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# Toward Sustainable Cultivation of *Pinus occidentalis* Swartz in Haiti: Effects of Alternative Growing Media and Containers on Seedling Growth and Foliar Chemistry

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**Abstract:** Haiti has suffered great losses from deforestation, with little forest cover remaining today. Current reforestation efforts focus on seedling quantity rather than quality. This study examined limitations to the production of high-quality seedlings of the endemic Hispaniolan pine (*Pinus occidentalis* Swartz). Recognizing the importance of applying sustainable development principles to pine forest restoration, the effects of growing media and container types on seedling growth were evaluated with the goal of developing a propagation protocol to produce high-quality seedlings using economically feasible nursery practices. With regard to growing media, seedlings grew best in compost-based media amended with sand. Topsoil, widely used in nurseries throughout Haiti, produced the smallest seedlings overall. Despite a low water holding capacity and limited manganese, compost-based media provided adequate levels of essential mineral nutrients (particularly nitrogen), which allowed for sufficient seedling nutrition. Seedling shoot and root growth, as well as the ratio of shoot biomass to root biomass, were greater in polybags relative to D40s. Results indicate that economically feasible improvements to existing nursery practices in Haiti can improve the early growth rates of *P. occidentalis* seedlings.

**Keywords:** compost; foliar nutrients; Hispaniolan pine; pine forests; seedling quality; sustainable development principles; tropical forest nursery

## 1. Introduction

Haiti, a tropical country with a landmass that was at one time 60% forested, has suffered great losses from deforestation [1], with little forest cover remaining today [2]. While generating new forests may seem a daunting task [3], the production of high-quality seedlings is an imperative first step. These seedlings must be grown specifically for reforestation, be economically accessible to local people, and be produced using locally available materials [4]. Nursery practices in Haiti, however, focus primarily on seedling quantity rather than quality. Multiple studies have shown that outplanting performance on reforestation sites correlates highly with seedling quality [5–7].

In a nursery system, seedling quality is often quantified by several morphological and physiological measurements. No single measurement can reliably predict performance; however, many studies suggest that seedling root-collar-diameter, shoot height, root volume, and the ratio of shoot-to-root biomass correlate highly with outplanting success [8–10].

Proper nursery culture has the greatest impact on seedling quality. In modern forest seedling nurseries, growers use high-quality growing media in concert with cutting-edge container technology and a wide array of fertilizers in a controlled environment. These ideal growing conditions and associated resources allow modern nurseries to easily implement the Target Plant Concept, which is defined as the specific physiological and morphological plant characteristics cultivated in the nursery that lead to the growth and survival of the outplanted seedling at a particular site [4]. The majority of nurseries located in areas of tropical deforestation, including Haiti, lack access to many of these resources; these nurseries must produce healthy seedlings using economically practical methods and available resources [11]. Effective use of the Target Plant Concept [4] in these resource-limited nurseries to better connect nursery cultural practices, and the resultant seedlings, with the anticipated field conditions, empowers reforestation managers with a framework that can lead to measurable success in post-planting seedling establishment.

Access to moisture and nutrients are critical to seedling quality [12,13] and are often managed through growing media. In Haiti, like many developing countries, topsoil is the primary component of available potting media [14,15], despite evidence that using topsoil in container nurseries often results in low outplanting success [16]. In addition to issues associated with poor drainage and compaction, topsoil is an unsustainable resource and is particularly valuable in heavily deforested regions already suffering from erosional soil losses [14]. Various alternative sources of growing media may be used [17], such as rice hulls, sand, compost, sawdust, or pine bark [5]. Incorporating compost as a component of growing media can prove highly valuable but requires batch testing to account for variability among feedstock sources [15,18–20]. Given the emerging compost industry in Haiti [21], local sources may be available. Amending potting media with vermicompost improved germination of container-grown maritime pine (*Pinus pinaster* Aiton) [18], and both germination and seedling growth of container-grown alleppo pine (*Pinus halepensis* Miller) were greater when activated sewage sludge was incorporated into peat-based media [19].

A second factor to consider when producing nursery-grown seedlings is container type [22,23]. Forest seedling nursery containers must perform a combination of functions, and the right container choice will vary by species [24]. Size (volume and depth) and design features work to mitigate root spiraling and influence overall root architecture [25]. In many developing countries, including Haiti, nursery growers typically use small plastic bags (i.e., polybags) as containers for growing tree seedlings. This container type is widely available and is lightweight and collapsible, which greatly reduces shipping costs. However, polybags have been associated with several plant growth and development issues, including root malformation, which can reduce outplanting success [26,27].

Hispaniolan pine (*Pinus occidentalis* Swartz), an endangered tree species native to the island of Hispaniola (comprising Haiti and the Dominican Republic), has been recognized as a species for over 200 years but has received limited scientific attention [28]. Given the location of the remaining pine forests at high elevation [29], the species also represents a critical component of restoration programs aimed at conserving soil and reducing damage during heavy rains. Since container type and growing media are two important considerations for the production of high-quality seedlings essential for reforestation success, the objective of this study was to examine the influence of growing media and container type on the development of Hispaniolan pine seedlings. We hypothesized that seedlings grown in nutrient rich, compost-based growing media would exhibit sufficient foliar nutrient levels and greater early seedling growth rates relative to unamended peat-based media or topsoil. We also hypothesized that seedling growth would be greater in rigid-walled D40 containers relative to polybags.

