

Article

Leaf Angle as a Criterion for Optimizing Irrigation in Forest Nurseries: Impacts on Physiological Seedling Quality and Performance after Planting in Pots

Richardson Barbosa Gomes da Silva ^{1,*} , Danilo Simões ¹ , Ivar Wendling ² , Débora Zanoni do Prado ³, Maria Márcia Pereira Sartori ¹ , Angelo Albano da Silva Bertholdi ¹ and Magali Ribeiro da Silva ¹ 

¹ School of Agriculture, São Paulo State University (Unesp), Botucatu 18610-034, Brazil; danilo.simoes@unesp.br (D.S.); maria.mp.sartori@unesp.br (M.M.P.S.); angelo.bertholdi@unesp.br (A.A.d.S.B.); magali.ribeiro@unesp.br (M.R.d.S.)

² Brazilian Agricultural Research Corporation (Embrapa Forestry), Colombo 83411-000, Brazil; ivar.wendling@embrapa.br

³ Institute of Biosciences, São Paulo State University (Unesp), Botucatu 18618-689, Brazil; deborazp@gmail.com

* Correspondence: richardson.silva@unesp.br

Abstract: Seedling species with different architectures, e.g., mean leaf angles, are often subjected to the same irrigation management in forest nurseries, resulting in wasted water and fertilizer and reduced seedling quality. We aimed to evaluate whether irrigation volumes applied to tree seedling species with different leaf angles affect the physiological quality in forest nurseries and, consequently, performance after potting. We submitted nine seedling species with different mean leaf angles to four daily water regimes (8, 10, 12, and 14 mm). In the nursery, the following physiological attributes were considered to assess seedling quality: leaf water potential, daily transpiration rate, SPAD value, chlorophyll *a* and *b*, anthocyanins, carotenoids, and total nutrient content. After potting, we evaluated height and stem diameter over 120 days. Leaf angle can be used as a criterion for optimizing irrigation in forest nurseries, avoiding water and fertilizer wastage, and increasing physiological seedling quality. Leaf angle measurements combined with concurrent assessments of leaf traits are helpful in further understanding the effects of leaf angle variation and water regime on seedling quality. For positive leaf angles, an irrigation volume of 8 mm is sufficient to increase physiological seedling quality. Conversely, seedlings with negative leaf angles show the opposite response, requiring the largest irrigation volume (14 mm) to increase physiological seedling quality, except when the mean leaf area is small and concentrated in the upper half of the stem, which facilitates the access of irrigation water to the substrate and thus satisfies seedling water requirements. For all species, up to 120 days after planting in pots, the effect of the irrigation volume that provides greater growth and physiological quality at the end of the nursery phase is not overcome by other irrigation volumes applied.

Keywords: forest restoration; transpiration; leaf pigment content; leaf water potential; total nutrient content; overhead microsprinkler irrigation



Citation: da Silva, R.B.G.; Simões, D.; Wendling, I.; do Prado, D.Z.; Sartori, M.M.P.; Bertholdi, A.A.d.S.; da Silva, M.R. Leaf Angle as a Criterion for Optimizing Irrigation in Forest Nurseries: Impacts on Physiological Seedling Quality and Performance after Planting in Pots. *Forests* **2023**, *14*, 1042. <https://doi.org/10.3390/f14051042>

Academic Editor: Cate Macinnis-Ng

Received: 12 March 2023

Revised: 15 May 2023

Accepted: 16 May 2023

Published: 18 May 2023



Copyright: © 2023 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

1. Introduction

To combat the global warming crisis, the United Nations has declared 2021–2030 as the “International Decade on Ecosystem Restoration” to accelerate the restoration of degraded ecosystems, including tropical forests. As such, active management techniques, such as planting seedlings, play an important role in forest restoration. Seedling production requires specialized knowledge to produce morphologically and physiologically high-quality plants to meet forest restoration goals.

High quality seedlings are often a critical requirement for implementing forest and landscape restoration programs [1]. Nursery cultural practices directly affect the physiological functioning and subsequent morphological development of seedlings [2].

A broad seedling quality evaluation program provides useful information to support grower decisions and to understand the effects of management [3]. Successful establishment depends on the use of seedlings whose morphological and physiological characteristics achieved at the end of nursery production support growth and survival under an expected range of site conditions [4].

Physiological attributes commonly measured during seedling development include nutrient status and plant water status. The use of physiological quality at the end of nursery production as a criterion for selecting species for ecological restoration is advantageous. The resistance of species to the effects of seasonality results in reduced mortality and restoration costs [5].

In nurseries, the main factors that affect the quality of seedlings are the quality of genetic material, irrigation, nutrition, container type, and substrate composition. In irrigation, overhead microsprinklers are the most commonly used system for tree seedlings and tend to grow several species with different architectures under the same water management [6,7].

Species with larger crown canopies may largely contribute to leaf water interception. This canopy effect, which depends on canopy characteristics, container size, and spacing, is not considered by many growers. As a result, many growers typically apply more water than needed when scheduling irrigation, preferring the risk of increased leaching losses to the consequences of seedling water deficits [8,9].

Plant species can vary widely in architecture, i.e., the disposition of their components in space [10], such as the leaf angles. Leaf angle is an important plant structural trait that affects light interception, as well as carbon and water fluxes [11]. Steeper leaf angles can help reduce exposure to excess radiation and consequent water stress during the middle of the day, thereby increasing water use efficiency and hence carbon gain [12,13].

On the other hand, lower leaf angles increase light interception when the sun is at a high angle. The large interception of radiation by lower leaf angles can result in a significant increase in leaf temperature to levels above those optimal for photosynthesis. High leaf temperatures also increase the vapor pressure difference with the atmosphere, which increases transpiration rate and decreases water use efficiency [14,15].

The authors [16] measured and compiled an extensive dataset of leaf angles for 138 deciduous broadleaf species commonly found in temperate and boreal ecoclimatic regions. Similarly, [17] provide a dataset of leaf angles for 71 different *Eucalyptus* species native to Australia. More recently, data on leaf angle distributions for 50 widespread forest broadleaf tree species in Europe were also reported [18].

Rather than focusing on seedlings and their physiological attributes in response to water management, most studies of leaf angles have been concerned with the study of forests and their effects on radiation [19–25], remote sensing of the environment [26–29], leaf wettability and rainfall distribution in forests [30,31], and genotypic and phenotypic plasticity [32,33].

Overcoming this knowledge gap will help growers improve seedling quality and avoid wasting inputs such as water and fertilizer. We aimed to evaluate whether irrigation volumes applied to tree species with different leaf angles affect the physiological seedling quality in forest nurseries and, consequently, the performance after planting in pots.

2. Materials and Methods

2.1. Site Description

We conducted this study during the summer months in a sector forest nursery located in the São Paulo State, Brazil, at coordinates 22°51' S 48°26' W. The climate is classified as Cfa (Köppen classification) and the original phytophysionomies in this region are tropical semi-deciduous forest and Cerrado, which are considered biodiversity hotspots [34].

2.2. Leaf Angle Measurement and Experimental Treatments

We measured the mean leaf angle of nine tree seedling species from tropical semi-deciduous forests and the Cerrado, typically used for implementing active forest restoration programs: *Croton floribundus* (-56°), *Heliocarpus popayanensis* (-54°), *Guazuma ulmifolia* (-14°), *Esenbeckia leiocarpa* (31°), *Lafoensia pacari* (38°), *Moquiniastrium polymorphum* (42°), *Psidium cattleianum* (55°), *Magnolia ovata* (57°), and *Genipa americana* (58°) using a 180-degree transparent protractor on the youngest fully expanded pair of leaves. According to [7,13], we measured the leaf angle from the horizontal plane (0° ~flat; 90° ~upward leaf; -90° ~downward leaf). For each species, we considered the mean value of 30 seedlings between 25 and 35 cm tall.

We tested water regimes typically used for seedling production (8, 10, 12, and 14 mm/day $^{-1}$), divided twice a day, and applied by an overhead microsprinkler system. To accurately measure the amount of water applied by the irrigation system, we placed 36 plastic containers equidistantly under the microsprinklers in each covered bed prior to the irrigation pulse. The test duration was 1.0 h. Irrigation volumes were measured using a 1.0 L graduated cylinder.

Thus, the irrigation volumes were applied by the irrigation system at an average rate of 32.4 mm h $^{-1}$, with an operating pressure of 300 kPa. From this reference, we programmed the control panel of the irrigation system to apply the specified irrigation volume of each treatment.

The experiment was conducted using a split-plot design. The main plots were assigned to four irrigation volumes: 8, 10, 12, and 14 mm/day $^{-1}$. Subplots were assigned to nine tree seedling species: *Croton floribundus*, *Heliocarpus popayanensis*, *Guazuma ulmifolia*, *Esenbeckia leiocarpa*, *Lafoensia pacari*, *Moquiniastrium polymorphum*, *Psidium cattleianum*, *Magnolia ovata*, and *Genipa americana*. Four replicates were set up for each treatment, giving a total of 144 subplots. The experimental unit (each subplot) included a total of 20 seedlings.

2.3. Nursery Management

We filled polyethylene trays with peat moss-based substrates, carbonized rice hull, and perlite (2:1:1; by volume) and sowed in a shade house (50% shade), where the seedlings received 8 mm daily irrigation volume. After one month, we transferred the emerged seedlings to polyethylene containers (92 cm 3). The occupancy of each polyethylene tray was one polyethylene container per 86.8 cm 2 .

Subsequently, we standardized the subplots of each species, measuring their heights: *Croton floribundus* (5.0 cm $\pm$ 0.8), *Heliocarpus popayanensis* (11.7 cm $\pm$ 1.6), *Guazuma ulmifolia* (9.6 cm $\pm$ 1.0), *Esenbeckia leiocarpa* (7.9 cm $\pm$ 1.0), *Lafoensia pacari* (13.3 cm $\pm$ 1.3), *Moquiniastrium polymorphum* (4.8 cm $\pm$ 1.2), *Psidium cattleianum* (8.8 cm $\pm$ 1.2), *Magnolia ovata* (4.5 cm $\pm$ 0.5), and *Genipa americana* (3.4 cm $\pm$ 0.9).

Thus, we randomly assigned the trays in suspended and covered beds with diffuse-light film and programmed the overhead sprinkler system to apply the irrigation volumes corresponding to the treatments. The classification of Christiansen's uniformity coefficient was high (90%) [35], and that of the uniform distribution coefficient was good (86%) [36].

At the beginning of the irrigation treatments, we started seedling fertilization by applying 8 mm of nutrient solution per week. This volume was applied equally to all treatments and was deducted from the irrigation volume. The macronutrient solution was composed of Ca(NO $_3$) $_2$, urea (CH $_4$ N $_2$ O), MAP (NH $_4$ H $_2$ PO $_4$), KNO $_3$, and MgSO $_4$ at concentrations (milligrams per liter) of 295 (N), 84 (P), 200 (K), 38 (Mg), 160 (Ca), and 52 (S), and micronutrient solution of H $_3$ BO $_3$, Na $_2$ MoO $_4$, MnSO $_4$, ZnSO $_4$, CuSO $_4$, and FeSO $_4$ at concentrations (milligrams per liter) of 25 (Fe), 4.6 (B), 3.9 (Mn), 1.2 (Zn), 0.6 (Cu), and 0.3 (Mo). We also applied 8 mm per week of hardening fertilization solution composed of KCl at 700 mg L $^{-1}$ for one month.

The nursery period of each species was different due to their different growth rhythms: *Heliocarpus popayanensis*, *Lafoensia pacari*, *Moquiniastrium polymorphum*, and *Psidium cattleianum*

was 90 days; *Croton floribundus*, *Esenbeckia leiocarpa*, and *Magnolia ovata* was 150 days; and *Guazuma ulmifolia* and *Genipa americana* was 120 days.

To determine the effects of irrigation volume on seedling species with different leaf angles, we evaluated the physiological attributes at the end of the nursery phase. To determine the growth performance of the seedlings after potting, we evaluated the attributes' height and stem diameter over 120 days.

2.4. Physiological Analysis

2.4.1. Leaf Water Potential and Daily Transpiration Rate

For each mean leaf angle, we evaluated leaf water potential (Ψ_{leaf}) in four seedlings of each irrigation volume with a dew point potentiometer (model WP4-T, Washington, USA), collecting two fully expanded leaves at midday.

