, or total biomass of the seedlings (Table 2).

As growing approached seven months, the seedling height ranged from 21.3 to 63.9 cm, while the root-collar diameter and height-to-diameter ratio varied from 5.2 to 8.2 mm and from 39.0 to 81.2, respectively. One-way ANOVA indicated that the effect of the growth medium and container type on seedling growth and height-to-diameter ratio was significant (Table 3, $p < 0.05$). It is very obvious that the larger container size could get great seedling growth under the same growth medium, whereas the F2 growth medium was relatively favorable to seedling growth of *C. paliurus* at the same container size. Moreover, it is noteworthy that there was a significant difference in the seedling growth between the same size containers with various textures (C2 and C4) in some cases (Table 3). For example, the seedling height in a non-woven container with 500 mL (C2) was 19.8% higher than that in the plastic container of same size (C4) on the growth medium of F1, while root-collar diameter on the F2 growth medium was 5.8% greater in the C4 than in the C2.

Growth medium formulas significantly influenced the shoot, root, and total biomass of seedlings. The least biomass production was observed on the F1 growth medium, but the seedling biomass was similar between the growing media of F2 and F3 (Figure 1A). Compared with the F1 growth medium, the shoot, root, and total biomass of *C. paliurus* seedlings grown on the F2 and F3 media increased by more than 33.0%, 29.1%, and 31.3%, respectively. The seedling biomass of *C. paliurus* was also significantly affected by the container types. The ranking of seedling biomass production by container size across the growth media was 760 mL (C1) > 500 mL (C2 and C4) > 280 mL (C3) (Figure 1B). Compared with the seedling biomass grown in the C3 container, the shoot, root and total biomass of *C. paliurus* seedlings grown in the C1 were 296.8%, 247.7%, and 274.9% higher, respectively, while only 109.8%, 89.5%, and 100.8% greater in the C2 and C4 containers. However, no significant difference in seedling biomass was detected between the same size containers with various textures (C2 and C4) (Figure 1B).