## 2. Materials and Methods

### 2.1. Experimental Design

The experiment followed a randomized complete block design (RCBD) with a factorial structure (five media treatments  $\times$  two container types) containing five replicates (i.e., blocks) per treatment. Each tray representing a growing medium and container treatment combination was considered as a block, and containers within each block were randomized weekly to minimize the effects of the nursery environment.

Five growing media mixtures were compared in this study: (1) 100% peat-based [Pe] (an unamended media consisting of 45% Canadian sphagnum peat moss, 45% coarse-grade vermiculite, and 10% fine-aged bark by volume; SunGro<sup>®</sup> Metro Mix, Agawam, MA, USA); (2) 100% “topsoil” [T] (60% topsoil, 20% animal manure, and 20% bark mulch; NuLife Topsoil, Waupaca, WI, USA); (3) 80:20 compost:topsoil [CT] (municipal biosolids and yard waste feedstock; Eko Compost, Lewiston, ID, USA); (4) 80:20 compost:grit [CG] (Grit size medium, 1.2–4.8 mm, Target<sup>®</sup> Forestry Nursery Grit, Burnaby, BC, USA); (5) 70:20:10 compost:topsoil:grit [CTG]. Pe is widely used in commercial forest tree seedling production nurseries in the US as well as other developed countries [30]. Topsoil is widely used in container nursery systems in developing countries, including Haiti [11,14,15]; however, our mix was a commercially available product. The three compost-based media types serve as potential alternatives.

Hispaniolan pine seeds from a government-funded seed bank in the Dominican Republic (Nigua Seed Bank, Santo Domingo, Dominican Republic, 18°22′34.644″ N, 70°4′9.7674″ W, provenance unknown) were soaked in distilled water for 12 h prior to sowing, as recommended by the Nigua Seed Bank. Since the number of seeds obtained was lower than anticipated and to ensure that all available seeds were used in the experiment and that each treatment received an equal number of seeds, 1–2 seeds were directly sown into each container on 12 June 2014 at the University of Idaho Pitkin Forest Nursery in Moscow, Idaho (46°43′32.0″ N, 116°57′20.4″ W). Plants were propagated in a greenhouse with daytime temperatures ranging from 10–27 °C and nighttime temperatures ranging from 5–16 °C; relative humidity ranged from 15%–100% over the growing season. No supplemental lighting was provided and daylength ranged from 9 h 31 min to 15 h 51 min over the course of the experiment. Seeds were sown into D40 (656 mL, 6.4 cm diam, 25.4 cm height; Stuewe and Sons Inc., Tangent, OR, USA) and polybag (946 mL, 7.6 cm diam, 19.1 cm height; Peaceful Valley Farm Supply, Grass Valley, CA, USA) containers. D40 containers are designed for growing tree seedlings and are rigid containers made of recycled polypropylene resin with internal longitudinal ridges and five bottom drainage holes. Polybag containers were modified to the same volume of D40 containers using a heat sealer (Uline, Pleasant Prairie, WI, USA). The heat sealer was used to close off excess container space vertically while avoiding loss of container depth. Each of the five growing media types was premixed and used to fill 100 containers of each container type, for a total of 1000 containers. After direct sowing, containers were covered with Deluxe Seed Guard germination cloth (Dewitt Company Inc., Sikeston, MO, USA) and irrigated using an overhead boom system three times daily with 3 passes per application until germination ceased. Seedlings in each treatment combination were then irrigated when block weights (one tray consisting of 5 to 20 seedlings) reached 80% of the weight at field capacity via the nursery manager method [31]. Using this method, D40-grown seedlings received 18, 9, 10, 6, and 7 irrigation events and polybag-grown seedlings received 15, 16, 12, 11, and 10 irrigation events over the 22-week growing period across Pe, T, CT, CG, and CTG treatments, respectively. No fertilizer was added at any point throughout the growing regime, representative of many situations where fertilizer is difficult to obtain in developing countries.

### 2.2. Sampling

Destructive sampling occurred during the week of 15 December 2014 for all seedlings. Measurements included morphological plant growth metrics of height (HT) and root-collar diameter (RCD), root volume (RV), root dry mass (RDM), and shoot dry mass (SDM). First, root systems were carefully washed clean of all growing media. Second, RV was determined by water displacement [32]. Next, seedlings were severed

at the root-collar, and roots and shoots were dried separately in paper bags at 70 °C for 72 h. Following drying, SDM and RDM were used to determine seedling shoot-to-root ratios (S:R). Tissue samples were collected at the time of destructive sampling from the entire shoot of each seedling and analyzed for nutrient concentrations (A & L Great Lakes Laboratories, Fort Wayne, IN, USA).