For each mean leaf angle, we evaluated the gravimetrically daily transpiration rate (milligram of water per square meter of leaf per second) [37] in sixteen seedlings of each irrigation volume. At 06:00 p.m., we completely saturated the substrate with water and then allowed it to drain. We attached a plastic bag to the polyethylene container and sealed it completely around the seedling with masking tape to prevent the evaporation of substrate water.

On a subsequent day (07:00 a.m.), we weighed the initial mass of the set (plastic bag, masking tape, and seedling) on a high-precision balance and kept it in full sunlight. After one day, we weighed the final mass of the set and then we detached leaves to determine the leaf area using a LI-3100 (Li-Cor, Lincoln, NE, USA). Thus, we calculated transpiration using Equation (1):

$$\frac{Im - Fm}{LA} \times \frac{1}{t} \quad (1)$$

where T is the daily transpiration rate, Im (mg) is the initial mass of set, Fm (mg) is the final mass of set, LA (m²) is the leaf area, and t is the time (s).

2.4.2. Leaf Pigment Contents Analysis and SPAD Value

For each mean leaf angle, we measured leaf pigment contents (chlorophyll *a* and *b*, anthocyanins, and carotenoids) in sixteen seedlings of each irrigation volume, collecting two fully expanded leaves, according to [38]. We homogenized leaves (0.100 g) in liquid nitrogen, and we performed all measurements in triplicate. We extracted pigments with 3 mL acetone/Tris pH 7.8 buffer solution (80/20, volume/volume). After the centrifugation of the extracts at 6000 rpm for five minutes, we read the absorbance at 663, 647, 537, and 470 nm. We calculated the pigment contents (mg g of fresh weight⁻¹) according to Equations (2)–(5), as follows:

$$Chl_a = 0.01373 \times A_{663} - 0.000897 \times A_{537} - 0.003046 \times A_{647} \quad (2)$$

$$Chl_b = 0.02405 \times A_{647} - 0.004305 \times A_{537} - 0.005507 \times A_{663} \quad (3)$$

$$Anthocyanins = 0.08173 \times A_{537} - 0.00697 \times A_{647} - 0.002228 \times A_{663} \quad (4)$$

$$Carotenoids = \frac{(A_{470} - (17.1 \times (Chl_a + Chl_b) - 9.479 \times Anthocyanins))}{119.26} \quad (5)$$

where Chl_a is the chlorophyll *a* content, Chl_b is the chlorophyll *b* content, and A is the sample absorbance at wavelength x in a 1 cm path length cuvette.

For each mean leaf angle, we considered the two fully expanded leaves and measured SPAD values in sixteen seedlings of each irrigation volume, using a chlorophyll concentration estimator (SPAD-502, Konica Minolta, Osaka, Japan).

2.4.3. Total Nutrient Content Analysis

For each mean leaf angle, we evaluated the total nutrient content in four seedlings of each irrigation volume, according to [39–41]. Seedlings were separated into roots and shoots and the substrate was washed from the roots. Both the shoot and root parts were dried in an air circulation oven at 70 °C until they reached a constant mass and then they were weighed for dry mass determination on a high-precision balance. The dried samples were separately ground in a Wiley mill (Arthur H. Thomas Company, Philadelphia, PA, USA) and used for determination of tissue macronutrient (g kg^{-1}) and micronutrient (mg kg^{-1}) concentrations for P, K, Mg, Ca, S, Zn, Fe, Cu, and Mn by nitroperchloric digestion, N by sulfuric perchloric digestion, and B by dry-ashing.

Tissue concentrations were multiplied by the oven-dried weight of the corresponding part to calculate nutrient content. Total macronutrient and micronutrient content were determined by summing the nutrient content of each seedling part.

2.5. Seedling Growth Performance after Planting in Pot

After finishing the nursery phase, we planted six seedling representatives of each species and irrigation volume in 7 L pots containing Oxisol medium texture with sand (3:1; by volume). In the soil, we added fertilizer with the N-P-K formulation (4:14:8) at a dosage of 2 kg m^{-3} soil. We randomized pots of seedlings in a covered greenhouse with diffuse-light film and irrigated them with 500 mL of water every five days. Immediately after planting and at 30, 60, 90, and 120 days after planting, we measured the growth characteristics: height (cm) and stem diameter (mm).

2.6. Data Analysis

The physiological data (leaf water potential, daily transpiration rate, SPAD value, chlorophyll *a* and *b*, anthocyanins, and carotenoids content) were submitted to the Shapiro–Wilk test [42] to verify the normality. Analysis of Variance (ANOVA) [43] was performed on these data ($p < 0.05$). The Scott–Knott test [44] was used for multiple comparisons ($p < 0.05$).

The relationship between each total nutrient content and irrigation volume at each leaf angle was evaluated using the Pearson correlation coefficient ($p < 0.05$ and $p < 0.01$).

To analyze the effect of the treatments applied during the nursery phase on seedling growth performance after potting, the ANOVA was performed on the growth data every 30 days ($p < 0.05$). Before the analysis, the normality of the data was verified with the Shapiro–Wilk test. The Scott–Knott test was used for multiple comparisons ($p < 0.05$). All statistical analyses were performed with the STATISTICA software package [45].

3. Results

3.1. Physiological Quality of Seedlings

In the ANOVA (Tables S1–S7 available in Supplementary Materials), the significant interaction between irrigation volume and mean leaf angle influenced the leaf water potential, daily transpiration rate, SPAD value, chlorophyll *a* and *b*, anthocyanins, and carotenoids content ($p < 0.05$).

Increasing irrigation volume did not significantly increase daily transpiration rate at 58°, 55°, 42°, and 38° leaf angles. For leaf angles of −54°, 31°, and 57°, the application of a 10 mm irrigation volume increased the daily transpiration rate, which was not significantly different from the larger irrigation volumes. On the other hand, for leaf angles of −14° and −56°, the largest irrigation volume (14 mm) was required to provide a greater daily transpiration rate (Figure 1).

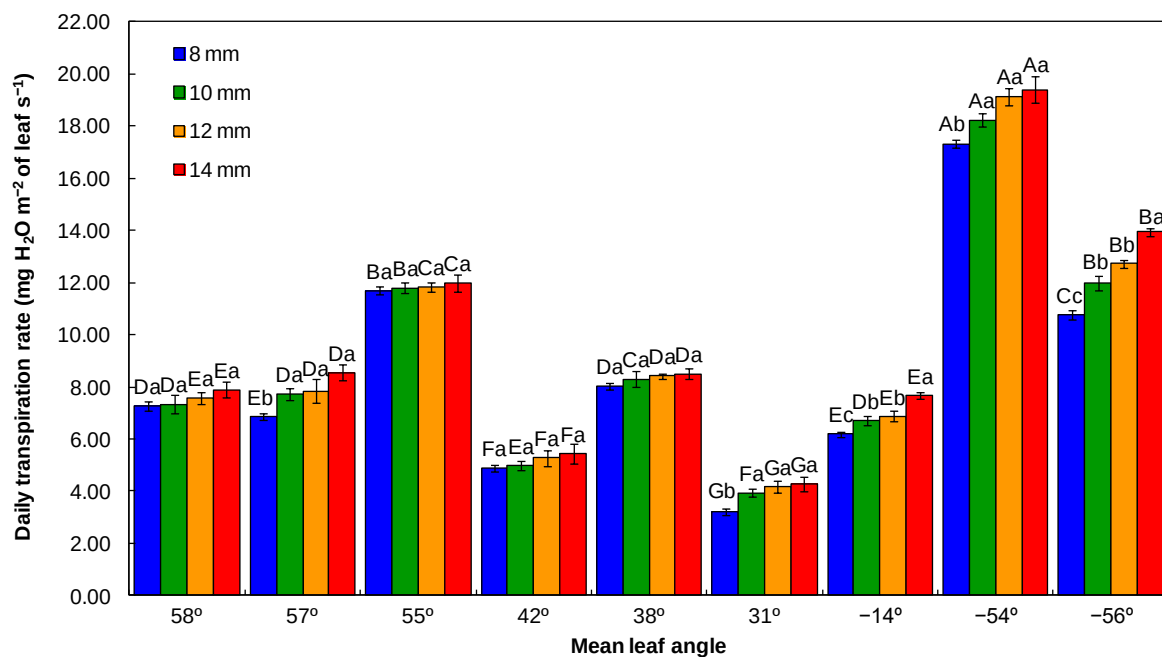


Figure 1. Daily transpiration rate of tree seedlings as influenced by mean leaf angles and irrigation volumes at end of the nursery phase. *Croton floribundus* (−56°), *Heliocarpus popayanensis* (−54°), *Guazuma ulmifolia* (−14°), *Esenbeckia leiocarpa* (31°), *Lafoensia pacari* (38°), *Moquiniastrum polymorphum* (42°), *Psidium cattleyanum* (55°), *Magnolia ovata* (57°), and *Genipa americana* (58°). Different capital letters on the bars indicate significant differences between mean leaf angles at the same irrigation volume by the Scott–Knott test at 5% probability. Different small letters on the bars indicate significant differences between irrigation volumes for the same mean leaf angle by the Scott–Knott test at 5% probability. Error bars denote the standard error.

In addition, species with negative leaf angles showed less negative leaf water potential at 14 mm irrigation volume, except for *Heliocarpus popayanensis* (−54°), for which this attribute did not significantly differ between irrigation volumes (Figure 2). It is important to highlight that *Heliocarpus popayanensis* had the smallest mean leaf area of the species tested (45.73 cm²; Table S8 available in Supplementary Materials). For species with mean leaf angles of 58°, 57°, 55°, 42°, 38°, and 31°, increasing irrigation volume did not significantly change leaf water potential.

Pigment contents and SPAD values were reduced in varying proportions with increasing irrigation volume when the leaf angle of the species was positive and −54°, except for leaf angle 38° where irrigation volume did not significantly change chlorophyll *a* and *b* or anthocyanins content. For the leaf angles of −14° and −56°, the larger irrigation volumes were required to produce higher carotenoids, chlorophyll *a* and *b*, and anthocyanins content (Figures 3–6) and a higher SPAD value (Figure 7).

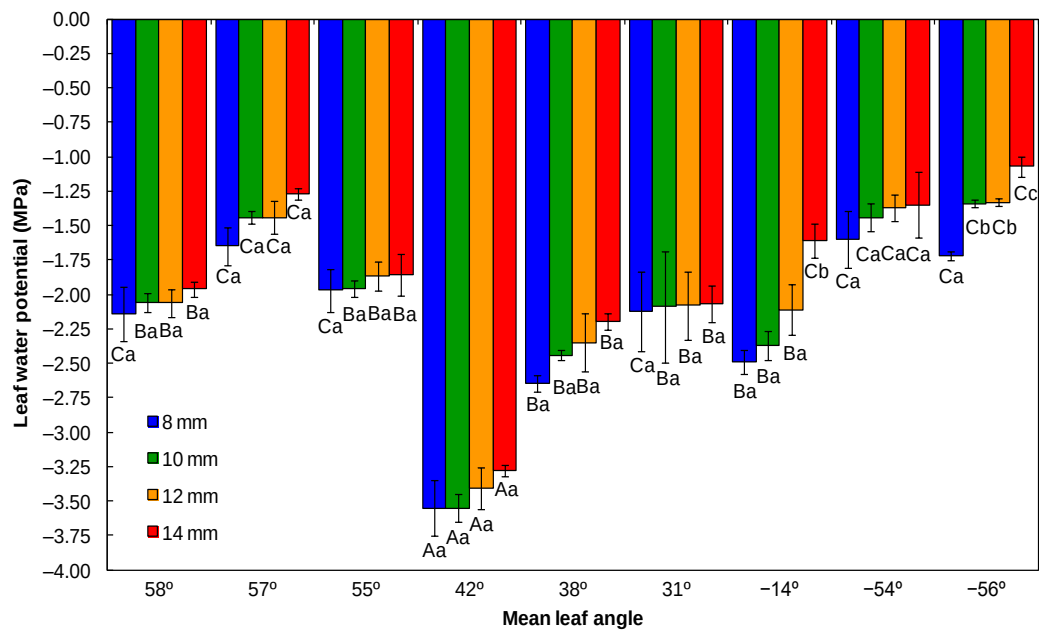


Figure 2. Leaf water potential as influenced by mean leaf angles and irrigation volumes at end of the nursery phase. *Croton floribundus* (-56°), *Heliocarpus popayanensis* (-54°), *Guazuma ulmifolia* (-14°), *Esenbeckia leiocarpa* (31°), *Lafoensia pacari* (38°), *Moquiniastrium polymorphum* (42°), *Psidium cattleyanum* (55°), *Magnolia ovata* (57°), and *Genipa americana* (58°). Different capital letters on the bars indicate significant differences between mean leaf angles at the same irrigation volume by the Scott–Knott test at 5% probability. Different small letters on the bars indicate significant differences between irrigation volumes for the same mean leaf angle by the Scott–Knott test at 5% probability. Error bars denote the standard error.