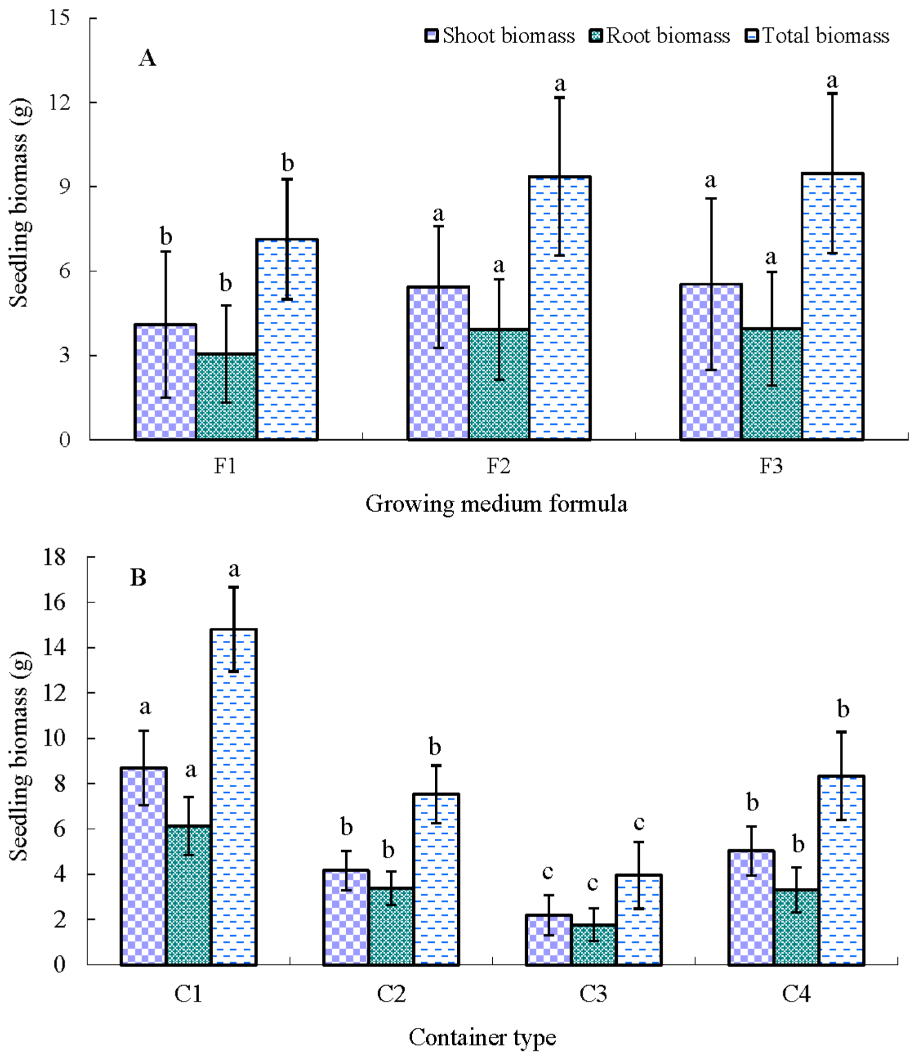

Figure 1. Effects of growth medium (**A**) and container type (**B**) on the biomass production of *Cyclocarya paliurus* seedlings sampled across the container types or growth media (means ± standard deviation). The means of the same biomass component followed by different letters are statistically different for container type or growth medium ($p < 0.05$, LSD test).

Table 2. *F*-values and probability levels from the GLM (general linear model) analysis of growth and biomass production of *Cyclocarya paliurus* seedlings.

Factors	*df*	Seedling Growth			Seedling Biomass			
		Height (cm)	Root-Collar Diameter (mm)	Ratio of Height to Diameter	Shoot (g)	Root (g)	Total (g)	Root-to-Shoot Ratio
Growth Medium (*F*)	2	19.37 ***	29.61 ***	17.35 ***	8.89 **	3.71 *	12.77 ***	0.62 ns
Container type (*C*)	3	333.94 ***	176.89 ***	158.44 ***	75.75 ***	34.16 ***	111.68 ***	2.62 ns
F × *C*	6	6.22 ***	2.86 **	10.02 ***	1.38 ns	0.57 ns	0.69 ns	3.65 *

ns: non-significant. *, ** and *** indicate significant effects at $p < 0.05$, $p < 0.01$, and $p < 0.001$, respectively.

Table 3. Effects of container type and growth medium on height growth, root-collar diameter, and the ratio of height to diameter of *Cyclocarya paliurus* container seedlings.

Container Type	Growing Medium	Seedling Height (cm)	Root-Collar Diameter (mm)	Ratio of Height to Diameter
C1	F1	63.92 ± 1.01 a	7.89 ± 0.17 ab	81.16 ± 1.27 a
C2	F1	43.44 ± 3.34 d	6.77 ± 0.28 e	64.88 ± 4.00 b
C3	F1	21.32 ± 2.64 h	5.23 ± 0.06 h	38.99 ± 1.71 f
C4	F1	36.25 ± 0.57 f	6.80 ± 0.33 de	54.23 ± 4.00 c
C1	F2	63.90 ± 3.51 a	8.16 ± 0.43 a	76.91 ± 4.83 a
C2	F2	45.16 ± 4.38 cd	7.18 ± 0.17 cd	64.51 ± 5.51 b
C3	F2	28.15 ± 0.11 g	6.17 ± 0.23 f	44.67 ± 0.16 e
C4	F2	48.80 ± 1.46 c	7.60 ± 0.15 b	64.52 ± 2.09 b
C1	F3	57.46 ± 2.99 b	8.22 ± 0.17 a	69.00 ± 5.08 b
C2	F3	38.67 ± 2.08 ef	7.54 ± 0.06 bc	50.11 ± 0.62 cd
C3	F3	25.77 ± 1.43 g	5.71 ± 0.12 g	46.45 ± 3.04 de
C4	F3	41.94 ± 2.05 de	7.71 ± 0.24 b	55.04 ± 0.01 c
Average of the treatments		42.9	7.08	59.21

Within the same column, the means followed by different letters are statistically different ($p < 0.05$, LSD test).

Owing to the interaction between growth medium and container type, we only evaluated the integrated effect of treatments on the root-to-shoot biomass ratio of the *C. paliurus* seedlings by one-way ANOVA, and the result was presented in Figure 2. The root-to-shoot biomass ratios ranged from 0.59 to 1.07, and followed the order of C2F3 ≥ C3F1 > C3F2 > C2F1 > C1F2 > C1F1 ≥ C1F3 > C4F1 > C4F3 > C4F2 > C2F2 > C3F3.

