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Emergence, growth, and quality of *Calycophyllum spruceanum* plants produced in different containers and substrates

Wilson Francisco Guerra-Arévalo[1] | **José Roy Cercado-Delgado**[2] | **Héctor Francisco Espinoza-García**[3] | **Tatiana Mildred Ucañay-Ayllon**[4] | **Diego Gonzalo García-Soria**[1] | **Carlos Abanto-Rodríguez**[1] | **Dennis del Castillo-Torres**[1] | **Luis Ernesto Freitas-Alvarado**[1] | **Rossana Díaz-Soria**[1] | **Héctor Guerra-Arévalo**[1]

[1]Departamento Loreto, Instituto de Investigaciones de la Amazonía Peruana (IIAP), Iquitos, Peru

[2]Departamento Cajamarca, Universidad Nacional Autónoma de Chota (UNACH), Chota, Peru

[3]Departamento Guatemala, Ministerio de Ambiente y Recursos Naturales, Ciudad de Guatemala, Guatemala

[4]Departamento Ucayali, Instituto Nacional de Innovación Agraria (INIA), Pucallpa, Peru

Correspondence
Wilson Francisco Guerra-Arévalo, Departamento Loreto, Instituto de Investigaciones de la Amazonía Peruana (IIAP), Iquitos, Loreto, Peru.
Email: wguerra@iiap.gob.pe

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Abstract

The growing demand for *Calycophyllum spruceanum* is causing strong pressure on natural populations due to anthropogenic activities. For this reason, it is urgent to develop propagation technologies and production of plants for reforestation activities and establishment of forest plantations for their use and conservation. The objective of this study was to determine the effect of different substrates and containers on the emergence and growth of *C. spruceanum*. For this purpose, two experiments were conducted. In the first, four substrates were tested to evaluate seedling emergence, while in the second, the growth and quality of plants in different containers and substrates were determined. The maximum emergence values of 62.3%, emergency speed index (ESI) of 2.2, and mean emergence time of 29.9 were determined using a combination of carbonized rice husk (CRH), chicken manure, and decomposed sawdust in a 1:1:1 ratio. The combination of the plastic bag container with CRH and poultry manure in a 1:1 ratio. Substrate gave the best results for total dry biomass (27.40), lignification index (0.19), robustness index (5.56), and Dickson's quality index (3.26). Therefore, the use of CRH, poultry manure, and decomposed sawdust in a 1:1:1 ratio is recommended for seedling emergence. Similarly, the use of a plastic bag-type container and the substrate CRH and chicken manure in a 1:1 ratio is recommended for the production of *C. spruceanum* plants.

1 | INTRODUCTION

Calycophyllum spruceanum is known as capirona, mulateiro, pau mulato, and capirona negra, and belongs to the Rubiaceae family (Martins, 2018). It is dispersed along the Amazon River and tributaries, where plants are mainly established in clayey, fertile, and floodable soils (Ugarte & Domínguez, 2010). It is distributed in the Amazon

Abbreviations: BAP, biomass of the aerial part; BRP, biomass of the root part; CRH, carbonized rice husk; DQI, Dickson quality index; EVI, emergence velocity index; ESI, emergency speed index; LI, lignification index; MET, mean emergence time; RI, robustness index; TDB, total dry biomass.

basin of Peru, Bolivia, and Brazil below 1200 m.a.s.l. in ecosystems with high and constant rainfall. In Ucayali (Peru), it thrives on the banks of the Aguaytía and Ucayali rivers, predominantly on alluvial and temporarily flooded soils (Loyola, 2019).

Calycophyllum spruceanum reaches heights of up to 35 m, and its DBH ranges from 50 to 120 cm. It has a regular cylindrical shaft, glossy, homogeneous green color, and smooth outer bark (Reynel et al., 2004). Its wood is used for flooring, doors, tongue and groove joints, furniture, heavy structures, beams, tool handles, posts, and charcoal, among others (Ugarte & Domínguez, 2010).

For this reason, due to its commercial importance in Loreto and Ucayali, around 84,000 m^3 $year^{-1}$ and 1700 m^3 $year^{-1}$ of roundwood were extracted, respectively (Pacheco & Vásquez, 2016).