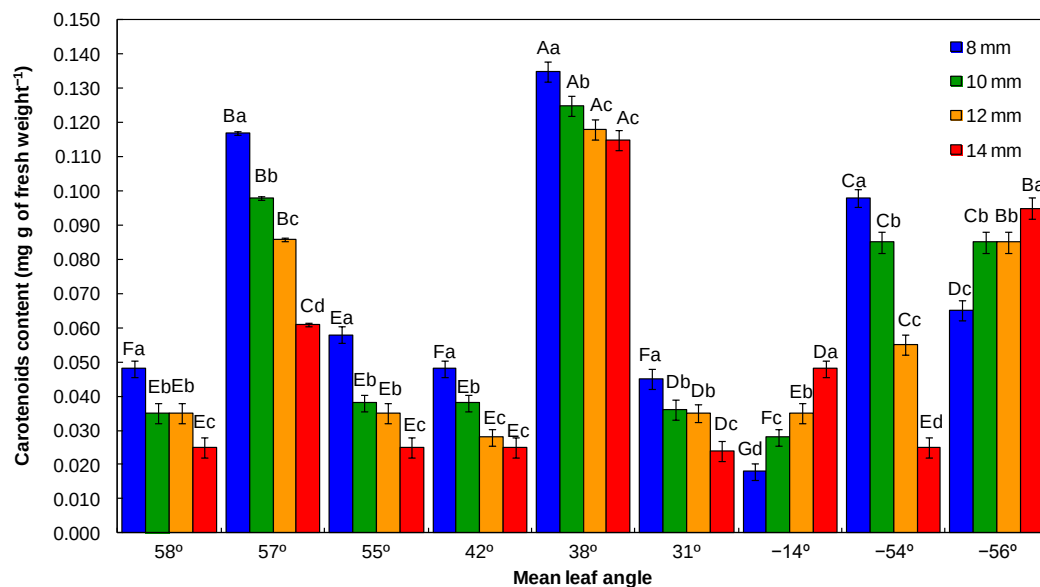


Figure 3. Carotenoids content of tree seedlings as influenced by mean leaf angles and irrigation volumes at the end of the nursery phase. *Croton floribundus* (-56°), *Heliocarpus popayanensis* (-54°), *Guazuma ulmifolia* (-14°), *Esenbeckia leiocarpa* (31°), *Lafoensia pacari* (38°), *Moquiniastrium polymorphum* (42°), *Psidium cattleyanum* (55°), *Magnolia ovata* (57°), and *Genipa americana* (58°). Different capital letters on the bars indicate significant differences between mean leaf angles at the same irrigation volume by the Scott–Knott test at 5% probability. Different small letters on the bars indicate significant differences between irrigation volumes for the same mean leaf angle by the Scott–Knott test at 5% probability. Error bars denote the standard error.

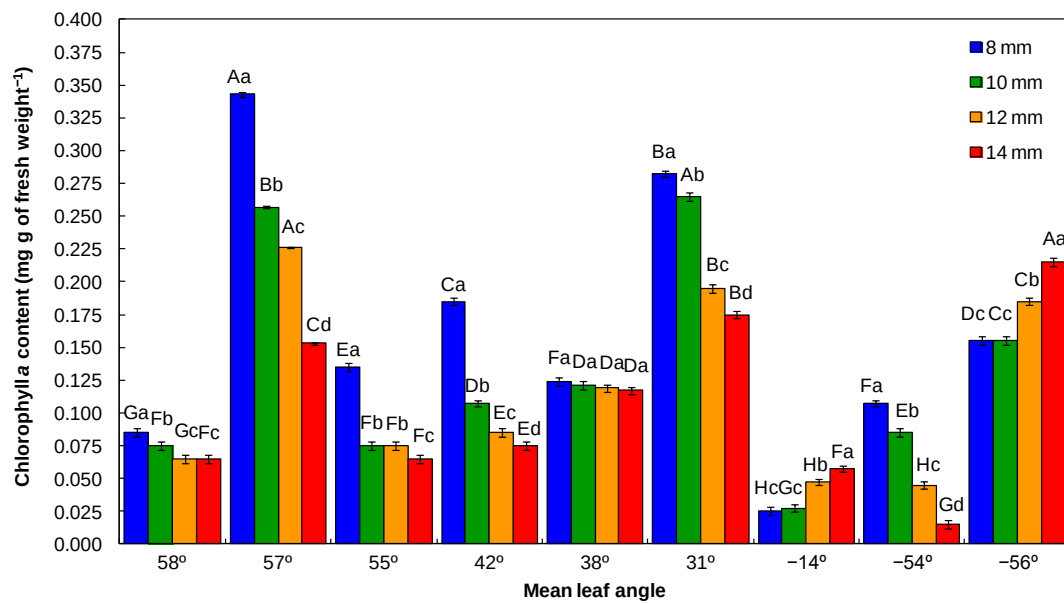


Figure 4. Chlorophyll *a* content of tree seedlings as influenced by mean leaf angles and irrigation volumes at the end of the nursery phase. *Croton floribundus* (−56°), *Heliocarpus popayanensis* (−54°), *Guazuma ulmifolia* (−14°), *Esenbeckia leiocarpa* (31°), *Lafoensia pacari* (38°), *Moquiniastrum polymorphum* (42°), *Psidium cattleianum* (55°), *Magnolia ovata* (57°), and *Genipa americana* (58°). Different capital letters on the bars indicate significant differences between mean leaf angles at the same irrigation volume by the Scott–Knott test at 5% probability. Different small letters on the bars indicate significant differences between irrigation volumes for the same mean leaf angle by the Scott–Knott test at 5% probability. Error bars denote the standard error.

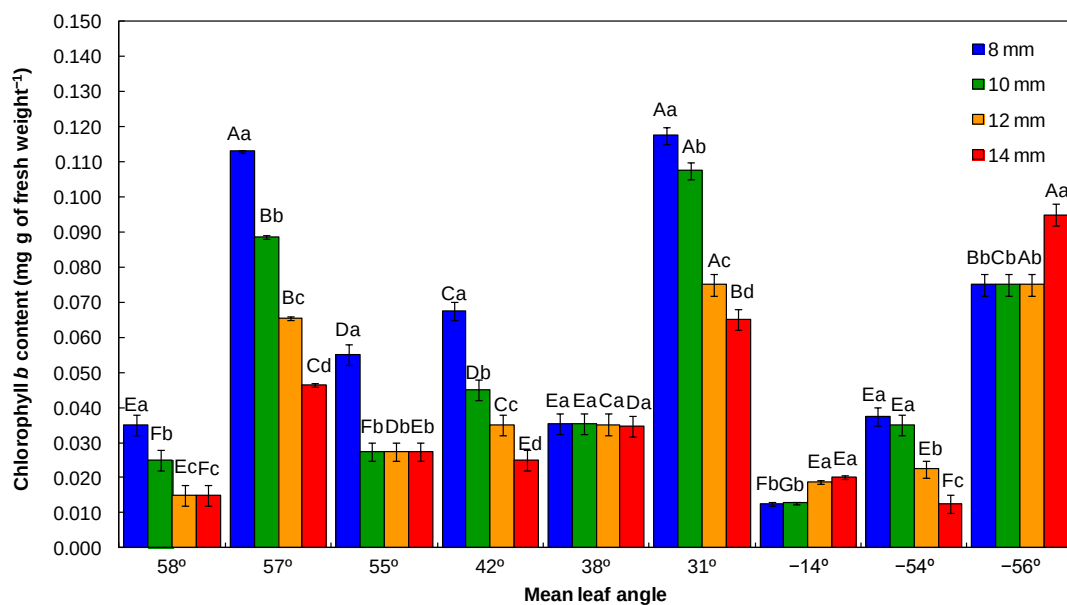


Figure 5. Chlorophyll *b* content of tree seedlings as influenced by mean leaf angles and irrigation volumes at the end of the nursery phase. *Croton floribundus* (−56°), *Heliocarpus popayanensis* (−54°), *Guazuma ulmifolia* (−14°), *Esenbeckia leiocarpa* (31°), *Lafoensia pacari* (38°), *Moquiniastrum polymorphum* (42°), *Psidium cattleianum* (55°), *Magnolia ovata* (57°), and *Genipa americana* (58°). Different capital letters on the bars indicate significant differences between mean leaf angles at the same irrigation volume by the Scott–Knott test at 5% probability. Different small letters on the bars indicate significant differences between irrigation volumes for the same mean leaf angle by the Scott–Knott test at 5% probability. Error bars denote the standard error.

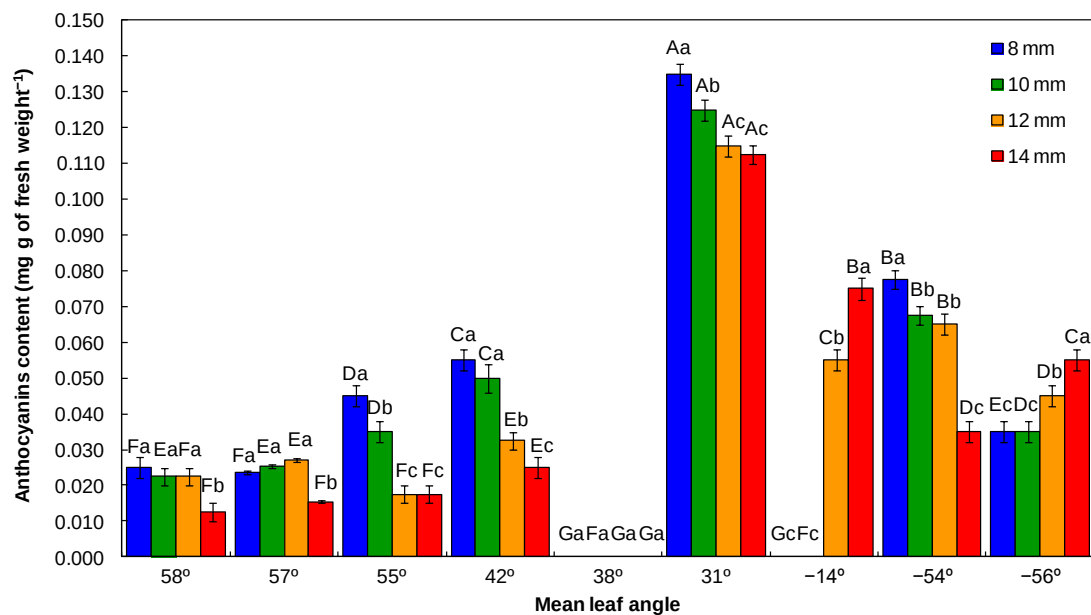


Figure 6. Anthocyanins content of tree seedlings as influenced by mean leaf angles and irrigation volumes at the end of the nursery phase. *Croton floribundus* (−56°), *Heliocarpus popayanensis* (−54°), *Guazuma ulmifolia* (−14°), *Esenbeckia leiocarpa* (31°), *Lafoensia pacari* (38°), *Moquiniastrium polymorphum* (42°), *Psidium cattleianum* (55°), *Magnolia ovata* (57°), and *Genipa americana* (58°). Different capital letters on the bars indicate significant differences between mean leaf angles at the same irrigation volume by the Scott–Knott test at 5% probability. Different small letters on the bars indicate significant differences between irrigation volumes for the same mean leaf angle by the Scott–Knott test at 5% probability. Error bars denote the standard error.