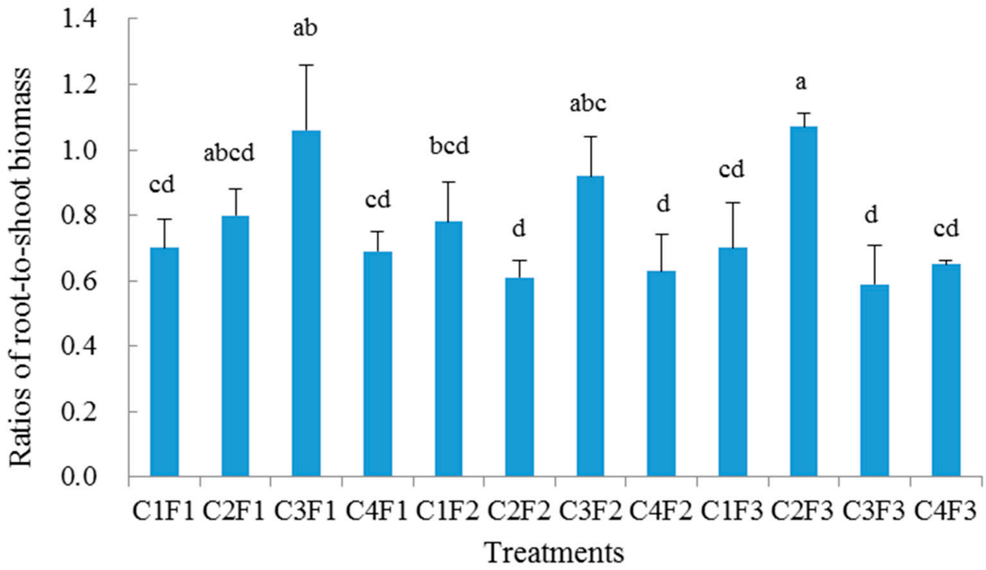

Figure 2. The integrated effect of treatments on the shoot-to-root biomass ratio of *Cyclocarya paliurus* seedlings (means ± standard deviation). The means followed by different letters are statistically different ($p < 0.05$, LSD test).

3.3. Seedling Root Morphology

Two-way analysis of variance showed that both growth media and container types significantly affected root morphological characteristics of the seedlings, except for mean root diameter, while no significant interaction between growth media and container types was observed for the root morphology (Table 4). Similarly to the seedling growth, it seems that container size exhibited larger effects on the total root length, root surface area, root volume, and mean root diameter of *C. paliurus* seedlings than the growth media (Table 4).

One-way ANOVA across the growth media indicated that container types significantly affected the root morphological characteristics, but there was no significant difference in the root morphology between the same size containers with various textures (C2 and C4) (Table 5). The seedlings grown in the large container had a more extended root system. Compared with the size of the 280 mL container (C3), the total root length grown in the 760 mL (C1) and 500 mL containers (C2 and C4) was increased by 214.7% and 90.4%, respectively, while the root surface area was about 300% and 200% greater. The root volume and mean root diameter grown in the C1, C2, and C4 containers were 68.7–247.0% and 45.7–77.0% greater than that of seedlings raised in C3 container. Although the root volume and the mean root diameter did not differ significantly between the C2 and C4 containers, it remained greater in the plastic containers (C4).

Table 4. *F*-values and probability levels from the GLM (general linear model) analysis of root morphology of *Cyclocarya paliurus* seedlings.

Factors	*df*	Root Morphology			
		Length of Total Root (m)	Root Surface Area (cm^2)	Root Volume (cm^3)	Mean Root Diameter (mm)
Growth Medium (*F*)	2	9.18 **	10.25 ***	4.20 *	2.37 ns
Container type (*C*)	3	19.02 ***	24.57 ***	12.78 **	3.90 *
$F \times C$	6	2.28 ns	2.37 ns	0.64 ns	1.05 ns

ns: non-significant. *, ** and *** indicate significant effects at $p < 0.05$, $p < 0.01$, and $p < 0.001$, respectively.