In this sense, their growing demand is causing strong pressure on *C. spruceanum* capirona, mainly due to different anthropogenic activities. Therefore, the recovery and enrichment of these ecosystems through reforestation activities and the establishment of forest plantations are urgent. However, for success in these activities, factors such as the origin of the seeds, plant quality, germination and emergence of the seedlings, sowing time, and transport, among others, must be taken into account. These factors determine the success of a forest plantation (Montoya & Cámara, 1996). In this regard, reforestation initiatives were carried out in Ucayali, such as the SHESHEA-ARAU 2019 reforestation project; however, many of these areas did prosper. Similar accounts were reported by Luis et al. (2004), mentioning that in Mexico, only 40% of the reforested areas had acceptable levels of quality and survival.

In relation to emergence, Romero (1989) mentions that this occurs after the seed has germinated, meaning it has completed its imbibition stage and emitted the radicle through the seed coat. Subsequently, the hypocotyl elongates, and the seedling appear on the soil or substrate. C*alycophyllum spruceanum*, germination is epigeal. The hypocotyl elongates and curves upward, causing the cotyledons to emerge from the soil, the integument to detach, and the plant stem to from along with the first leaves (Floriano, 2004).

Meanwhile, Moraes et al. (2007) mention that seedling emergence is sometimes limited by various factors such as texture, structure, aeration, and water retention in the substrate. Therefore, the quality of this input is among the most relevant aspects affecting the growth and development of seedlings from germination through emergence and growth (Dutra et al., 2012). Caldeira et al. (2012) indicate that the presence of organic matter (OM) in the substrate enhances seed germination and seedling emergence, due to the physical, biological, and chemical properties it provides.

Additionally, Caldeira et al. (2012) emphasize that, in plant production, apart from the substrate, the container's role should be considered, as it can influence the roots system's development during the plant's growth phase in the nursery. Regarding plant quality of the plants, Ortega et al. (2006) highlight its correlation with the possibility of survival after transplanting into the final field. Thus, reforesting with high-quality plants can mitigate effects of climatic factors.

A plant is considered of good quality when there is a balanced growth or organs (roots, stems, and leaves). Therefore, the success of plants in the final field largely depends on the root system´s quality to absorb nutrients and water from the soil and the aerial part's ability to utilize light energy through chlorophyll pigments and uptake carbon dioxide (CO_2) during photosynthesis (Devlin, 1980). In this context, the objective of this work was to determine the effect of different substrates and containers on the emergence and growth of *C. spruceanum* plants in Ucayali, Peru.

Core Ideas

- Availability of organic substrates for the production of *Calycophyllum spruceanum* plants.
- Arrangement of the best combination of organic substrates and suitable container to produce quality plants.
- Environmental decontamination occurs through the use of residues from the rice industry, poultry farming, and wood sawing.
- Generation of low-cost technology for the production of native species for Amazonian riverside farmers.

2 | MATERIALS AND METHODS

Two experiments were conducted. The first aimed to assess the influence of various substrates on seedling emergence, while the second focused on evaluating the effect of different containers and substrates on the growth of *C. spruceanum* plants.

2.1 | Experimental site

The studies were carried out at the Instituto de Investigaciones de la Amazonía Peruana (IIAP), located at 12,400 km of the Federico Basadre highways, Yarinacocha district, Coronel Portillo province. The site's coordinates are latitude 8°22′31″ south and longitude 74°34′35″ west, with an altitude of 154 m above sea level. This region features a tropical rainforest climate characterized by a type B(r)A climate, known for its abundant rainfall warmth. The recorded temperatures include maximum, average, and minimum values of 33˚C, 26˚C, and 18.7˚C, respectively. Annual rainfall and relative humidity are reported as 2090 mm and 91.3%, respectively (SENAMHI, 2022).

2.2 | Seed production of *C. spruceanum*

Seed was manually harvested from four seed trees at ICRAF (International Centre for Agroforestry Research). These trees are currently under the custody of the Association of Forest Producers PROSEMA, located at 98 km of the Federico Basadre road, 100 m to the right margin. The selected trees exhibited similar characteristics in terms of vigor, crown, height, and the same phenological stage of dissemination. Seeds were selected based on their size, color, and phytosanitary condition to standardize the material and prevent potential effects that could introduce the experimental errors.