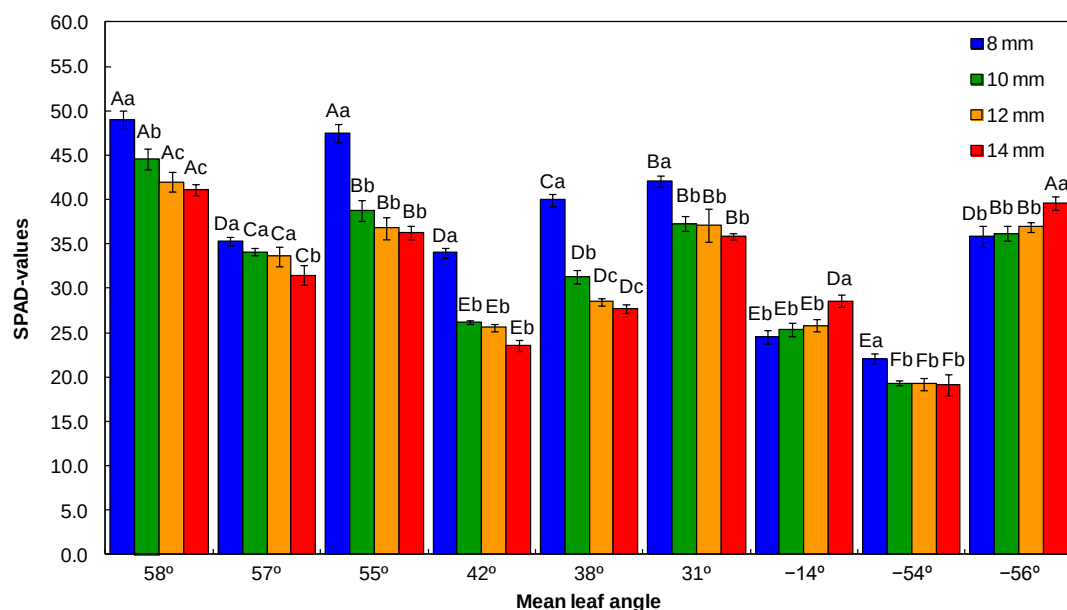


Figure 7. SPAD values of tree seedlings as influenced by mean leaf angles and irrigation volumes at the end of the nursery phase. *Croton floribundus* (−56°), *Heliocarpus popayanensis* (−54°), *Guazuma ulmifolia* (−14°), *Esenbeckia leiocarpa* (31°), *Lafoensia pacari* (38°), *Moquiniastrium polymorphum* (42°), *Psidium cattleianum* (55°), *Magnolia ovata* (57°), and *Genipa americana* (58°). Different capital letters on the bars indicate significant differences between mean leaf angles at the same irrigation volume by the Scott–Knott test at 5% probability. Different small letters on the bars indicate significant differences between irrigation volumes for the same mean leaf angle by the Scott–Knott test at 5% probability. Error bars denote the standard error.

For each mean leaf angle, the Pearson's correlation coefficient between each total nutrient content and irrigation volume was significant for most nutrients. When significant, for leaf angles of -14° and -56° , the correlation coefficient was positive, i.e., an increase in irrigation volume increased the total nutrient content. For leaf angles of 58° , 57° , 55° , 42° , 38° , 31° , and -54° , the correlation coefficient was negative, i.e., total nutrient content decreased as irrigation volume increased (Figure 8).

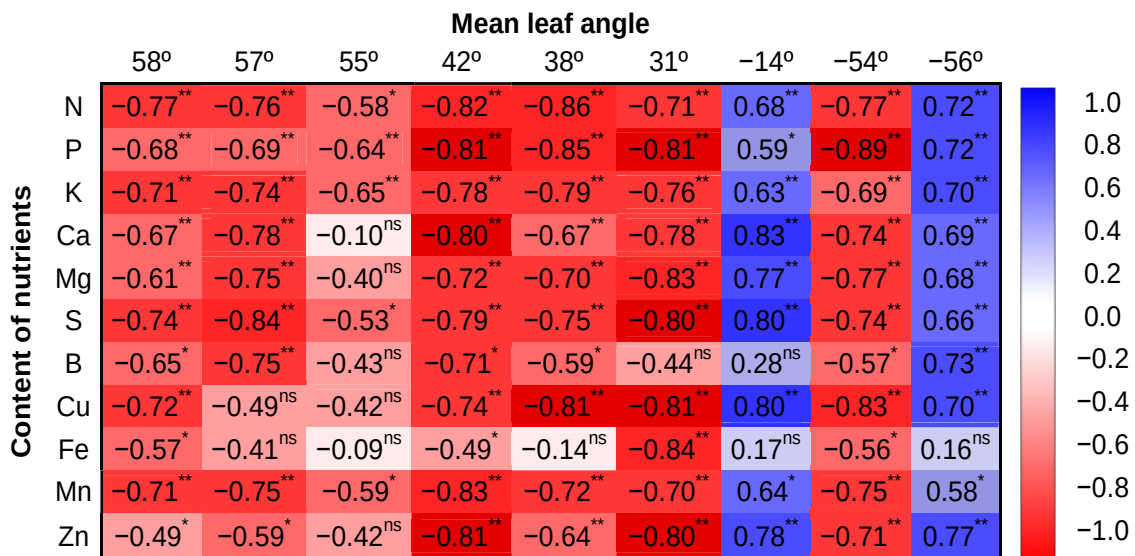


Figure 8. Pearson's correlation coefficient between irrigation volumes and content of nutrients in each mean leaf angle at the end of the nursery phase. *Croton floribundus* (-56°), *Heliocarpus popayanensis* (-54°), *Guazuma ulmifolia* (-14°), *Esenbeckia leiocarpa* (31°), *Lafoensia pacari* (38°), *Moquiniastrum polymorphum* (42°), *Psidium cattleianum* (55°), *Magnolia ovata* (57°), and *Genipa americana* (58°). ^{ns} = no significant correlation, * = significant correlation at 5% probability, and ** = significant correlation at 1% probability.

3.2. Seedling Growth Performance after Planting in Pot

The height and stem diameter of the species at the end of the nursery phase followed a pattern similar to that of the physiological attributes in response to the irrigation volumes applied, i.e., the 8 mm irrigation volume produced higher growth seedlings when the leaf angles were 58° , 57° , 55° , 42° , 38° , 31° , and -54° , while a 14 mm irrigation volume was necessary for the mean leaf angles of -14° and -56° .

After planting, the effect of the treatments applied during the nursery phase influenced the performance of the potted seedlings to different extents. Regarding height, the effect of irrigation volumes remained significant up to 120 days after planting at leaf angles of 57° and 58° ; 42° (90 days); 38° , 31° , and -14° (60 days); and -54° and -56° (30 days). The 55° mean leaf angle was the only one where irrigation volumes differed only at the time of planting (Figure 9).

For stem diameter, there was a difference between the irrigation volumes applied during the nursery phase until 120 days after planting for the species with an angle of 42° ; -14° and 57° (60 days); and -54° and 55° (30 days). For leaf angles of 58° and 31° , irrigation volumes did not differ between 30 and 90 days after planting; however, they differed again at 120 days. The species with angles of -56° and 38° were the only species in which the irrigation volumes applied during the nursery phase differed only at the time of planting (Figure 10).

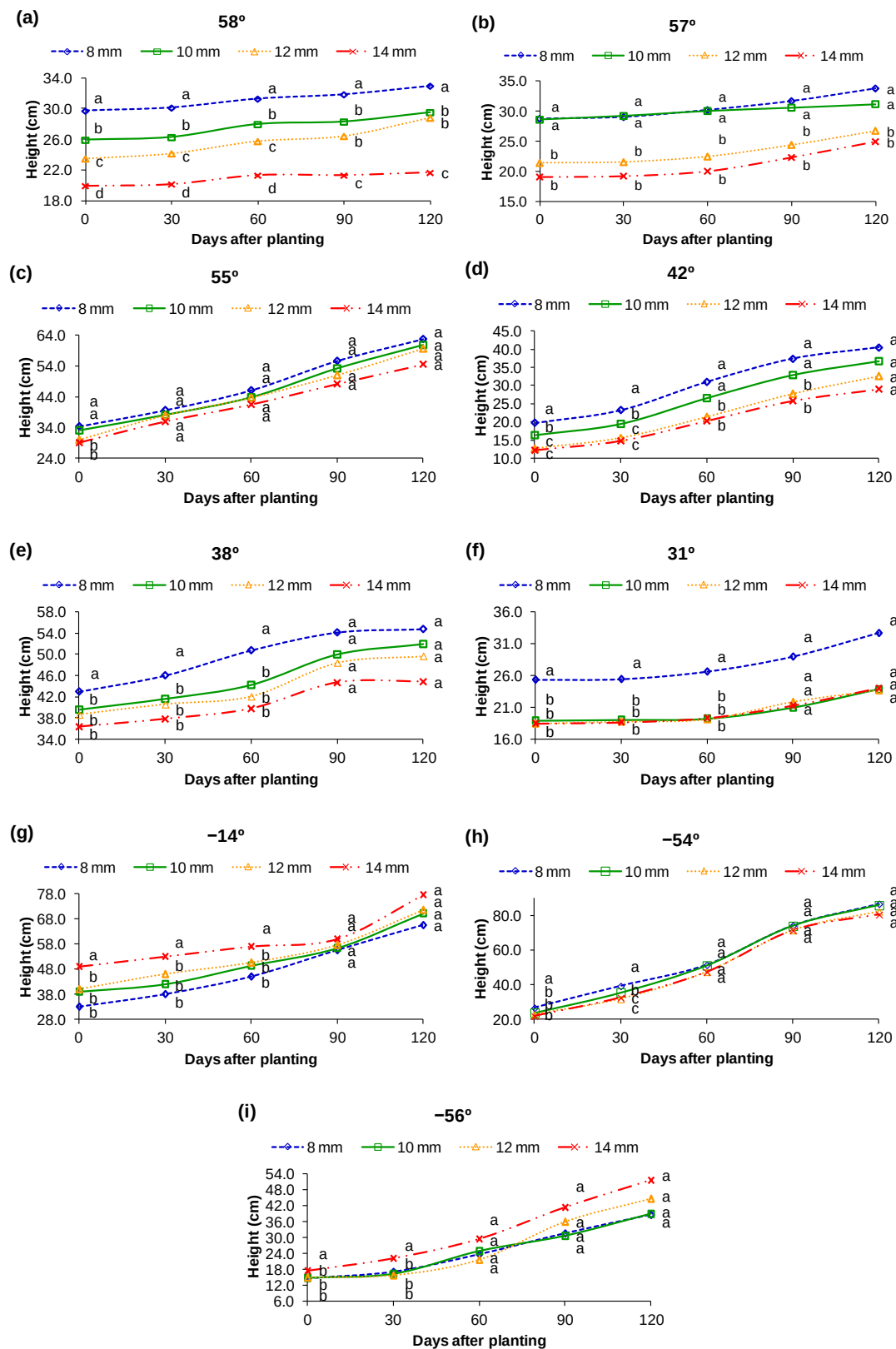


Figure 9. Height (cm) of tree seedlings until 120 days after planting in pots as influenced by treatments applied during the nursery phase. (a) *Genipa americana* (58°), (b) *Magnolia ovata* (57°), (c) *Psidium cattleianum* (55°), (d) *Moquiniastrum polymorphum* (42°), (e) *Lafoensia pacari* (38°), (f) *Esenbeckia leiocarpa* (31°), (g) *Guazuma ulmifolia* (−14°), (h) *Heliocarpus popayanensis* (−54°), and (i) *Croton floribundus* (−56°). Different letters on the lines indicate significant differences between irrigation volumes in the same month by the Scott–Knott test at 5% probability.