Table 5. The effect of container type and growth medium on root parameters of *Cyclocarya paliurus* container seedlings.

Treatment	Length of Total Root (m)	Root Surface Area (m^2)	Root Volume (cm^3)	Mean Root Diameter (mm)
		Container type		
C1	51.71 ± 19.39 a	0.08 ± 0.02 a	12.18 ± 4.33 a	3.61 ± 2.02 a
C2	31.45 ± 10.97 b	0.04 ± 0.01 b	5.92 ± 2.19 bc	3.01 ± 0.61 ab
C3	16.43 ± 8.19 c	0.02 ± 0.10 c	3.51 ± 1.38 c	2.04 ± 1.20 b
C4	31.11 ± 11.49 b	0.04 ± 0.01 b	7.59 ± 4.15 b	3.38 ± 0.55 a
		Growth medium		
F1	23.36 ± 8.74 b	0.03 ± 0.01 b	5.23 ± 2.96 b	2.83 ± 1.57 a
F2	40.65 ± 17.91 a	0.06 ± 0.03 a	8.01 ± 4.98 a	2.74 ± 0.76 a
F3	34.00 ± 21.41 a	0.05 ± 0.03 a	8.66 ± 4.82 a	3.49 ± 1.54 a

Within the same column the means followed by different letters are statistically different for container type or growth medium ($p < 0.05$, LSD test).

Except for mean root diameter, the other root morphological characteristics of the seedlings were significantly influenced by growth media (Table 5). The LSD test across the container types showed that the root morphological characteristics of the seedlings grown in the F2 and F3 media did not vary significantly (Table 5). However, the largest total root length and root surface area were observed on the F2 growth medium, whereas the greatest root volume and mean root diameter were found on the F3 growth medium. Meanwhile, the seedlings grown on the F1 growth medium had the smallest root system; also, the total root length, the root surface area and the root volume decreased by 31.3%, 40.0%, and 34.7% or more respectively, compared with those raised on F2 and F3 growth media.

4. Discussion

4.1. Effects of Container Type and Growth Medium on Seedling Quality

Seedling quality reflects the integration of a multitude of physiological and morphological characteristics of the seedling. Our results showed that both container type and growth medium significantly affected the growth, biomass, and root morphological characteristics of *C. paliurus* seedlings, except for mean root diameter (Tables 2 and 4). If we defined the seedlings with a height above 45 cm and root-collar diameter of more than 7.0 mm as qualified seedlings, seedlings from the combined treatments of C1F1, C1F2, C2F2, C4F2, and C1F3 satisfied the criteria, and the height-to-diameter ratios ranged from 64.5 to 81.2. However, no seedlings from the small size container (C3) reached the defined seedling quality. Results from the present study also showed that the root-to-shoot biomass ratio of the seedling was not significantly impacted by the container type and growth medium (Table 2), which suggests a similar phenotypic response to culturing practices during the nursery stage, in agreement with the result reported by Aghai et al. [11] for western larch. However, a higher root-to-shoot biomass ratio is preferred when seedling size is similar, and a higher total biomass with a similar root-to-shoot biomass ratio is favored. Haase [32] indicated that quality container seedlings should have a shoot-to-root ratio of 2:1 or less, and the values from this study were within that range (Figure 2). At the physiological level, a higher root-to-shoot biomass ratio may result in more favorable water relations, lower shoot maintenance requirements, and thus faster growth rates, while a higher total biomass may result in larger total carbohydrate stores available for remobilization and rapid growth soon after planting [33].

The obviously positive effect of increasing container size on seedling growth was reported for many woody plant species. For example, it was observed that seedling height, root-collar diameter, leaf production, and biomass increased with an increase of container size for *Elaeis guineensis* [7], *Acacia koa* [10], *Eucalyptus citriodora* [34], and *Pinus pinea* [35], which is in agreement with our results for the *C. paliurus*. Given the observed greater effects of container size on *C. paliurus* seedling growth and root morphology than those provided by growth medium formulas (Tables 2 and 4), our findings suggest that container size effects cannot be mediated by selecting appropriate growth media, and container size is a very important consideration during the tree seedling production in nurseries. For instance, the seedling height and root-collar diameter grown in the large container size with less nutrient medium (C1F1) were more than 127.1% and 27.9% larger than these grown in the small container size with more abundant nutrient media (C3F3 and C3F2), respectively. Our results also confirmed the results reported by Akpo et al. [7], where they concluded that fertilizer addition or substrate selection cannot overrule container size effects for the container seedlings of oil palm.