2.3 | Procurement of substrates

In the first study, four substrates were used: (1) fine river sand (control), (2) alluvial soil, (3) decomposed sawdust, and (4) carbonized rice husk (CRH) [charred rice husk] + chicken manure + decomposed sawdust (1:1:1). In the second study, three substrates were used: (1) control substrate: agricultural soil + fine sand (1:1), (2) CRH + chicken manure (1:1), and (3) CRH + chicken manure + sawdust (2:1:1).

Decomposed poultry manure was purchased from local suppliers specialized in organic fertilizers. Similarly, decomposed sawdust was purchased from local sawmills and alluvial soil was obtained from the *C. spruceanum* capironales of the Panaillo hamlet, Yarinacocha district, Ucayali region. Before collection, the leaf litter was separated from the surface, and then the soil from the first 10 cm of depth was obtained with a straight shovel. Additionally, fine sand was collected from the banks of the Ucayali River. The rice husks were carbonized in a cone burner at the IIAP Ucayali headquarters.

Prior to mixing the substrates, each input was sieved using a 0.5-mm sieve to remove impurities (stones, leaves, pieces of wood, large clods of earth, etc.). They were disinfected and sterilized with steam, using metal cylinders heated with a wood fire for a period of 2 h. Following this, the substrates were disinfected with CUPRAVIT to eliminate fungi and bacteria.

After obtaining the different substrates for experiments 1 and 2, a 1 kg sample was obtained for physicochemical analysis. These samples were packed, coded, and sent to the soil laboratory of the National Agrarian University La Molina-NAUM for respective analysis. Regarding the containers used in the second experiment, the tubes had a capacity of 180 cm^3, and the plastic bags had the following dimensions: 4″ × 7″ × 2 mm. Both containers were purchased from local nursery companies.

2.4 | Experimental design

Experiment 1: It was conducted using a completely randomized design (CRD) with four treatments (T1-fine river sand [control], T2-alluvial soil, T3-decomposed sawdust, and T4-CRH + chicken manure + decomposed sawdust [1:1:1]) distributed in three replications with 100 seeds per experimental plot. The variables evaluated from the third day after seed sowing were: emergence percentage (Equation 1), determined by the ratio between the number of seedlings that emerged and the number of seeds sown multiplied by 100.

$$\mathrm{E} = \frac{\text{Number of seedlings emerged at last count}}{\text{Number of seeds sown}} \times 100 \quad (1)$$

where E is the percentage of seedling emergence.

The emergency speed index (ESI) was also evaluated (Equation 2), for which normal seedlings were counted daily and divided by the number of days.

$$ESI = \frac{N1}{d1} + \frac{N2}{d2} + \frac{N3}{d3} \cdots\cdots\cdots \frac{Nn}{dn} \quad (2)$$

where Nn is the number of seedlings emerged from the first to the last count, dn is the number of days after sowing, from the first to the last count.

In addition, the mean emergence time (MET) was determined (Equation 3) through the ratio of daily counts and emerged plants to the total number of emerged seedlings.

$$\mathrm{MET} = \frac{g1ni + g2nii \ldots gn}{N1 + N2 \ldots Gn} \quad (3)$$

where $g1$, $g2 \ldots gn$ is the number of days from sowing to the respective count, and $n1 + n2 \ldots Gn$ is the number of seedlings emerged at times $g1$, $g2 \ldots gn$.