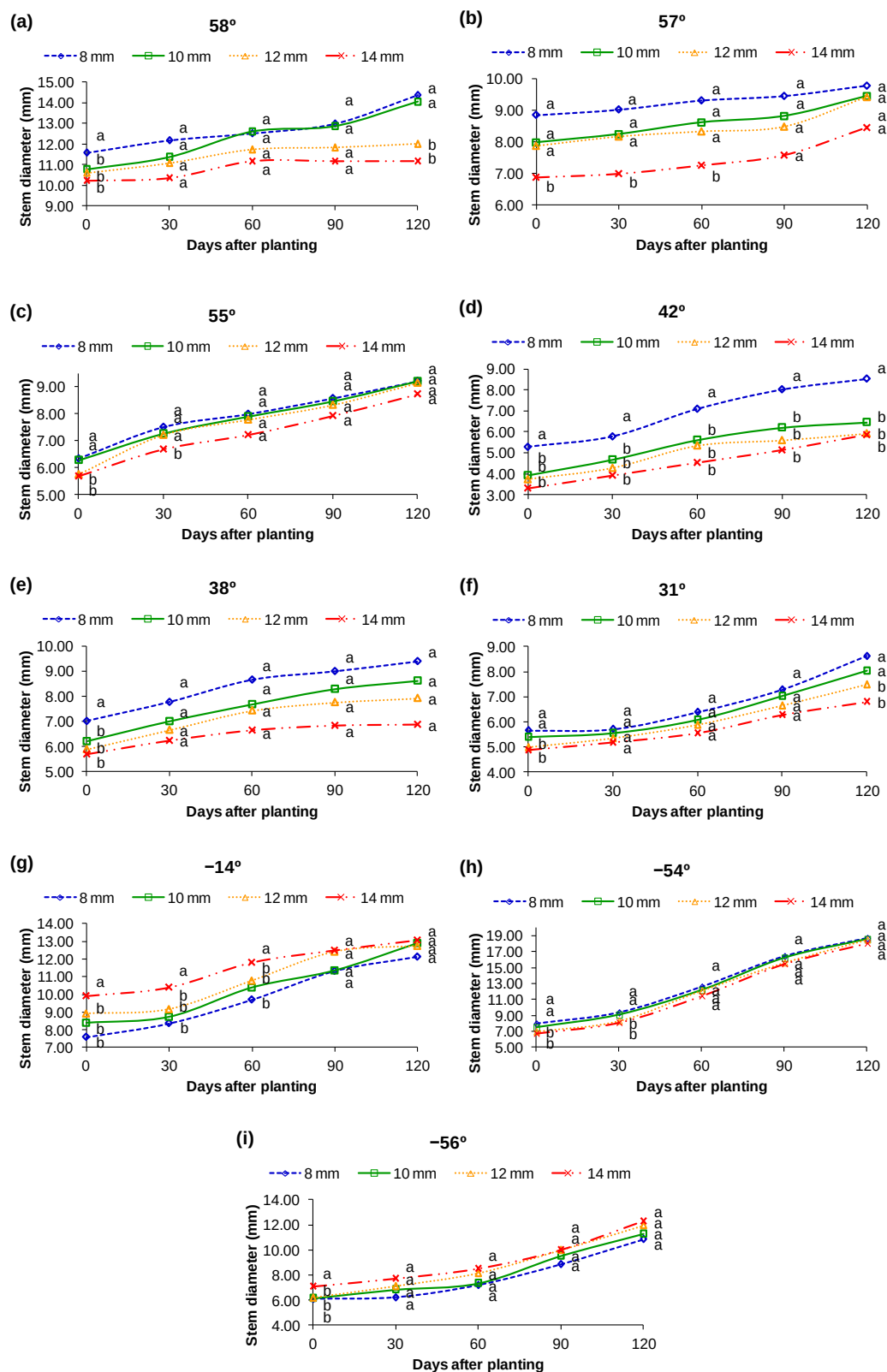


Figure 10. Stem diameter (mm) of tree seedlings until 120 days after planting in pots as influenced by treatments applied during the nursery phase. (a) *Genipa americana* (58°), (b) *Magnolia ovata* (57°), (c) *Psidium cattleianum* (55°), (d) *Moquiniastrium polymorphum* (42°), (e) *Lafoensia pacari* (38°), (f) *Esenbeckia leiocarpa* (31°), (g) *Guazuma ulmifolia* (−14°), (h) *Heliocarpus popayanensis* (−54°), and (i) *Croton floribundus* (−56°). Different letters on the lines indicate significant differences between irrigation volumes in the same month by the Scott-Knott test at 5% probability.

4. Discussion

4.1. Physiological Quality of Seedlings

Irrigation water capture and its direction to the substrate is hindered by seedling species with negative mean leaf angles (-14° and -56°), their leaf area, and the spatial distribution of leaves (extended their leaves downwards of the stem) (Figure 11a,b); therefore, to achieve less negative leaf water potential and to increase the daily transpiration rate, a 14 mm irrigation volume is required to compensate for the uncaptured water.

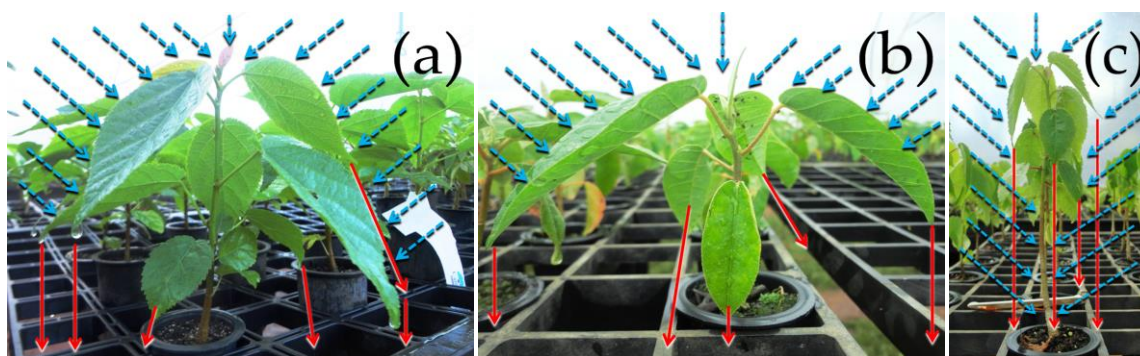


Figure 11. Irrigation water capture by seedling species with negative mean leaf angles. (a) Leaf angle, leaf area, and spatial distribution of leaves in *Guazuma ulmifolia* (-14°) and (b) *Croton floribundus* (-56°) hinder the access of irrigation water to the substrate. (c) Leaf angle, small leaf area, and spatial distribution of leaves in *Heliocarpus popayanensis* (-54°) facilitate the access of irrigation water to the substrate.

These results are in accordance with the study of [46], i.e., for species with negative mean leaf angles of -1° and -64° , the smallest irrigation volume (6 mm) reduced the leaf water potential, stomatal conductance, and daily transpiration rate. According to [47], the leaf water potential indicates whether the physiological status of seedlings has deteriorated. In their review, leaf water potential had a 100% positive correlation with growth, highlighting the importance of this attribute in assessing seedling quality. Furthermore, leaf water potential shows interactions between water supply and plant water demand. Even moderate stress, as caused by some of the treatments in this study, can cause stomatal closure, reducing transpiration rates and photosynthesis [48–50].

Seedlings of *Heliocarpus popayanensis* (-54°) showed different behavior from other negative leaf angles, i.e., it was not necessary to apply a 14 mm irrigation volume to provide less negative leaf water potential and increase the daily transpiration rate. This was due to the fact that this species had the smaller mean leaf area of all species tested here (45.73 cm^2 ; Table S8 available in Supplementary Materials) and concentrated their leaf area in the upper half of the stem (Figure 11c), which facilitated the access of irrigation water to the substrate and thus fully satisfied the water requirements of the seedlings with the smaller irrigation volumes. These results are in accordance with [51], who recommended that leaf angle measurements be combined with concurrent assessments of leaf traits (e.g., leaf area and spatial distribution of leaves). These combinations would be helpful in further understanding the effects of leaf angle variation and water regime on seedling quality.

For mean leaf angles of 57° and 31° , the daily transpiration rate increased as the irrigation volume increased from 8 mm to 10 mm, which did not differ significantly from the other irrigation volumes. For mean leaf angles of 58° , 55° , 42° , and 38° , the 8 mm irrigation volume was sufficient to maintain an adequate volume of water in the container, so that daily transpiration rate and leaf water potential were not significantly different from the larger irrigation volumes. Among these species, *Moquiniastrium polymorphum* (42°) had the higher midday leaf water potential values (ranging from -3.28 to -3.55 MPa). *Moquiniastrium polymorphum* is a common species in dry forests and Cerrado, occurring both

in the more open and in the more closed vegetation types of typical or forested savanna and riparian forests [52] with fire resistance [53]. Drought-tolerant species function at low plant water potentials by maintaining low osmotic potentials or by accumulating solutes in response to stress [54]. They have the ability to maintain turgor, growth, and gas exchange in very dry environments [55].

For mean leaf angles of -56° and -14° , greater irrigation volumes were necessary to achieve higher pigment contents and SPAD values. This indicates that the application of smaller irrigation volumes could not fully meet the water and nutrient requirements of these seedlings (Figure 11a,b). During the stress period, carotenoids are associated with several important physiological functions, including light harvesting and photoprotection [56,57]. In *Erythrina velutina* and *Poincianella pyramidalis* seedlings, the full recovery of photosynthesis after rewatering was associated with enhanced photoprotection by carotenoids, ensuring the resilience of these species in the face of periodic drought [58].

The effect of irrigation on chlorophyll and carotenoids content is controversial. In seedlings of *Picea abies* [59], *Pistacia lentiscus* [60], *Pachira aquatica* [61], *Prunus sargentii* [62], *Populus nigra* and *Quercus brantii* [63], drought stress caused a decrease in the concentrations of chlorophylls and carotenoids, which may have a protective role in protecting the photosynthetic apparatus against drought-induced damage. In seedlings of *Acacia arabica* [64] and *Larix kaempferi* [62], with higher water content, carotenoids and leaf chlorophyll were not affected. Thus, pigment response to irrigation and water stress may vary among species.

For seedlings with leaf angles of 58° , 57° , 55° , 42° , 38° , 31° , and -54° , the smallest irrigation volume (8 mm) was sufficient to produce higher pigment contents and SPAD values. The increase in irrigation volume at these leaf angles resulted in a loss of ions and probably affected the biosynthesis of chlorophyll, carotenoids, and anthocyanins. Mineral nutrition can affect the synthesis of these pigments. It also significantly affects the dynamics of leaf surface formation and its extent, which is reflected in the photosynthetic potential and net photosynthetic productivity. The only exception was the seedlings of *Lafoensia pacari* (38°), which did not produce any anthocyanins content in any of the irrigation volumes. This result is in accordance with the studies of [65,66], where no anthocyanins and anthocyanidins (non-glycosylated form of anthocyanins) were found in the phytochemical profile of the hydroalcoholic extract of the leaves of *Lafoensia pacari*.

Of all the macronutrients, nitrogen has the greatest influence on plant development in general [67]. Furthermore, in tree species, the nitrogen concentration has been correlated with the chlorophyll content and the SPAD value [68–74], allowing the use of SPAD as a rapid and reliable diagnostic tool to predict nitrogen status in forest nurseries [75].

To develop large-scale nursery production practices that improve seedling quality at reduced cost, more knowledge is needed on irrigation and nutrient management in container seedlings [76–79]. For mean leaf angles of -14° and -56° , an increase in irrigation volume increased all the total macronutrient and micronutrient contents, except B and Fe, which were not significantly correlated. This indicates that the application of smaller irrigation volumes at these angles has difficulty reaching the substrate surface, which likely causes a reduction in ion contact with the roots (Figure 11a,b).

According to [80], it is necessary to apply greater irrigation volumes (e.g., 10, 12, and 14 mm) in *Eucalyptus grandis* seedlings to achieve higher nutrient contents. Optimal nutrient reserves can increase shoot growth during the field establishment phase. Seedlings with optimum nutrient reserves before planting had a positive relationship with shoot growth in 78% of the studies reviewed by [47]. In addition, high nutrient content in pine seedlings developed larger root systems by maintaining a greater number of growing roots rather than by increasing the elongation rate of individual roots, which can be interpreted as a strategy to maximize foraging efficiency [81].

At mean leaf angles of 58° , 57° , 55° , 42° , 38° , 31° , and -54° , all total macronutrient contents, except Ca and Mg in *Psidium cattleyanum* (55°), were reduced with greater irrigation volumes. In these species, 75.6% of the micronutrients were also reduced with increased irrigation volumes. It is not easy to perform efficient fertilization practices that deliver

nutrients at the right time; however, new irrigation and fertilization strategies are being sought by commercial forest nurseries and researchers to improve fertilizer application and minimize negative adverse effects [82].