Media samples ( $n = 5$ ) from each treatment were also analyzed for nutrient concentrations (C, N,  $\text{NO}_3$ , P, K, Ca, Mg, and Na), pH, and electrical conductivity (EC) at the beginning and end of the growing season (A & L Great Lakes Laboratories, Fort Wayne, IN, USA). Nutrient concentrations were determined via the saturated media extract (SME) method, whereby growing media samples were saturated with distilled water and allowed to equilibrate for one hour. After equilibration, pH measurements were taken directly from the media slurry. All other analyses were performed on the extracted leachate from the slurry obtained via a Buchner funnel lined with filter paper [33].

Media bulk density was approximated for each treatment by filling five of each container type for each media type and then oven-drying the media from each container in paper bags at 100 °C for 48 h [34] prior to recording weights. Bulk density ( $\text{g cm}^{-3}$ ) was calculated by dividing the dry weight of the media (g) by the volume of the media ( $\text{cm}^3$ ).

The media water holding capacity (WHC) was calculated separately at the University of Idaho's Soil laboratory. A high-range pressure system with ceramic plates was used to determine the water holding capacity for all five media types at two water potentials:  $-0.033$  MPa (field capacity) and  $-1.5$  MPa (wilting point) [35]. Five samples of each media type were analyzed for field capacity and wilting point. Bulk density and the gravimetric water content of each sample were obtained. Gravimetric soil water content (SWC) was calculated as

$$\text{SWC} = (\text{Db} \times \theta\text{m})/\text{Dw} \quad (1)$$

where  $\text{Db}$  = media bulk density,  $\theta\text{m}$  = gravimetric water content, and  $\text{Dw}$  = water density ( $\text{Dw} = 1 \text{ g cm}^{-3}$ ). Once SWC was determined for each media type, both at field capacity and wilting point, WHC was calculated:

$$\text{WHC} = \text{field capacity SWC} - \text{wilting point SWC} \quad (2)$$

### 2.3. Statistical Analysis

Data were analyzed using SAS software (version 9.4; SAS Institute, Cary, NC, USA) via PROC GLIMMIX. Models included the main effects of growing media and container type, as well as their interaction, with replicate included as a random effect. Where main effects did not interact ( $p > 0.05$ ), the interaction term was omitted from the model. Treatment comparisons were evaluated at  $\alpha = 0.05$ .

## 3. Results

### 3.1. Media Characterization

Bulk density (BD) differed significantly across media types ( $p < 0.0001$ ) but not between container types ( $p = 0.8870$ ) and the main effects did not interact ( $p = 0.0919$ ). BD ranked as follows:  $\text{Pe} < \text{T} = \text{CT} < \text{CG} = \text{CTG}$  (0.131, 0.375, 0.398, 0.545, 0.546  $\text{g cm}^{-3}$ , respectively). Water holding capacity (WHC) varied across growing media types with Pe having the highest water-holding capacity at 62%, followed by T at 32%, CT at 30%, CTG at 16%, and finally CG at 7%.

Analyses revealed that Pe was low in  $\text{NO}_3$ , K, Ca, and Mg but initially provided acceptable levels of P, although this declined to inadequate levels in the absence of fertilizer by the end of the growing season (Table 1). While T initially provided optimal levels of  $\text{NO}_3$  and acceptable levels of P, K, Ca, and Mg, all nutrient levels were inadequate by the end of the growing season. Compost-based media amended with topsoil (CT) or grit (CG) initially provided acceptable levels of  $\text{NO}_3$ , very high levels of P, high levels of K, and low levels of Ca and Mg. Compost-based media amended with both topsoil and grit (CTG) initially provided optimal levels of  $\text{NO}_3$ , very high levels of P, high levels of K, and acceptable levels of Ca and Mg. By the end of the growing season,  $\text{NO}_3$  levels had become inadequate for all compost-based media, while P levels remained high, levels of K were adequate, and Ca and Mg were inadequate.

**Table 1.** Chemical properties of media used to grow Hispaniolan pine seedlings measured before (top row) and after (bottom row) seedling production. Note: C, N, and C:N were assessed before seedling production but not after. Means (SE) are presented ( $n = 5$ ); different letters within a row indicate a significant difference across growing media types at  $\alpha = 0.05$ . EC = electrical conductivity; Pe = peat; T = topsoil; CT = compost–topsoil; CG = compost–grit; CTG = compost–topsoil–grit.