Owing to the differences in physical and chemical properties of the growing media (Table 1), different growth media also obviously influenced the seedling quality of *C. paliurus*, even if the observed effects on seedling growth, biomass, and root morphological characteristics were not as larger as the container size, especially as the container size decreased. For example, in the small container size (C3), the seedling height and root-collar diameter grown on the F1 growth medium were 20.9–30.0% and 9.2–18.0% less than those on the F2 and F3 growth media, respectively (Table 3). A possible explanation for this growth difference is the nutrient shortage on the F1 growth medium.

It is also worth mentioning that in the most cases, seedling growth, biomass, and root morphological characteristics grown in the black plastic container (C4) with the same growth medium were slightly larger than these grown in the non-woven container. The possible reason is that the non-woven container is permeable and allows water and soluble nutrients to move laterally, which could affect the water and nutrient availability for each seedling and thus impact the seedling growth. Moreover, the possible other reason is that the black plastic containers may absorb more solar radiation, which can increase the root temperature during the early spring.

4.2. Correlation among the Seedling Morphological Characteristics

It was reported that well-developed and well-structured root systems with numerous first order laterals are one of the most essential attributes of broad-leaved tree species [13]. However, measurements of seedling height and root-collar diameter are operationally used in the nursery practices to determine seedling quality, and can be used to project seedling potential following outplanting [36]. Our results indicated that both seedling height and root-collar diameter were highly correlated, with the different morphological variables of the one-year old seedlings ($p < 0.01$, Table 6), except for the ratio of root-to-shoot biomass. The highest correlation between variables was observed for shoot biomass and total biomass ($R = 0.978$), followed by root biomass and total biomass ($R = 0.955$). In most cases, there were no significant correlations between the ratio of root-to-shoot biomass and other morphological variables, except for a negatively significant correlation with shoot biomass ($R = -0.430$, Table 6).

The relations between seedling biomass, height, and root-collar diameter variables were curvilinear (Figure 3), but the correlation of total biomass was higher than the shoot biomass in regards to seedling root-collar diameter and height. Moreover, our study showed that a more closed relationship was found between root-collar diameter and seedling biomass (Figure 3), which is not in agreement with the result reported by Akpo et al. [7] for container seedlings of oil palm, where the highest correlation of seedling biomass was observed with seedling height.

Furthermore, both seedling root-collar diameter and height were positively and significantly correlated with the biomass variables and root morphological variables ($p < 0.01$, Table 6). These observed high correlations have two implications for evaluating the seedling quality of *C. paliurus*. First, it is unnecessary to consider all the seedling essential attributes to evaluate the seedling quality. The nursery manager or researcher would come to the similar conclusion by selecting seedling root-collar diameter and height, or one of these two allometric variables. Second, instead of measuring the total biomass of *C. paliurus* seedlings, the researcher could consider only the aboveground biomass, which is easier to collect and less time-consuming to process.