4.2. Seedling Growth Performance after Planting in Pot

Seedling quality is the key to successful active forest restoration programs, and especially under climate change [83–85], it has an important function in plant survival and growth [86–89]. Seedling quality assessment has evolved to consider various morphological and physiological measurement protocols in the nursery to predict future planting performance [90,91]. Thus, intrinsic growth potential is related to physiological and morphological seedling attributes and their eco-physiological behavior toward site characteristics [92].

We show that the duration of treatment effects applied at the nursery phase on the seedling growth performance after potting was variable. On the other hand, it is important to emphasize that for all species, up to 120 days after planting in pots, the effect of the irrigation volume that provided greater growth and physiological quality at the end of the nursery phase was not overcome by the other irrigation volumes applied ($p < 0.05$).

A positive correlation between the initial seedling height and stem diameter and subsequent growth after planting was found in 70% and 91% of the studies reviewed by [47], respectively. The height advantage is relevant on soils without water and nutrient deficits because the competition for light between seedlings and the site vegetation is the main factor limiting seedling development [93,94]. As a result of limited sunlight and depleted upper-profile soil moisture in areas of high vegetative competition, reductions in seedling carbon gain corresponded to substantial increases in seedling mortality and establishment failure [95,96]. Researchers agree on the importance of considering stem diameter as the best morphological attribute to predict seedling growth [97–101], especially under dry soil conditions [102].

The height and stem diameter of each species at the end of the nursery phase followed a similar pattern to that of the physiological attributes in response to the irrigation volumes applied. This correspondence between the physiological and morphological attributes was a relevant finding because quantifying the benefits of physiological plant attributes is a very narrow window.

For most nursery management programs, rapid and non-destructive morphological measurements are still best suited [103]. This is because seedling survival and subsequent establishment after planting depend not only on their physiological status but also on their morphological characteristics, ability to root, and eco-physiological response [104]. With this understanding, growers can implement appropriate nursery practices to improve seedling physiological functioning and morphological development [105].

5. Conclusions

Leaf angle can be used as a criterion to optimize irrigation in forest nurseries, prevent water and fertilizer wastage, and increase physiological tree seedling quality. Leaf angle measurements combined with concurrent assessments of leaf traits (e.g., leaf area and spatial distribution of leaves) are helpful in further understanding the effects of leaf angle variation and water regime on seedling quality.

Water access to the substrate is facilitated by tree seedlings with positive mean leaf angles; thus, an irrigation volume of 8 mm is sufficient to increase physiological tree seedling quality. Conversely, negative mean leaf angles show the opposite response, requiring the largest irrigation volume (14 mm) to increase physiological tree seedling quality, except when the mean leaf area is small and concentrated in the upper half of the stem, which facilitates the access of irrigation water to the substrate and thus satisfies seedling water requirements.

For all species, up to 120 days after planting in pots, the effect of the irrigation volume that provides greater growth and physiological quality at the end of the nursery phase is not overcome by the other irrigation volumes applied, highlighting the benefits of applying

the correct irrigation volume in tree seedlings. Furthermore, the height and stem diameter of tree seedling species at the end of the nursery phase in response to irrigation volume show a similar pattern to physiological attributes, facilitating the creation of operational procedures for active forest restoration.

Supplementary Materials: The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/f14051042/s1>: Table S1: Analysis of variance (ANOVA) for the daily transpiration rate of tree seedlings as influenced by mean leaf angles and irrigation volumes at the end of the nursery phase; Table S2: Analysis of variance (ANOVA) for the leaf water potential of tree seedlings as influenced by mean leaf angles and irrigation volumes at the end of the nursery phase; Table S3: Analysis of variance (ANOVA) for the carotenoids content of tree seedlings as influenced by mean leaf angles and irrigation volumes at the end of the nursery phase; Table S4: Analysis of variance (ANOVA) for the chlorophyll *a* content of tree seedlings as influenced by mean leaf angles and irrigation volumes at the end of the nursery phase; Table S5: Analysis of variance (ANOVA) for the chlorophyll *b* content of tree seedlings as influenced by mean leaf angles and irrigation volumes at the end of the nursery phase; Table S6: Analysis of variance (ANOVA) for the anthocyanins content of tree seedlings as influenced by mean leaf angles and irrigation volumes at the end of the nursery phase; Table S7: Analysis of variance (ANOVA) for the SPAD values of tree seedlings as influenced by mean leaf angles and irrigation volumes at the end of the nursery phase; Table S8: Mean leaf area (cm²) at the end of the nursery phase of nine tree seedling species with different mean leaf angles submitted to four daily irrigation volumes.

Author Contributions: Conceptualization, R.B.G.d.S. and M.R.d.S.; methodology, R.B.G.d.S., D.Z.d.P., M.M.P.S. and M.R.d.S.; software, R.B.G.d.S., M.M.P.S. and D.S.; formal analysis, R.B.G.d.S., D.Z.d.P., M.M.P.S., A.A.d.S.B., D.S. and M.R.d.S.; investigation, R.B.G.d.S.; writing—original draft preparation, R.B.G.d.S.; writing—review and editing, R.B.G.d.S., I.W., D.Z.d.P., D.S., M.M.P.S., A.A.d.S.B. and M.R.d.S.; visualization, R.B.G.d.S., D.S., I.W. and D.Z.d.P.; supervision, M.R.d.S.; project administration, M.R.d.S. and R.B.G.d.S. All authors have read and agreed to the published version of the manuscript.

Funding: This study was supported by São Paulo Research Foundation (FAPESP)—Grant Number 2013/17447-8.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: All data are provided in the main manuscript. Contact the corresponding author if further explanation is needed.

Acknowledgments: We wish to thank Giuseppina Pace Pereira Lima, Luiz Fernando Rolim de Almeida, Angélica Lino Rodrigues, Cláudio Roberto Ribeiro da Silva, José Aparecido Benedito, João Marques Rodrigues, Rosângela Cristina Moreci, Ludmila Ribeiro Roder, Thaís Randazzo Enz, Leonardo Albiero Gomes, and Maurício Torloni Neto.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Haase, D.; Davis, A. Developing and Supporting Quality Nursery Facilities and Staff Are Necessary to Meet Global Forest and Landscape Restoration Needs. *Reforesta* **2017**, *4*, 69–93. [\[CrossRef\]](#)
2. Grossnickle, S.C.; Kiiskila, S.B.; Haase, D.L. Seedling Ecophysiology—Five Questions To Explore in the Nursery for Optimizing Subsequent Field Success. *Tree Plant. Notes* **2020**, *63*, 112–127.
3. Haase, D.L. Understanding Forest Seedling Quality: Measurements and Interpretation. *Tree Plant. Notes* **2008**, *52*, 24–30.
4. Davis, A.S.; Jacobs, D.F. Quantifying Root System Quality of Nursery Seedlings and Relationship to Outplanting Performance. *New For.* **2005**, *30*, 295–311. [\[CrossRef\]](#)
5. Rodrigues, A.L.; Bertholdi, A.A.S.; Mantoan, L.P.B.; Vasconcellos, G.M.; Almeida, L.F.R. Photochemistry and Hydric Responses of Congeneric Croton Species at Restoration Sites under Dry Season: Implications for Species Selection. *Theor. Exp. Plant Physiol.* **2019**, *31*, 329–339. [\[CrossRef\]](#)
6. da Silva, R.B.G.; Simões, D.; da Silva, M.R. Qualidade de Mudas Clonais de Eucalyptus Urophylla x E. Grandis Em Função Do Substrato. *Rev. Bras. Eng. Agrícola Ambient.* **2012**, *16*, 297–302. [\[CrossRef\]](#)

7. da Silva, R.B.G.; Gabira, M.M.; Prado, D.Z.D.; Uesugi, G.; Simões, D.; da Silva, M.R. Influence of Mean Leaf Angles and Irrigation Volumes on Water Capture, Leaching, and Growth of Tropical Tree Seedlings. *Forests* **2020**, *11*, 1198. [\[CrossRef\]](#)
8. Beeson, R.C.; Yeager, T.H. Plant Canopy Affects Sprinkler Irrigation Application Efficiency of Container-Grown Ornamentals. *HortScience* **2003**, *38*, 1373–1377. [\[CrossRef\]](#)
9. Yeager, T.; Million, J.; Larsen, C.; Stamps, B. Florida Nursery Best Management Practices: Past, Present, and Future. *Horttechnology* **2010**, *20*, 82–88. [\[CrossRef\]](#)
10. Reinhardt, D.; Kuhlemeier, C. Plant Architecture. *EMBO Rep.* **2002**, *3*, 846–851. [\[CrossRef\]](#)
11. Liu, J.; Skidmore, A.K.; Wang, T.; Zhu, X.; Premier, J.; Heurich, M.; Beudert, B.; Jones, S. Variation of Leaf Angle Distribution Quantified by Terrestrial LiDAR in Natural European Beech Forest. *ISPRS J. Photogramm. Remote Sens.* **2019**, *148*, 208–220. [\[CrossRef\]](#)
12. King, D.A. The Functional Significance of Leaf Angle in Eucalyptus. *Aust. J. Bot.* **1997**, *45*, 619. [\[CrossRef\]](#)
13. Falster, D.S.; Westoby, M. Leaf Size and Angle Vary Widely across Species: What Consequences for Light Interception? *New Phytol.* **2003**, *158*, 509–525. [\[CrossRef\]](#)
14. van Zanten, M.; Pons, T.L.; Janssen, J.A.M.; Voisenek, L.A.C.J.; Peeters, A.J.M. On the Relevance and Control of Leaf Angle. *Crit. Rev. Plant Sci.* **2010**, *29*, 300–316. [\[CrossRef\]](#)
15. Hagemeyer, M.; Leuschner, C. Functional Crown Architecture of Five Temperate Broadleaf Tree Species: Vertical Gradients in Leaf Morphology, Leaf Angle, and Leaf Area Density. *Forests* **2019**, *10*, 265. [\[CrossRef\]](#)
16. Chianucci, F.; Pisek, J.; Raabe, K.; Marchino, L.; Ferrara, C.; Corona, P. A Dataset of Leaf Inclination Angles for Temperate and Boreal Broadleaf Woody Species. *Ann. For. Sci.* **2018**, *75*, 50. [\[CrossRef\]](#)
17. Pisek, J.; Adamson, K. Dataset of Leaf Inclination Angles for 71 Different Eucalyptus Species. *Data Br.* **2020**, *33*, 106391. [\[CrossRef\]](#)
18. Pisek, J.; Diaz-Pines, E.; Matteucci, G.; Noe, S.; Rebmann, C. On the Leaf Inclination Angle Distribution as a Plant Trait for the Most Abundant Broadleaf Tree Species in Europe. *Agric. For. Meteorol.* **2022**, *323*, 109030. [\[CrossRef\]](#)
19. Niinemets, Ü. A Review of Light Interception in Plant Stands from Leaf to Canopy in Different Plant Functional Types and in Species with Varying Shade Tolerance. *Ecol. Res.* **2010**, *25*, 693–714. [\[CrossRef\]](#)
20. Zou, J.; Zhong, P.; Hou, W.; Zuo, Y.; Leng, P. Estimating Needle and Shoot Inclination Angle Distributions and Projection Functions in Five Larix Principis-Rupprechtii Plots via Leveled Digital Camera Photography. *Forests* **2020**, *12*, 30. [\[CrossRef\]](#)
21. Deguchi, R.; Koyama, K. Photosynthetic and Morphological Acclimation to High and Low Light Environments in Petasites Japonicus Subsp. Giganteus. *Forests* **2020**, *11*, 1365. [\[CrossRef\]](#)
22. de Mattos, E.M.; Binkley, D.; Campoe, O.C.; Alvares, C.A.; Stape, J.L. Variation in Canopy Structure, Leaf Area, Light Interception and Light Use Efficiency among Eucalyptus Clones. *For. Ecol. Manag.* **2020**, *463*, 118038. [\[CrossRef\]](#)
23. Peguero-Pina, J.J.; Vilagrosa, A.; Alonso-Forn, D.; Ferrio, J.P.; Sancho-Knapik, D.; Gil-Pelegrín, E. Living in Drylands: Functional Adaptations of Trees and Shrubs to Cope with High Temperatures and Water Scarcity. *Forests* **2020**, *11*, 1028. [\[CrossRef\]](#)
24. Wu, X.; Fan, W.; Du, H.; Ge, H.; Huang, F.; Xu, X. Estimating Crown Structure Parameters of Moso Bamboo: Leaf Area and Leaf Angle Distribution. *Forests* **2019**, *10*, 686. [\[CrossRef\]](#)
25. Liu, Q.; Xie, L.; Li, F. Dynamic Simulation of the Crown Net Photosynthetic Rate for Young Larix Olgensis Henry Trees. *Forests* **2019**, *10*, 321. [\[CrossRef\]](#)
26. Liu, Y.; Shan, Y.; Ying, H.; Wala, D.; Zhang, X.; Ruhan, A.; Rina, S.; Rina, S. Examining the Angular Effects of UAV-LS on Vegetation Metrics Using a Framework for Mediating Effects. *Forests* **2022**, *13*, 1221. [\[CrossRef\]](#)
27. Wen, Y.; Zhuang, L.; Wang, H.; Hu, T.; Fan, W. An Automated Hemispherical Scanner for Monitoring the Leaf Area Index of Forest Canopies. *Forests* **2022**, *13*, 1355. [\[CrossRef\]](#)
28. Zhang, B.; Wang, X.; Yuan, X.; An, F.; Zhang, H.; Zhou, L.; Shi, J.; Yun, T. Simulating Wind Disturbances over Rubber Trees with Phenotypic Trait Analysis Using Terrestrial Laser Scanning. *Forests* **2022**, *13*, 1298. [\[CrossRef\]](#)
29. Qu, Y.; Wang, J.; Song, J.; Wang, J. Potential and Limits of Retrieving Conifer Leaf Area Index Using Smartphone-Based Method. *Forests* **2017**, *8*, 217. [\[CrossRef\]](#)
30. Anna, K.-I.; Sylwia, Ł.; Marcin, Z.; Ewa, S.-O.; Wojtan, B. Variability of Leaf Wetting and Water Storage Capacity of Branches of 12 Deciduous Tree Species. *Forests* **2020**, *11*, 1158. [\[CrossRef\]](#)
31. Dong, L.; Han, H.; Kang, F.; Cheng, X.; Zhao, J.; Song, X. Rainfall Partitioning in Chinese Pine (Pinus Tabuliformis Carr.) Stands at Three Different Ages. *Forests* **2020**, *11*, 243. [\[CrossRef\]](#)
32. Fladung, M. Targeted CRISPR/Cas9-Based Knock-Out of the Rice Orthologs TILLER ANGLE CONTROL 1 (TAC1) in Poplar Induces Erect Leaf Habit and Shoot Growth. *Forests* **2021**, *12*, 1615. [\[CrossRef\]](#)
33. Zhang, L.; Yang, J.; Huang, Y.; Jia, Z.; Fang, Y. Leaf Venation Variation and Phenotypic Plasticity in Response to Environmental Heterogeneity in Parrotia Subaequalis (H. T. Chang) R. M. Hao et H. T. Wei, An Endemic and Endangered Tree Species from China. *Forests* **2018**, *9*, 247. [\[CrossRef\]](#)
34. Roder, L.R.; Guerrini, I.A.; Lozano Sivilsaca, D.C.; Yaguana Puglla, C.A.; Góes de Moraes, F.; Pinheiro da Silva, J.; Batista Fonseca, R.C.; Umbelino, M.T.; James, J.N.; Capra, G.F.; et al. Atlantic Rainforest Natural Regeneration in Fragmented Formations Affected by Increasing Human Disturbance. *J. Environ. Manag.* **2023**, *325*, 116521. [\[CrossRef\]](#) [\[PubMed\]](#)
35. Christiansen, J.E. *Irrigation by Sprinkling*; University of California: Berkeley, CA, USA, 1942.
36. Merriam, J.L.; Keller, J. *Farm Irrigation System Evaluation: A Guide for Management*; Utah State University: Logan, UT, USA, 1978.