	F Ratio ( <i>p</i> Value)	Pe	T	CT	CG	CTG
pH	69.92 (<0.0001)	5.16 (0.14) b	5.50 (0.06) b	6.98 (0.10) a	7.16 (0.07) a	7.00 (0.16) a
	196.52 (<0.0001)	6.04 (0.06) c	6.70 (0.03) b	7.40 (0.03) a	7.38 (0.06) a	7.44 (0.02) a
EC (mmho cm <sup>-1</sup> )	5.17 (0.0050)	0.10 (<0.01) b	1.22 (0.29) ab	1.05 (0.28) ab	1.17 (0.14) ab	2.11 (0.56) a
	80.38 (<0.0001)	0.12 (<0.01) b	0.16 (<0.01) b	0.59 (0.04) a	0.66 (0.04) a	0.58 (0.02) a
C (%)	65.77 (<0.0001)	29.45 (1.51) a	12.52 (0.55) c	19.25 (0.41) b	18.36 (0.27) b	16.35 (0.42) b
N (%)	523.52 (<0.0001)	0.48 (0.03) d	0.42 (0.01) d	1.75 (0.03) b	1.97 (0.04) a	1.54 (0.05) c
NO <sub>3</sub> <sup>-</sup> (ppm)	3.58 (0.0233)	1.2 (0.20) b	184.0 (14.60) ab	51.6 (28.61) ab	56.0 (14.11) ab	198.0 (97.36) a
	9.97 (0.0001)	1.0 (<0.01) b	1.0 (<0.01) b	9.2 (3.48) a	15.2 (1.62) a	7.2 (1.80) ab
P—H <sub>2</sub> PO <sub>4</sub> <sup>-</sup> (ppm)	13.86 (<0.0001)	3.52 (0.33) b	3.38 (0.30) b	31.86 (6.20) a	37.94 (5.27) a	27.16 (5.39) a
	49.08 (<0.0001)	1.18 (0.02) b	1.64 (0.07) b	16.58 (0.69) a	18.08 (2.00) a	15.54 (1.67) a
K <sup>+</sup> (ppm)	12.47 (<0.0001)	13.0 (1.14) c	82.8 (4.49) bc	300.0 (71.64) ab	351.6 (45.28) a	551.8 (107.68) a
	36.45 (<0.0001)	11.8 (0.20) b	14.6 (0.40) b	141.8 (14.74) a	163.80 (18.11) a	132.0 (14.04) a
Ca <sup>2+</sup> (ppm)	7.48 (0.0007)	3.4 (0.51) c	110.8 (12.50) a	45.6 (9.47) bc	49.4 (2.46) abc	86.8 (29.78) ab
	96.27 (<0.0001)	7.2 (0.80) c	22.4 (0.81) b	42.2 (2.54) a	38.4 (1.36) a	38.4 (1.29) a
Mg <sup>2+</sup> (ppm)	3.80 (0.0187)	2.4 (0.24) b	46.6 (4.77) a	19.4 (5.44) ab	18.2 (1.59) ab	44.6 (20.40) a
	68.23 (<0.0001)	3.8 (0.20) c	7.0 (0.32) b	16.4 (0.98) a	17.2 (0.97) a	17.6 (1.03) a
Na <sup>+</sup> (ppm)	20.60 (<0.0001)	22.4 (0.93) c	295.0 (18.28) a	112.6 (25.49) bc	105.2 (9.00) bc	191.6 (38.76) b
	20.79 (<0.0001)	57.6 (0.93) b	58.8 (0.80) b	85.2 (5.95) a	92.0 (4.49) a	81.6 (1.83) a
C:N	3583.72 (<0.0001)	61.60 (0.23) a	29.74 (0.77) b	11.02 (0.15) c	9.34 (0.05) d	10.60 (0.15) cd

Significant differences were found in chemical properties across growing media types (Table 1). Prior to seedling production, Pe was characterized by low pH, very low EC, high C, low N, NO<sub>3</sub>, P, K, Ca, Mg, and Na, and a very high C:N relative to other media types. T was characterized by low pH, intermediate EC, low C, N, P, and K, and high NO<sub>3</sub>, Ca, Mg, and Na, with an intermediate C:N relative to other media types. CT was characterized by neutral pH, intermediate EC, high P, and intermediate C, N, NO<sub>3</sub>, K, Ca, Mg, and Na, and a low C:N relative to other media types. Similarly, CG was characterized by neutral pH, intermediate EC, high N, P, and K, intermediate C, NO<sub>3</sub>, Ca, Mg, and Na, and a low C:N relative to other media types. Finally, CTG was also characterized by neutral pH, high EC, high NO<sub>3</sub>, P, K, and Mg, intermediate C, N, Ca, and Na, and a low C:N relative to other media types. Trends across media types persisted following seedling production (Table 1). Relative to initial values, pH of all media types increased. For Pe, EC and concentrations of Ca, Mg, and Na increased but for all other growing media types, EC and concentrations of NO<sub>3</sub>, P, K, Ca, Mg, and Na declined following seedling production.

### 3.2. Seedling Morphology

Seedling morphology differed significantly between container types (with the exception of RV) and across growing media types, but the main effects did not interact ( $p \geq 0.2969$ ; Table 2). Seedlings grown in polybags were 10% taller and 11% thicker in RCD compared to seedlings grown in D40s. SDM was 38% greater and RDM was 8% greater for seedlings grown in polybags relative to those grown in D40s. S:R was 47% greater for seedlings grown in polybags compared to those grown in D40 containers.

With regard to growing media, seedling height and SDM were greatest when grown in CG, followed by CTG, CT, and Pe, with the shortest and lightest seedlings grown in T. Similarly, RCD and RDM of seedlings grown in CG were significantly greater than seedlings grown in all other media types. RV was greatest for seedlings grown in CG, followed by those grown in Pe, with seedlings grown in T, CT, and CTG having the smallest RV. S:R was 115% higher for seedlings grown in CTG compared to seedlings grown in T.