Experiment 2: It was conducted using a CRD in a factorial scheme (2C × 3S), where the R factor compromised containers with two levels: (1) polyethylene plastic bags with a capacity of 7″ × 4″ × 2 mm, and (2) tubes with a capacity of 180 cm^3, and the S factor includes substrates with three levels: (1) control (agricultural soil + sand in a 1:1 ratio), (2) CRH + chicken manure 1:1 ratio, and (3) CRH + chicken manure + decomposed sawdust 2:1:1 ratio. The experiment was distributed in four replicates, each containing 20 seedlings per experimental plot. The evaluated variables were as follows: plant height (H, cm), basal diameter (DB, mm), number of leaves (NL), and critical chlorophyll index (CCI). Plant height (H) was measured from the substrate level to the apex of the last leaf using a millimeter ruler. DB was measured with a digital vernier 1.0 cm above the substrate level. Then NL was determined through simple counting. Chlorophyll content was determined with a portable chlorophyll meter SPAD-502 from the second month after replanting, for which five leaves from the middle part of 10 plants per replicate were evaluated.

At 90 days after planting, a sample of 10 plants from each repotting and treatment was collected. The substrate was washed away from the roots under running water, and the aerial part's were separated from the root system with a cloth scissors previously disinfected with 90° alcohol. Both parts were coded and placed separately in paper envelopes. Subsequently, the samples were placed in a forced circulation air oven at 60°C until a constant dry biomass was obtained. After 3 days, the samples were removed from the oven and weighed to obtain the biomass of the aerial part (BAP) and biomass of the root part (BRP), thus the total dry biomass (TDB, g) was obtained. Using the data obtained before and after drying the samples, the following indices were calculated: the lignification index (LI = TDB/TWB [total wet biomass]), the robustness index (RI = H/BD), and the Dickson quality index (DQI = (TDB/H/BD + BAP/BRP).

2.5 | Data analysis

The data were tabulated in Microsoft Excel, and then assessed for the assumptions of normality and homogeneity of variances using the Shapiro Wilk and Barlett methods, respectively. Since the data met the criteria of normality and homogeneity an analysis of variance (ANOVA) was conducted using the *F*-test, and the treatment means were compared using Tukey's 5% probability test. Statistical analyses were performed using RStudio Team and Info stat.

3 | RESULTS AND DISCUSSION

3.1 | Experiment 1

According to the ANOVA, the different substrates resulted in significant statistical differences ($p \leq 0.05$) in the emergence percentage at 33 days after sowing and MET. Specifically, the treatments using river sand (T1) and CRH + chicken manure + decomposed sawdust (1:1:1) (T4) yielded the most favorable results, with an emergence of 61.3% and an ESI of 2.2 for *C. spruceanum* seedlings. On the other hand, the lowest emergence of 2.3% and an ESI of 0.1 were observed with the alluvial soil substrate (T2) (Table 1).

In relation to the MET, it was observed that the highest number of seedlings emerged at 33 days with the alluvial soil, indicating a delay. In contrast, when utilizing the CRH + chicken manure + decomposed sawdust (1:1:1) substrate, emergence occurred in fewer days (29.9–30 days). Therefore, a lower MET suggests a higher ESI (Table 1).

TABLE 1 Mean test for percentage emergence (%E), emergency speed index (ESI), and mean emergence time (MET) of *Calycophyllum spruceanum* seedlings by effect of different substrates in conventional nursery.

Substrates	%E	ESI	MET
(T2) Alluvial soil	2.3c	0.1d	33.0a
(T3) Decomposed sawdust	37.7b	1.3c	31.4ab
(T1) River sand	60.3a	2.0b	31.2ab
(T4) CRH + chicken manure + decomposed sawdust (1:1:1)	62.3a	2.2a	29.2b
Average	40.6	1.4	31.4

Note: Equal means in the column do not differ statistically according to Tukey's test at 5% probability.
Abbreviation: CRH, carbonized rice husk.

Similar results of 78.0% were reported by Onofre (2011) for *C. spruceanum* seedling emergence. Conversely, Nogueira et al. (2003) found lower emergence rates below 0.4% while studying *Hetaeria oblongifolia* Huber on sand substrate. Leal et al. (2020) reported emergence rates from 1.7% to 0.5% in *Amburana cearensis* using humus and coconut fiber substrates, respectively.

In this study, it is evident that the "alluvial soil" substrate was ineffective for the emergence, ESI, and MET of *C. spruceanum* seedlings. Contributing factors included particle cohesion and slight plastic due to the substrate's clay content (26%), and OM (9.71%). These characteristics led to substrate compaction observed through surface cracking, acting as a physical barrier to seeding emergence during the 44-day evaluation period.