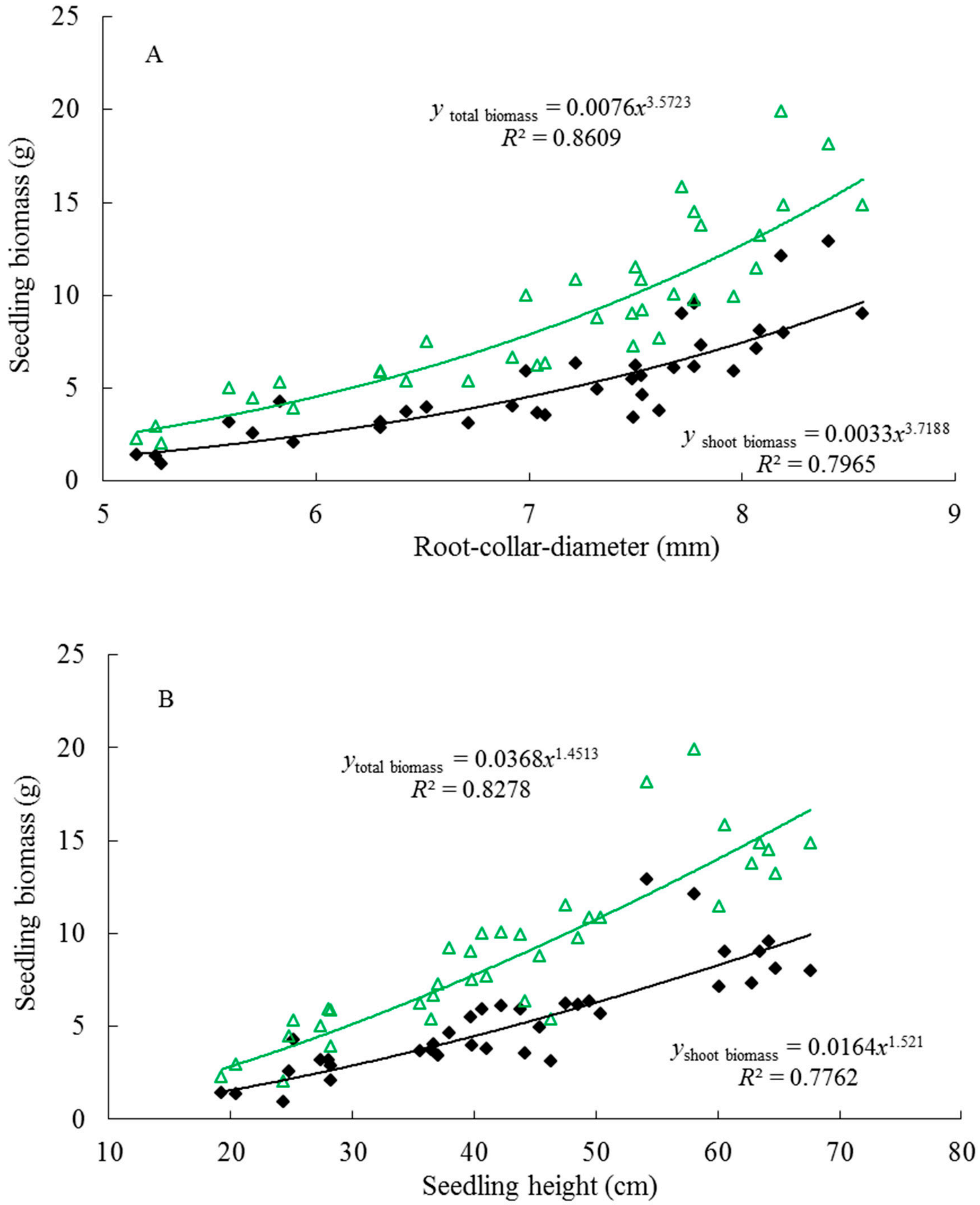

Figure 3. The relationship of seedling biomass with root-collar diameter (**A**) and height (**B**) of *Cyclocarya paliurus* seedlings (n = 36).

Table 6. Correlation matrix of seedling morphological variables measured (Pearson coefficients, two-tailed, *n* = 36).

Variables	Height	RCD	HDR	Shoot Biomass	Root Biomass	Total Biomass	RSR	TRL	RSA	RV
RCD	0.886 **									
HDR	0.925 **	0.765 **								
Shoot biomass	0.841 **	0.826 **	0.803 **							
Root biomass	0.875 **	0.860 **	0.808 **	0.872 **						
Total biomass	0.885 **	0.869 **	0.835 **	0.978 **	0.955 **					
RSR	−0.227	−0.198	−0.299	−0.430 **	−0.023	−0.278				
TRL	0.721 **	0.726 **	0.593 **	0.712 **	0.760 **	0.754 **	−0.076			
RSA	0.719 **	0.720 **	0.620 **	0.742 **	0.797 **	0.788 **	−0.057	0.936 **		
RV	0.686 **	0.693 **	0.587 **	0.670 **	0.722 **	0.714 **	−0.074	0.724 **	0.875 **	
MRD	0.450 **	0.498 **	0.308	0.362 *	0.345 *	0.369 *	−0.171	0.421 *	0.410 *	0.575 **

** and * Indicate significant correlation between variables at $p < 0.01$ and $p < 0.05$, respectively. RCD: root-collar diameter; HDR: Height-to-RCD ratio; RSR: root-to-shoot biomass ratio; TRL: total root length; RSA: root surface area; RV: root volume; MRD: mean root diameter.