**Table 2.** Nursery growth of Hispaniolan pine seedlings across growing media and container treatments. Means (SE) are presented; different letters within a main effect indicate a significant difference at  $\alpha = 0.05$ . HT = height; RCD = root-collar diameter; RV = root volume; RDM = root dry mass; SDM = shoot dry mass; S:R = SDM/RDM; Pe = peat; T = topsoil; CT = compost-topsoil; CG = compost-grit; CTG = compost-topsoil-grit.

		HT (cm)	RCD (mm)	RV (cm <sup>3</sup> )	RDM (g)	SDM (g)	S:R
Container	D40s	3.94 (0.05) b	0.98 (0.01) b	0.91 (0.03)	0.13 (0.01) b	0.13 (0.01) b	1.13 (0.05) b
	Polybags	4.32 (0.07) a	1.09 (0.02) a	0.96 (0.04)	0.14 (0.01) a	0.18 (0.01) a	1.58 (0.07) a
Media	Pe	3.80 (0.07) bc	0.93 (0.02) b	1.03 (0.04) b	0.09 (0.01) b	0.09 (0.01) bc	1.32 (0.10) ab
	T	3.68 (0.07) c	0.91 (0.02) b	0.72 (0.03) c	0.11 (0.01) b	0.08 (0.01) c	0.83 (0.04) b
	CT	4.04 (0.10) b	0.94 (0.03) b	0.72 (0.04) c	0.11 (0.01) b	0.14 (0.01) b	1.57 (0.08) ab
	CG	4.92 (0.08) a	1.40 (0.03) a	1.56 (0.05) a	0.27 (0.01) a	0.33 (0.01) a	1.35 (0.04) ab
	CTG	4.24 (0.11) b	1.02 (0.03) b	0.61 (0.04) c	0.12 (0.01) b	0.15 (0.01) b	1.72 (0.15) a
Type III tests of fixed effects		F ratio ( <i>p</i> value)					
Container	1/40 <sup>1</sup>	24.30 (<0.0001)	26.19 (<0.0001)	3.13 (0.0846)	4.71 (0.0360)	21.43 (<0.0001)	8.87 (0.0049)
Media	4/40	28.29 (<0.0001)	48.99 (<0.0001)	30.84 (<0.0001)	48.26 (<0.0001)	46.91 (<0.0001)	3.24 (0.0215)

<sup>1</sup> degrees of freedom.

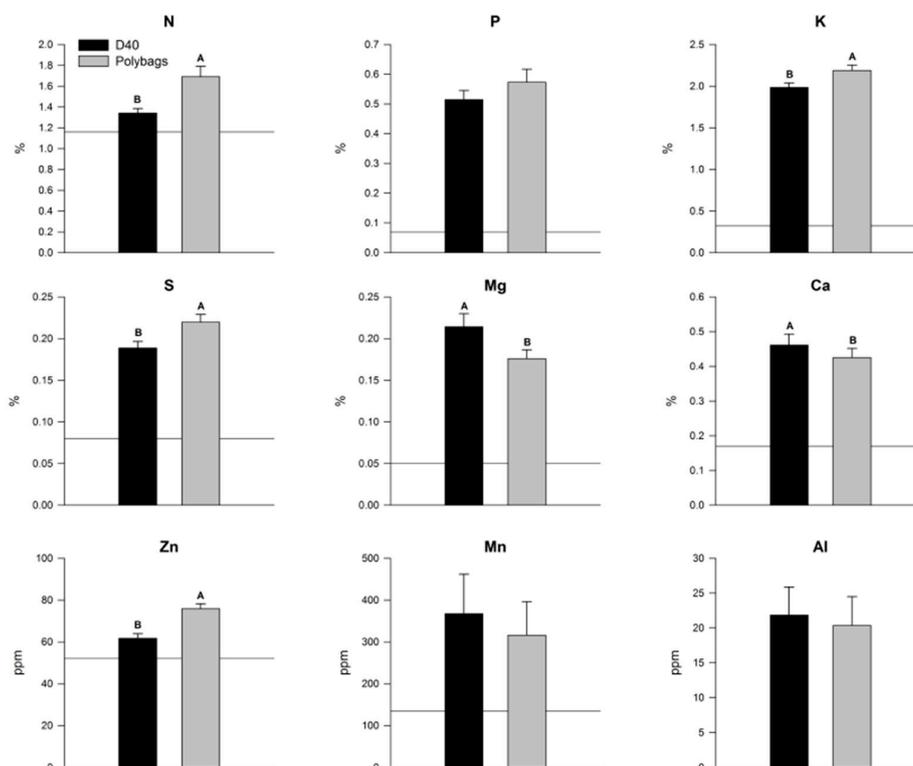
### 3.3. Foliar Chemistry

Plant tissue analyses revealed significant differences in the levels of all nutrients examined among growing media types, and in levels of most nutrients examined between container types, with significant interactions between the two main effects for Na, B, Fe, and Cu (Table 3 and Figures 1–3). Seedlings grown in polybags yielded significantly greater foliar concentrations of N, K, S and Zn and lower foliar concentrations of Mg and Ca relative to those grown in D40s, with no significant difference in foliar P, Mn, or Al concentrations between container types (Figure 1).