Additionally, it was observed that the irrigation water did not permeate the substrate, causing waterlogging on the surface (Figure 1).

In this regard, Rego et al. (2007) mention that it is necessary for the substrates to have adequate humidity conditions for seed hydration and rehydration in order for germination and emergence to occur. Therefore, to obtain adequate IVE values, it is best to use substrates with a low percentage of clay to avoid compaction, since these substrates are less porous, less permeable, and have low infiltration (Park et al., 2011).

In relation to the treatment with decomposed sawdust (T3), the results were not very satisfactory due to its high moisture storage capacity. Oliveira et al. (2007) affirm that substrates with high humidity negatively affect the emergence of seedlings due to the absence of oxygen and the absence of adequate temperatures for the germination process; therefore, sawdust should be mixed with CRH and sand, among other organic materials to favor seedlings emergence (Peñuelas, 2008). In this study, the emergence of *C. spruceanum* seeds with substrates whose sawdust content exceeded 50% presented unsatisfactory results in emergence due to the fact

FIGURE 1 Emergence of plants *Calycophyllum spruceanum* from in substrate (A) soil alluvial (T2) and (B) substrate with organic matter (T4).

that it retains excess moisture. Therefore, Hernández et al. (2014) propose that 30% and 50% sawdust should be used in a substrate, or in any case avoid its use and look for other alternatives.

The MET in this study indicates that the emergence of the seedlings occurred in a prolonged time, indicating that the seeds of this species have some physical dormancy or tegument, in this sense, it is recommended to carry out studies to reduce the MET as pre-germinative treatments or subject them to different environments and temperatures. Since the optimal temperature is that where maximum germination occurs in the shortest time (Mayer & Poljakoff, 1989).

In this regard, Martins (2018) recommends that the emergence of *C. spruceanum* seedlings should be in controlled environments where the temperature and relative humidity are constant. In this study, these characteristics were not controlled, and it was carried out at ambient temperature and relative humidity; therefore, there were variations from 22˚C to 36˚C and from 54% to 84%, respectively. Thompson et al. (1977) indicate that diurnal temperature fluctuations can initiate or accelerate seed germination of many species.

3.2 | Experiment 2

After replanting, substrates, containers, and the interaction of these factors caused significant effects ($p \leq 0.01$) on height (*H*, cm), DB, mm, and TDB (g); on the other hand, the factor containers and the interaction did not produce significant effects on NL and CCI.

In general, it was determined that the height (*H*, cm) and DB of the plants produced in bags were statistically superior to the plants produced in a tube in all substrates (Table 4). However, the plants that were treated with the substrate: CRH + chicken manure (1:1) and in bags presented the best results of 25.75 cm and 4.72 mm in height and DB, respectively, being statistically superior to the other treatments. Meanwhile, the lowest results of 10.34 cm and 3.15 mm were verified in the bag and in control substrate: agricultural soil + fine sand (1:1) (Table 2).

The plants produced in tubes had the same growth trend, but with lower values; thus, both in CRH + chicken manure (1:1) and in the control substrate-agricultural soil + fine sand (1:1), they reached a basal height and diameter of 19.25 cm and 8.12 cm, and 3.92 mm and 2.43 mm, respectively, at 120 days after planting (Table 2).

In this study, plant height was higher than the results of 21.9 cm reported by Loyola (2019). In short, the substrate CRH + chicken manure (1:1) and the bag container type were the most efficient for the growth of *C. sppruceanum* plants at 120 days after planting. In relation to the control substrate—agricultural land + fine sand (1:1) in both containers, it was observed that plant growth in height and DB of *C. spruceanum* was statistically inferior to all treatments. In this regard, Carvalho Filho et al. (2003) indicate that the type of substrate and the size of the container are the first aspects that should be considered to guarantee the production of good-quality plants.

The substrate CRH + chicken manure + sawdust (2:1:1) caused intermediate results due to its composition; thus, the presence of chicken manure influenced plant growth, but apparently the amount was not sufficient, since a greater amount of CRH was added instead of OM. Gonçalves et al. (2005) state that a substrate should have a lower proportion of CRH with values ranging between 20% and 30%, while the organic material should range between 70% and 80% in the final composition of the substrate.