4.3. Potential Option for the Practical Implications

Field performance of the plants and reforestation success were determined by seedling quality and the environmental conditions at the planting site. At sites where bareroot stock or natural or direct seeding is not expected to do well, foresters and managers considered container seedlings [33]. However, both the forester and nursery manager would like to develop a procedure of container seedling production with the least amount of environmental control necessary to produce the required seedling characteristics at the lowest production costs. Our results indicated that both container type and growth medium formula obviously affected the seedling size and root morphology of *C. paliurus*, which suggests that selections of container type and growth medium are very important for the quality seedling production of *C. paliurus*. Puértolas et al. [6] reported a threshold existed in the relative importance of seedling size on survival, but many studies indicated that the seedlings produced in larger volume containers can outperform those raised in smaller volume containers after planting [37,38], though seedlings raised in smaller volume containers are less costly to produce per unit. However, Close et al. [1] reported no observed effect of container type on *Eucalyptus globulus* growth four years after planting, despite the significant early effects of container type, which indicates that container type does not affect growth in the longer term.

The choice of seedling size and container type and their effects on post-planting performance are of economic importance to production forestry [1,8]. In view of the morphological indexes and biomass accumulation in the container seedlings of *C. paliurus*, the non-woven container C1 (760 mL) performed the best for the container seedlings, followed by containers C2 and C4 (500 mL). The seedlings of *C. paliurus* produced in the three container types (C1, C2, and C4), were healthy, and all of them approximately reached the appropriate dimensions for planting defined before. However, no seedlings from the small size container (C3, 280 mL) reached the defined seedling height and root-collar diameter (Table 3), which relates to difficulties competing with weeds after planting in sub-tropical areas. The use of the 760 mL non-woven container can produce a large size of *C. paliurus* seedlings, but nursery managers may consider that to be too large and require too much growth medium and labor to fill. Although the containers of C2 and C4 had a similar effect on the growth and quality of *C. paliurus* seedlings, it may be important that the seedlings produced in the non-woven container (C2) were planted with the containers and the container is degradable. Consequently, the seedlings had their roots protected not only during the planting work, but also throughout the whole first year after outplanting, until the roots increased and penetrated the soil. From the view of the economic, environmental, and transportation factors, the C2 container (500 mL, non-woven container) would be the most ideal choice for producing one-year-old *C. paliurus* seedlings in the practice.

Recently, more local growth medium components are preferred by seedling growers in order to reduce the increased transportation costs of growing media [7,14,19]. For instance, it is recommended that nursery managers use forest soil or household waste as substrate [7]. Our results showed that the seedlings grown on the growth media of F2 and F3 were superior to the seedlings raised on the F1 growth medium when the same container types were used. Both the yellow subsoil and fermented chicken manure used in this study are cheaper than peat, and transported at low costs, because the two substrates are easy to obtain locally. Although the effects of the F2 and F3 growth media on the seedling quality of *C. paliurus* were similar (Tables 3 and 5, and Figure 1), the F2 growth medium is more expensive than the F3 medium, because more peat was used in the F2 medium. Therefore, the F3 growth medium (yellow subsoil: perlite: fermented chicken manure: peat by volume = 2:2:3:3) would be recommended to put into practice in the future, even if the F2 growth medium would be also suitable for growing *C. paliurus* container seedlings and can be an alternative.

5. Conclusions

Both container type and growth medium significantly affected the growth, biomass, and root morphological characteristics of *C. paliurus* seedlings, but the selection of growth media cannot overrule container size effects for the container seedlings. The root-collar diameter and height of the seedlings were positively and significantly correlated with the biomass variables and root morphological variables, and can be used as essential attributes to evaluate the seedling quality. From the view of resource, environment, and cost benefits, the used management regime for *C. paliurus* seedling production is suggested to ensure good quality seedling delivery. The recommendations include the use of a non-woven container with 500 mL (C2), the selection of more local growth medium (such as F3), and producing one-year-old *C. paliurus* seedlings at the growing density of 100 container seedlings m^{-2} with air root pruning. Based on such a procedure, the nursery managers can optimize their *C. paliurus* seedling production more efficiently.

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