**Table 3.** Effects of growing media and container type on Hispaniolan pine seedling foliar nutrient concentrations.

	Media F Ratio ( <i>p</i> Value)	Container F Ratio ( <i>p</i> Value)	Media   Container F Ratio ( <i>p</i> Value)
N (%)	5.06 (0.0019)	14.57 (0.0004)	ns *
P (%)	16.12 (<0.0001)	2.70 (0.1076)	ns
K (%)	9.90 (<0.0001)	9.53 (0.0035)	ns
S (%)	14.42 (<0.0001)	14.64 (0.0004)	ns
Mg (%)	26.40 (<0.0001)	14.78 (0.0004)	ns
Ca (%)	45.12 (<0.0001)	5.61 (0.0223)	ns
Na (%)	35.69 (<0.0001)	0.70 (0.4081)	6.37 (0.0005)
B (ppm)	24.02 (<0.0001)	10.46 (0.0025)	3.79 (0.0106)
Zn (ppm)	6.08 (0.0006)	28.17 (<0.0001)	ns
Mn (ppm)	210.98 (<0.0001)	2.19 (0.1457)	ns
Fe (ppm)	37.42 (<0.0001)	7.41 (0.0096)	5.36 (0.0015)
Cu (ppm)	20.55 (<0.0001)	19.04 (<0.0001)	2.82 (0.0377)
Al (ppm)	19.85 (<0.0001)	0.20 (0.6607)	ns

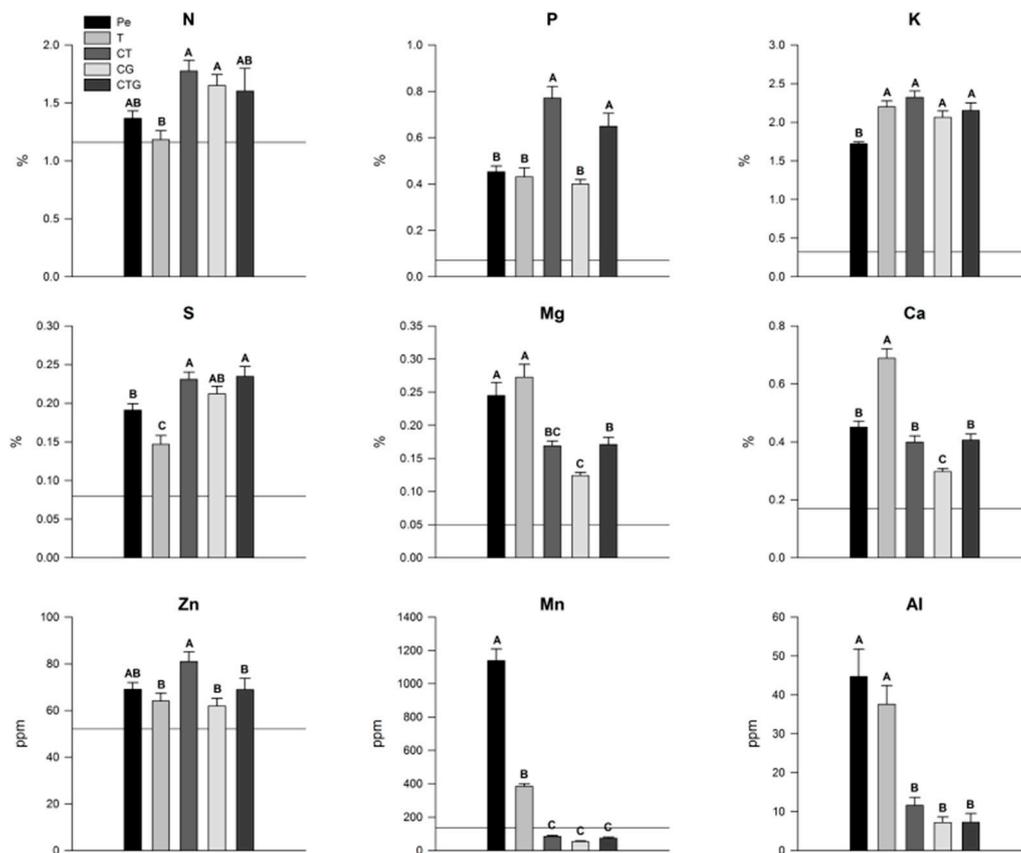
\* ns = interaction term was not significant at  $\alpha = 0.05$  and therefore dropped from the model.