The plants produced with the substrate composed of CRH + chicken manure (1:1) are suitable for the final field and showed adequate growth in height and DB. Gomes et al. (2002) mention that plants for final field should have 25–35 cm height and 5–10 mm DB, respectively. On the other hand, Sáenz et al. (2014) point out that plants should be of 12–15 cm height and 2.5–4.9 mm DB, respectively. Therefore, plants produced from *C. spruceanum* with these dimensions will have a greater capacity for survival because they are good indicators of survival and growth in the final field.

The plants produced in the substrate CRH + chicken manure (1:1) emitted the greater NH (14.7); on the other hand, the plants produced in the substrate: agricultural land + sand

TABLE 2 Tukey's statistical test for plant height and basal diameter of *Calycophyllum spruceanum* plant by effect of different containers and substrates in a conventional nursery.

	Container			
	Plant height (*H*, cm)		Basal diameter (BD, mm)	
Substrates	Bag	Tube	Bag	Tube
Control: agricultural soil + fine sand (1:1)	10.3Ac	8.1Bc	3.2Ab	2.4Bc
CRH + chicken manure + sawdust (2:1:1)	16.4Ab	12.4Bb	3.3Ab	3.0Bb
CRH + chicken manure (1:1)	25.8Aa	19.3Ba	4.7Aa	3.9Ba
Average	17.5	13.3	3.7	3.1

Note: Equal lowercase letters in the column and uppercase letters in the line do not significant statistical difference, according to the Tukey's test at 1% probability.
Abbreviation: CRH, carbonized rice husk.

FIGURE 2 *Calycophyllum spruceanum* plants were produced in (A) control substrate and in tube (T1) and in (B) carbonized rice husk (CRH) + chicken manure in bag (T3).

TABLE 3 Tukey's statistical test for the number of leaves (NL) of *Calycophyllum spruceanum* plants by simple effects of substrates and containers in conventional nursery.

Substrates	Media
Control: agricultural soil + fine sand (1:1) (T1)	12.4c
CRH + chicken manure + sawdust (2:1:1) (T2)	14.2b
CRH + chicken manure (1:1) (T3)	14.7a
Average	14.48
Containers	
Tube	13.4b
Bag	14.3a
Average	13.8

Note: Equal letters in the column do not show statistically significant differences according to Tukey's test at 5% probability.
Abbreviation: CRH, carbonized rice husk.

TABLE 4 Tukey's statistical test for critical chlorophyll index (CCI) of *Calycophyllum spruceanum* plants by effect of different containers and substrates in conventional nursery.

Substrates	Media
Control: agricultural soil + fine sand (1:1) (T1)	24.4b
CRH + chicken manure + sawdust (2:1:1) (T2)	29.9a
CRH + chicken manure (1:1) (T3)	30.4a
Average	28.4

Note: Equal letters in the column do not show statistically significant differences according to Tukey's test at 1% probability
Abbreviation: CRH, carbonized rice husk.

(1:1) presented the lowest results. of 12.4 leaves at 90 days after planting (Table 3). When the containers were analyzed, it was found that the plants produced in bags emitted a greater NL compared to the tube (Table 3; Figure 2).

The greater or lesser emission of the NH in plants is very important because it indicates the efficiency of the substrates with which they are produced; on the other hand, leaves are considered the organs that best represent the nutritional status of the plants as a result of the availability of macro- and micronutrients in the substrates (Floss, 2006).

Table 4 shows the Tukey's test ($p \leq 0.01$) for the CCI. It is observed that the substrates composed of CRH + chicken manure (1:1) and CRH + chicken manure + sawdust (2:1:1) statistically showed the same results of 30.15 CCI. Meanwhile, they were statistically superior to the values of the plants produced in the control substrate: agricultural land + fine sand (1:1), which on average reached 24.43 CCI at 90 days after planting.

According to Yang et al. (2015), the analysis of chlorophyll content allows monitoring plant development and thereby

TABLE 5 Chemical analysis of the substrates used in experiment 2.