37. Aroca, R.; Porcel, R.; Ruiz-Lozano, J.M. How Does Arbuscular Mycorrhizal Symbiosis Regulate Root Hydraulic Properties and Plasma Membrane Aquaporins in *Phaseolus Vulgaris* under Drought, Cold or Salinity Stresses? *New Phytol.* **2007**, *173*, 808–816. [\[CrossRef\]](#) [\[PubMed\]](#)
38. Sims, D.A.; Gamon, J.A. Relationships between Leaf Pigment Content and Spectral Reflectance across a Wide Range of Species, Leaf Structures and Developmental Stages. *Remote Sens. Environ.* **2002**, *81*, 337–354. [\[CrossRef\]](#)
39. Landis, T.D. Mineral Nutrition as an Index of Seedling Quality: Principles and Applications. In *Evaluating Seedling Quality: Principles, Procedures, and Predictive Abilities of Major Tests*; Duryea, M.L., Ed.; Forest Research Laboratory, Oregon State University: Corvallis, OR, USA, 1985; pp. 29–48.
40. Oliet, J.A.; Tejada, M.; Salifu, K.F.; Collazos, A.; Jacobs, D.F. Performance and Nutrient Dynamics of Holm Oak (*Quercus Ilex* L.) Seedlings in Relation to Nursery Nutrient Loading and Post-Transplant Fertility. *Eur. J. For. Res.* **2009**, *128*, 253–263. [\[CrossRef\]](#)
41. Xu, L.; Zhang, X.; Zhang, D.; Wei, H.; Guo, J. Using Morphological Attributes for the Fast Assessment of Nutritional Responses of Buddhist Pine (*Podocarpus Macrophyllus* [Thunb.] D. Don) Seedlings to Exponential Fertilization. *PLoS ONE* **2019**, *14*, e0225708. [\[CrossRef\]](#)
42. Shapiro, S.S.; Wilk, M.B. An Analysis of Variance Test for Normality (Complete Samples). *Biometrika* **1965**, *52*, 591–611. [\[CrossRef\]](#)
43. Fisher, R.A. *Statistical Methods For Research Workers*; Oliver and Boyd: London, UK, 1925.
44. Scott, A.J.; Knott, M. A Cluster Analysis Method for Grouping Means in the Analysis of Variance. *Biometrics* **1974**, *30*, 507–512. [\[CrossRef\]](#)
45. StatSoft, Inc. *STATISTICA (Data Analysis Software System), version 8*; StatSoft, Inc.: Tulsa, OK, USA, 2007.
46. Delgado, L.G.M.; da Silva, R.B.G.; Gabira, M.M.; Rodrigues, A.L.; Simões, D.; de Almeida, L.F.R.; da Silva, M.R. Mean Leaf Angles Affect Irrigation Efficiency and Physiological Responses of Tropical Species Seedling. *Forests* **2022**, *13*, 832. [\[CrossRef\]](#)
47. Grossnickle, S.C.; MacDonald, J.E. Why Seedlings Grow: Influence of Plant Attributes. *New For.* **2018**, *49*, 1–34. [\[CrossRef\]](#)
48. Taiz, L.; Zeiger, E. *Plant Physiology*; ArtMed: Porto Alegre, Brazil, 2017.
49. Vishwakarma, K.; Upadhyay, N.; Kumar, N.; Yadav, G.; Singh, J.; Mishra, R.K.; Kumar, V.; Verma, R.; Upadhyay, R.G.; Pandey, M.; et al. Absciscic Acid Signaling and Abiotic Stress Tolerance in Plants: A Review on Current Knowledge and Future Prospects. *Front. Plant Sci.* **2017**, *8*, 161. [\[CrossRef\]](#)
50. Yuan, Z.-S.; Liu, F.; Xie, B.-G.; Zhang, G.-F. The Growth-Promoting Effects of Endophytic Bacteria on *Phyllostachys Edulis*. *Arch. Microbiol.* **2018**, *200*, 921–927. [\[CrossRef\]](#)
51. Yang, X.; Li, R.; Jablonski, A.; Stovall, A.; Kim, J.; Yi, K.; Ma, Y.; Beverly, D.; Phillips, R.; Novick, K.; et al. Leaf Angle as a Leaf and Canopy Trait: Rejuvenating Its Role in Ecology with New Technology. *Ecol. Lett.* **2023**, 1–16. [\[CrossRef\]](#) [\[PubMed\]](#)
52. Durigan, G.; Baitello, J.B.; Franco, G.A.D.C.; Siqueira, M.F. *Plantas Do Cerrado Paulista: Imagens de Uma Paisagem Ameaçada*; Páginas & Letras: São Paulo, Brazil, 2004; ISBN 85-86508-32-2.
53. Brandes, A.F.D.N.; Sánchez-Tapia, A.; Sansevero, J.B.B.; Albuquerque, R.P.; Barros, C.F. Fire Records in Tree Rings of *Moquiniastrum Polymorphum*: Potential for Reconstructing Fire History in the Brazilian Atlantic Forest. *Acta Bot. Bras.* **2019**, *33*, 61–66. [\[CrossRef\]](#)
54. Morgan, J.M. Osmoregulation and Water Stress in Higher Plants. *Annu. Rev. Plant Physiol.* **1984**, *35*, 299–319. [\[CrossRef\]](#)
55. White, D.A.; Turner, N.C.; Galbraith, J.H. Leaf Water Relations and Stomatal Behavior of Four Allopatric Eucalyptus Species Planted in Mediterranean Southwestern Australia. *Tree Physiol.* **2000**, *20*, 1157–1165. [\[CrossRef\]](#)
56. Lichtenthaler, H.K. Biosynthesis, Accumulation and Emission of Carotenoids, α -Tocopherol, Plastoquinone, and Isoprene in Leaves under High Photosynthetic Irradiance. *Photosynth. Res.* **2007**, *92*, 163–179. [\[CrossRef\]](#)
57. Nisar, N.; Li, L.; Lu, S.; Khin, N.C.; Pogson, B.J. Carotenoid Metabolism in Plants. *Mol. Plant* **2015**, *8*, 68–82. [\[CrossRef\]](#)
58. de Sousa Leite, T.; da Silva Dias, N.; Oliveira de Freitas, R.M.; Dallabona Dombroski, J.L.; de Sousa Leite, M.; Martins de Farias, R. Ecophysiological and Biochemical Responses of Two Tree Species from a Tropical Dry Forest to Drought Stress and Recovery. *J. Arid Environ.* **2022**, *200*, 104720. [\[CrossRef\]](#)
59. Pukacki, P.M.; Kamińska-Rożek, E. Effect of Drought Stress on Chlorophyll a Fluorescence and Electrical Admittance of Shoots in Norway Spruce Seedlings. *Trees* **2005**, *19*, 539–544. [\[CrossRef\]](#)
60. Vasques, A.R.; Pinto, G.; Dias, M.C.; Correia, C.M.; Moutinho-Pereira, J.M.; Vallejo, V.R.; Santos, C.; Keizer, J.J. Physiological Response to Drought in Seedlings of *Pistacia Lentiscus* (Mastic Tree). *New For.* **2016**, *47*, 119–130. [\[CrossRef\]](#)
61. Frosi, G.; Harand, W.; de Oliveira, M.T.; Pereira, S.; Cabral, S.P.; Montenegro, A.A.D.A.; Santos, M.G. Different Physiological Responses under Drought Stress Result in Different Recovery Abilities of Two Tropical Woody Evergreen Species. *Acta Bot. Bras.* **2017**, *31*, 153–160. [\[CrossRef\]](#)
62. Bhusal, N.; Lee, M.; Reum Han, A.; Han, A.; Kim, H.S. Responses to Drought Stress in *Prunus Sargentii* and *Larix Kaempferi* Seedlings Using Morphological and Physiological Parameters. *For. Ecol. Manag.* **2020**, *465*, 118099. [\[CrossRef\]](#)
63. Karimi, A.; Tabari, M.; Javanmard, Z.; Bader, M.K.-F. Drought Effects on Morpho-Physiological and Biochemical Traits in Persian Oak and Black Poplar Seedlings. *Forests* **2022**, *13*, 399. [\[CrossRef\]](#)
64. Lassouane, N.; Aïd, F.; Lutts, S. Water Stress Impact on Young Seedling Growth of *Acacia Arabica*. *Acta Physiol. Plant.* **2013**, *35*, 2157–2169. [\[CrossRef\]](#)
65. Firmo, W.C.A.; Miranda, M.V.; Coutinho, G.S.L.; Silveira, L.M.S.; Olea, R.S.G. Estudo Fitoquímico e Avaliação Da Atividade Antibacteriana de *Lafoensia Pacari* (Lythraceae). *Publ. UEPG Cienc. Biol. Saude* **2014**, *20*, 7–12. [\[CrossRef\]](#)