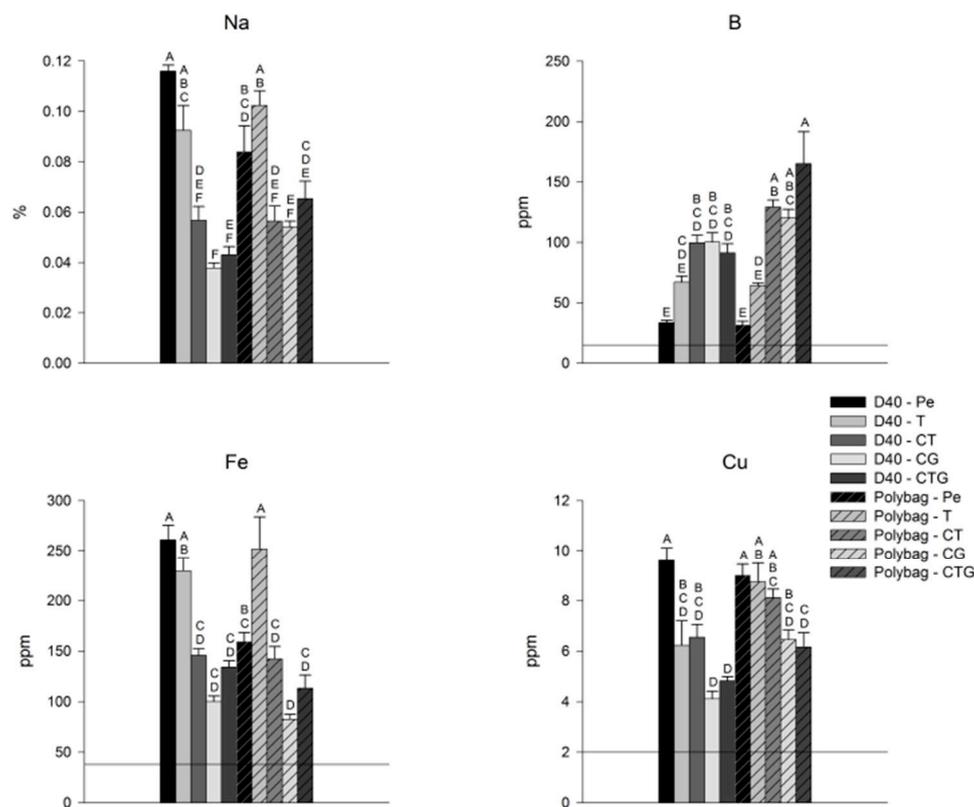
**Figure 1.** Foliar nutrient concentrations of Hispaniolan pine seedlings between container types. Horizontal lines indicate normal nutrient levels reported for *Pinus strobus* L. from <http://agsci.psu.edu/aasl/plant-analysis/plant-tissue-total-analysis/interpretive-nutrient-levels-for-plant-analysis/pine-white> (accessed 2 February 2018).

With regard to growing media (Figure 2), foliar N was greater in seedlings grown in CT and CG relative to T. Foliar P was greater in seedlings grown in CT and CTG relative to Pe, T, and CG. Foliar K was greater in seedlings grown in T, CT, CG, and CTG relative to Pe. Foliar S was greatest in seedlings grown in CT and CTG, followed by CG, then Pe, and finally T. Foliar Mg was greatest in seedlings grown in T and Pe, followed by those grown in CT and CTG, and finally those grown in CG. Foliar Ca was greater in seedlings grown in T, followed by those grown in Pe, CT, and CTG, and finally those grown in CG. Foliar Mn was greater in seedlings grown in Pe, followed by those grown in T, with those grown in CT, CG, and CTG yielding the lowest levels of Mn. Foliar Al was greater in seedlings grown in Pe and T relative to those grown in CT, CG, and CTG.

The main effects of growing media and container type interacted to impact foliar concentrations of Na, B, Fe, and Cu (Figure 3). Foliar Na and foliar Fe were greatest among seedlings grown in Pe in D40s or in T in either D40s or polybags. Foliar B was greatest among seedlings grown in polybags containing compost-amended growing media (CT, CG, and CTG). Foliar Cu was greatest among seedlings grown in Pe (in either D40s or polybags) and in polybags containing T or CT.



**Figure 2.** Foliar nutrient concentrations of Hispaniolan pine seedlings among growing media treatments. Pe = peat; T = topsoil; CT = compost-topsoil; CG = compost-grit; CTG = compost-topsoil-grit. Horizontal lines indicate normal nutrient levels reported for *Pinus strobus* from <http://agsci.psu.edu/aasl/plant-analysis/plant-tissue-total-analysis/interpretive-nutrient-levels-for-plant-analysis/pine-white> (accessed 2 February 2018).



**Figure 3.** Foliar nutrient concentrations of Hispaniolan pine seedlings across interacting container and growing media treatment levels where significant ( $\alpha = 0.05$ ). Horizontal lines indicate normal nutrient levels reported for *Pinus strobus* from <http://agsci.psu.edu/aasl/plant-analysis/plant-tissue-total-analysis/interpretive-nutrient-levels-for-plant-analysis/pine-white> (accessed 2 February 2018).