66. Lima, P.C.; Santos, M.G.; Calabrese, K.S.; Silva, A.L.A.; Almeida, F. Avaliação Da Capacidade Leishmanicida de Espécies Vegetais Do Cerrado. *Rev. Patol. Trop.* **2015**, *44*, 45–55. [\[CrossRef\]](#)
67. Bojovic, B.; Stojanovic, J. Chlorophyll and Carotenoid Content in Wheat Cultivars as a Function of Mineral Nutrition. *Arch. Biol. Sci.* **2005**, *57*, 283–290. [\[CrossRef\]](#)
68. Richardson, A.D.; Duigan, S.P.; Berlyn, G.P. An Evaluation of Noninvasive Methods to Estimate Foliar Chlorophyll Content. *New Phytol.* **2002**, *153*, 185–194. [\[CrossRef\]](#)
69. Chenard, C.H.; Kopsell, D.A.; Kopsell, D.E. Nitrogen Concentration Affects Nutrient and Carotenoid Accumulation in Parsley. *J. Plant Nutr.* **2005**, *28*, 285–297. [\[CrossRef\]](#)
70. Madeira, A.C.; Madeira, M.; Fabião, A.; Marques, P.; Carneiro, M. Avaliação Da Nutrição de Plantações Jovens de Eucalipto Por Análise Foliar e Métodos Não Destrutivos. *Rev. Ciências Agrárias* **2009**, *32*, 139–153. [\[CrossRef\]](#)
71. Júnior, O.A.D.O.; Cairo, P.A.R.; De Novaes, A.B. Características Morfofisiológicas Associadas à Qualidade de Mudanças de Eucalyptus Urophylla Produzidas Em Diferentes Substratos. *Rev. Árvore* **2011**, *35*, 1173–1180. [\[CrossRef\]](#)
72. De Carvalho, R.P.; Davide, L.M.C.; Borges, F.L.G.; Davide, A.C.; Daniel, O. Respostas Morfofisiológicas Entre Procedências de Canafistula Submetidas a Diferentes Condições Hídricas e Nutricionais. *Pesqui. Florest. Bras.* **2015**, *35*, 179–188. [\[CrossRef\]](#)
73. Uesugi, G.; Favan, J.R.; De Moraes, C.B.; Wanginiak, T.C.R.; da Silva, M.R. Utilização Do SPAD-502 Para a Predição Dos Teores de Nitrogênio Em Mudanças de Croton Urucurana Baill. (Nota Científica). *Rev. do Inst. Florest.* **2015**, *27*, 177–181. [\[CrossRef\]](#)
74. de Moura, A.R.; Nogueira, R.J.M.C.; da Silva, J.A.A.; de Lima, T.V. Relações Hídricas e Solutos Orgânicos Em Plantas Jovens de Jatropha Curcas L. Sob Diferentes Regimes Hídricos. *Ciência Florest.* **2016**, *26*, 345–354. [\[CrossRef\]](#)
75. Acevedo, M.; Rubilar, R.; Dumroese, R.K.; Ovalle, J.F.; Sandoval, S.; Chassin-Trubert, R. Nitrogen Loading of Eucalyptus Globulus Seedlings: Nutritional Dynamics and Influence on Morphology and Root Growth Potential. *New For.* **2021**, *52*, 31–46. [\[CrossRef\]](#)
76. Scagel, C.F.; Bi, G.; Fuchigami, L.H.; Regan, R.P. Irrigation Frequency Alters Nutrient Uptake in Container-Grown Rhododendron Plants Grown with Different Rates of Nitrogen. *HortScience* **2012**, *47*, 189–197. [\[CrossRef\]](#)
77. Silva, D.; Stuepp, C.A.; Wenfling, I.; Helm, C.; Angelo, A.C. Influence of Seed Storage Conditions on Quality of Torresea Acreana Seedlings. *Cerne* **2019**, *25*, 60–67. [\[CrossRef\]](#)
78. Manguera, J.R.S.A.; Holl, K.D.; Rodrigues, R.R. Enrichment Planting to Restore Degraded Tropical Forest Fragments in Brazil. *Ecosyst. People* **2019**, *15*, 3–10. [\[CrossRef\]](#)
79. Simões, D.; Gil, J.F.S.; da Silva, R.B.G.; Munis, R.A.; da Silva, M.R. Stochastic Economic Analysis of Investment Projects in Forest Restoration Involving Containerized Tree Seedlings in Brazil. *Forests* **2021**, *12*, 1381. [\[CrossRef\]](#)
80. Lopes, J.L.W.; Guerrini, I.A.; Saad, J.C.C.; da Silva, M.R. Nutrição Mineral de Mudanças de Eucalipto Produzidas Sob Diferentes Lâminas de Irrigação e Substratos. *Rev. Bras. Ciência Do Solo* **2007**, *31*, 713–722. [\[CrossRef\]](#)
81. Toca, A.; Oliet, J.A.; Villar-Salvador, P.; Martínez Catalán, R.A.; Jacobs, D.F. Ecologically Distinct Pine Species Show Differential Root Development after Outplanting in Response to Nursery Nutrient Cultivation. *For. Ecol. Manag.* **2019**, *451*, 117562. [\[CrossRef\]](#)
82. Oliet, J.A.; Salazar, J.M.; Villar, R.; Robredo, E.; Valladares, F. Fall Fertilization of Holm Oak Affects N and P Dynamics, Root Growth Potential, and Post-Planting Phenology and Growth. *Ann. For. Sci.* **2011**, *68*, 647–656. [\[CrossRef\]](#)
83. del Campo, A.D.; Segura-Orenga, G.; Molina, A.J.; González-Sanchis, M.; Reyna, S.; Hermoso, J.; Ceacero, C.J. On the Need to Further Refine Stock Quality Specifications to Improve Reforestation under Climatic Extremes. *Forests* **2022**, *13*, 168. [\[CrossRef\]](#)
84. Vieira, D.L.M.; Rodrigues, S.B.; Jakovac, C.C.; da Rocha, G.P.E.; Reis, F.; Borges, A. Active Restoration Initiates High Quality Forest Succession in a Deforested Landscape in Amazonia. *Forests* **2021**, *12*, 1022. [\[CrossRef\]](#)
85. Fiore, N.V.; Ferreira, C.C.; Dzedzej, M.; Massi, K.G. Monitoring of a Seedling Planting Restoration in a Permanent Preservation Area of the Southeast Atlantic Forest Biome, Brazil. *Forests* **2019**, *10*, 768. [\[CrossRef\]](#)
86. Stape, J.L.; Gonçalves, J.L.M.; Gonçalves, A.N. Relationships between Nursery Practices and Field Performance for Eucalyptus Plantations in Brazil. *New For.* **2001**, *22*, 19–41. [\[CrossRef\]](#)
87. Campbell, K.A.; Hawkins, C.D.B. Effect of Seed Source and Nursery Culture on Paper Birch (Betula Papyrifera) Uprooting Resistance and Field Performance. *For. Ecol. Manag.* **2004**, *196*, 425–433. [\[CrossRef\]](#)
88. Olivo, V.B.; Buduba, C.G. Influencia de Seis Sustratos En El Crecimiento de Pinus Ponderosa Producido En Contenedores Bajo Condiciones de Invernáculo. *Bosque (Valdivia)* **2006**, *27*, 267–271. [\[CrossRef\]](#)
89. Devetaković, J.R.; Pavlović, S.; Krinulović, L.; Janković, I.K. Field Performance of Austrian Pine Bareroot Seedlings in Comparison to Seedlings Pattern and Density in the Nursery. *Reforest* **2021**, *11*, 27–35.
90. Jacobs, D.F.; Salifu, K.F.; Seifert, J.R. Relative Contribution of Initial Root and Shoot Morphology in Predicting Field Performance of Hardwood Seedlings. *New For.* **2005**, *30*, 235–251. [\[CrossRef\]](#)
91. Jacobs, D.F.; Davis, A.S.; Dumroese, R.K.; Burney, O.T. Nursery Cultural Techniques Facilitate Restoration of Acacia Koa Competing with Invasive Grass in a Dry Tropical Forest. *Forests* **2020**, *11*, 1124. [\[CrossRef\]](#)
92. Grossnickle, S. Restoration Silviculture: An Ecophysiological Perspective—Lessons Learned across 40 Years. *Reforest* **2016**, *1*, 1–36. [\[CrossRef\]](#)
93. Thiffault, N.; Jobidon, R.; Munson, A.D. Comparing Large Containerized and Bareroot Conifer Stock on Sites of Contrasting Vegetation Composition in a Non-Herbicide Scenario. *New For.* **2014**, *45*, 875–891. [\[CrossRef\]](#)
94. Gardiner, R.; Shoo, L.P.; Dwyer, J.M. Look to Seedling Heights, Rather than Functional Traits, to Explain Survival during Extreme Heat Stress in the Early Stages of Subtropical Rainforest Restoration. *J. Appl. Ecol.* **2019**, *56*, 2687–2697. [\[CrossRef\]](#)

95. Johnson, D.M.; Smith, W.K. Refugial Forests of the Southern Appalachians: Photosynthesis and Survival in Current-Year Abies Fraseri Seedlings. *Tree Physiol.* **2005**, *25*, 1379–1387. [[CrossRef](#)]
96. Pinto, J.; McNassar, B.; Kildisheva, O.; Davis, A. Stocktype and Vegetative Competition Influences on Pseudotsuga Menziesii and Larix Occidentalis Seedling Establishment. *Forests* **2018**, *9*, 228. [[CrossRef](#)]
97. Gomes, J.M.; Couto, L.; Leite, H.G.; Xavier, A.; Garcia, S.L.R. Parâmetros Morfológicos Na Avaliação de Qualidade de Mudanças de Eucalyptus Grandis. *Rev. Árvore* **2002**, *26*, 655–664. [[CrossRef](#)]
98. de Souza, C.A.M.; de Oliveira, R.B.; Filho, S.M.; Lima, J.S.D.S. Crescimento Em Campo de Espécies Florestais Em Diferentes Condições de Adubações. *Ciência Florest.* **2006**, *16*, 243–249. [[CrossRef](#)]
99. Downes, G.M.; Drew, D.; Battaglia, M.; Schulze, D. Measuring and Modelling Stem Growth and Wood Formation: An Overview. *Dendrochronologia* **2009**, *27*, 147–157. [[CrossRef](#)]
100. Binotto, A.F.; Lúcio, A.D.C.; Lopes, S.J. Correlations between Growth Variables and the Dickson Quality Index in Forest Seedlings. *Cerne* **2010**, *16*, 457–464. [[CrossRef](#)]
101. Speck, T.; Burgert, I. Plant Stems: Functional Design and Mechanics. *Annu. Rev. Mater. Res.* **2011**, *41*, 169–193. [[CrossRef](#)]
102. South, D.B.; Rakestraw, J.L.; Lowerts, G.A. Early Gains from Planting Large-Diameter Seedlings and Intensive Management Are Additive for Loblolly Pine. *New For.* **2001**, *22*, 97–110. [[CrossRef](#)]
103. Ivetić, V.; Grossnickle, S.; Škorić, M. Forecasting the Field Performance of Austrian Pine Seedlings Using Morphological Attributes. *iFor. Biogeosci. For.* **2016**, *10*, 99. [[CrossRef](#)]
104. Grossnickle, S.C. Why Seedlings Survive: Influence of Plant Attributes. *New For.* **2012**, *43*, 711–738. [[CrossRef](#)]
105. Grossnickle, S. Seedling Establishment on a Forest Restoration Site—An Ecophysiological Perspective. *Reforesta* **2018**, *6*, 110–139. [[CrossRef](#)]

Disclaimer/Publisher’s Note: The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.