#### 4. Discussion

The growing media and container type did not interact to affect seedling morphology, but each independently influenced seedling growth, providing evidence that these nursery cultural practices can be used to cultivate *P. occidentalis* seedlings suited to particular outplanting conditions. It is well documented that seedling morphology and early outplanting performance can differ among container types [36,37], but it remains unknown exactly why Hispaniolan pine seedling growth differed between polybags and D40s. Although the diameter of both container types was similar, seedlings grown in polybags had greater shoot and root growth, but also higher S:R. Despite the known growth and development issues associated with the use of polybags [16], Hispaniolan pine seedling morphology and foliar chemistry indicate that this container is a suitable choice for the species. This is promising given that polybags are currently widely used, and a conversion to rigid plastic containers would likely be impractical as it would require a concomitant shift in the entire nursery system, from the use of uniform, artificial growing media and the consequent need for fertilization and frequent irrigation to the need for raised benches to promote root pruning [16].

One of the main issues with the use of polybags for the production of tree seedlings relates to poor root growth, particularly when seedlings are held in their containers for too long [16]. The higher S:R suggests that the rate of root growth relative to shoot growth was lower for seedlings grown in polybags compared to those grown in D40s, perhaps indicating that root growth was constrained in polybags by the end of the 22-week growing season. Moreover, research suggests that seedlings with low S:R tend to have increased survival rates when outplanted to harsh sites, such as those in Haiti, because of the increased uptake of water and nutrients afforded by the larger root system relative to lower demands made by the smaller shoot [38]. Thus, even though seedlings grown in D40s were smaller

overall, the S:R was potentially more favorable for seedlings that are likely to encounter periodic dry conditions following outplanting. Further study into how specific characteristics of different container types influence growth of seedlings of this species, and how this relates to outplanting performance, is warranted.

With regard to growing media, differences in chemical and physical properties among types were likely the primary drivers for the noted differences in seedling response variables. Soil bulk density and water holding capacity influence the amount of water available for plant uptake [39] as well as fine root proliferation. Soils that are highly compressed tend to lack the pore spaces necessary for holding water and air. Thus, limited pore space may lead to stunted growth [40]. Field-sourced soils can be highly variable in their physical characteristics and tropical soils in particular can contain high amounts of clay and silt, which have lower macropore space that may limit gas exchange, water drainage, and potentially plant-available water, since clays hold more water at high tension [17]. In some instances, packing topsoil in containers and the settling of the soils in the container may contribute to higher bulk densities, thereby making rooting a problem [17]. None of the growing media types had bulk densities beyond that of root penetration, which is approximately  $1.5 \text{ g cm}^{-3}$  [41], so bulk density likely did not inhibit root penetration. Studies have shown that as bulk density increases, available water holding capacity decreases [42]. Media water holding capacity also varied across treatments, with an 8-fold difference between CG and Pe (7% and 62%, respectively). Media types can be selected and designed to have higher water holding capacities and provide increased moisture availability [43]; however, depending on soil texture and other properties, additional factors such as oxygen availability may be affected [30].

Nonetheless, early seedling growth was not better in Pe but rather in CG, which may be explained by media chemistry. Although seedlings likely had adequate foliar nutrient levels across all growing media, based on recommended nutrient levels for *Pinus strobus* L. (<http://agsci.psu.edu/aasl/plant-analysis/plant-tissue-total-analysis/interpretive-nutrient-levels-for-plant-analysis/pine-white>), both Pe and T were low in %N, and seedlings grown using these media types likely bordered on nitrogen deficient. While T may have had a relatively high amount of  $\text{NO}_3$  initially (most likely due to the manure and bark mulch; Table 1), we suspect a large fraction of it leached out before the roots could exploit the full capacity of the container. Nitrogen is a macronutrient which is essential to all plant physiological processes, and a lack of access to this nutrient may have contributed to the stunted growth of these seedlings. While amending T with compost improved growing conditions, it was when seedlings were grown in a combination of compost and grit (CG) that better growth was observed. CG was characterized by neutral pH, higher levels of N, P, and K, and lower C:N compared to Pe which had a slightly acidic pH, lower levels of N, P, and K, and very high C:N. Yet, seedlings grown in compost-based growing media (CT, CG, and CTG) showed low levels of foliar manganese, which is essential for the synthesis of chlorophyll and also serves as an enzyme activator [44]. Manganese absorption may have been inhibited by pH [45] and high concentrations of iron, calcium and aluminum in compost-based media types [46] (Figure 2). Similar results have been found for *Pinus sylvestris* seedling tissue nutrient concentrations grown in compost [47].

## 5. Conclusions

Improvements can be made to existing nursery practices in Haiti to enhance the production of *P. occidentalis* seedlings, particularly if viewed through the framework of the Target Plant Concept [4]. Compost-based growing media, particularly when amended with forestry grit or similar coarse-grained sand, show promise for early seedling growth. Despite low water holding capacity and limited manganese, this growing medium provided adequate levels of essential mineral nutrients (particularly nitrogen). Seedling morphology and foliar chemistry indicate that polybags remain a suitable choice for the species, although higher S:R among seedlings grown in polybags relative to D40s suggest that further work is needed to determine the specific container characteristics that are optimal (i.e., meet the target specifications for a given objective) for the production of *P. occidentalis* seedlings for reforestation

and restoration in Haiti. Given that the seedlings in this study were relatively small, an examination of the nutritional needs of *P. occidentalis* as well as a field component to determine what are the most important seedling attributes that influence post-planting establishment is needed to advance reforestation success in Haiti.

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