Treatments	pH (1:1)	EC (dS/m)	M.O (%)	N (%)	P (%)	K (%)	CaO (%)
River sand + agricultural soil (1:1) (T1)	6.8	0.19	0.97	0.07	0.007	0.007	1.43
CRH + chicken manure + sawdust (2:1:1) (T2)	5.3	0.98	28.6	1.03	1.07	0.33	1.95
CRH + chicken manure (1:1) (T3)	5.8	4.54	18.8	1.13	2.38	0.87	3.61

Source: INIA.
Abbreviation: CRH, carbonized rice husk.

TABLE 6 Tukey's statistical test for total dry biomass (TDB) of *Calycophyllum spruceanum* plants by effect of different containers and substrates in nursery in conventional nursery.

	Container	
	Bag	Tub
Substrates	Total dry biomass	
Control: agricultural land + fine sand (1:1)	5.45Ac	3.53Ac
CRH + chicken manure + sawdust (2:1:1)	9.62Ab	7.05Ab
CRH + chicken manure (1:1)	27.40Aa	13.60Ba
Promedio	14.16	8.06

Note: Equal uppercase letters in the row and lowercase letters in the column do not show statistically significant differences according to Tukey's test at 1% probability.
Abbreviation: CRH, carbonized rice husk.

obtaining information on physiological state and nitrogen content, among others. In the same sense, secondary metabolites, such as photosynthetic pigments, are influenced by the quantity and composition of OM according to Pereira (2015).

Kampf (2005) mentions that, when there is no addition of OM in the substrates, there is a lower water retention capacity, low CEC values, and consequently low nutrient availability, which is reflected in lower chlorophyll values. Therefore, in this work, the substrates composed of CRH + chicken manure + decomposed sawdust (2:1:1) and CRH + chicken manure (1:1) presented the best results of chlorophyll index since the MO values were 28.64% and 18.81%, respectively, and also presented the best values of N, being 1.03% and 1.13%, respectively. On the contrary, when analyzing the control substrate, river sand + agricultural land (1:1) was determined to be only 0.97% of MO and 0.069% of N (Table 5).

In relation to the TDB, between the treatments: control-agricultural land + fine sand (1:1) and CRH + chicken manure + sawdust (2:1:1), there were no statistical differences, both in the bag and tube type containers; on the contrary, when the substrate CRH + chicken manure (1:1) was used in the bag type substrate, the dry biomass was statistically superior to the dry biomass of the plants produced in tube (Table 6). In this sense, the plants produced in the CRH + chicken manure substrate (1:1) presented 27.4 g, being statistically higher than the other substrates. However, the lowest values of 5.45 g of dry biomass were obtained by the plants produced in the control substrate and in the bag (Table 6).

Similar results were reported by Almeida et al. (2020) working in the production of plants *Handroanthus impetiginosus* (Mart. ex DC) with different substrates.

ANOVA for quality indices revealed that the substrates, containers, and the interaction of the factors caused significant statistical differences for the RI and DQI; on the contrary, it was only observed that the LI was significantly affected by the substrates factor. In this sense, in Table 7, it is observed that the plants produced in the CRH + chicken manure substrate (1:1) presented the best LI of 0.19. Meanwhile, the plants that were produced in the substrate composed of agricultural soil + fine sand (1:1) only reached values of 0.16. On the other hand, the values of the RI and DQI of the plants produced in the control and CRH substrates + chicken manure + sawdust (2:1:1) and in the bag container were statistically similar; however, when the substrate CRH + chicken manure (1:1) was used in the bag, the plants presented better quality indexes, being statistically superior to the values obtained in a tube (Table 7).

The quality indexes of the plants produced in CRH + chicken manure (1:1) were statistically superior to those obtained in the other substrates in bags and tubes, reaching 5.56 and 4.92 IR, and 3.26 and 1.79 DQI, respectively. However, the lowest quality indexes were determined in the control substrate, reaching values of 3.29 and 3.21 for IR and 0.89 and 0.63 for DQI in bag and tube, respectively (Table 7).

Similar results were reported by Aguirre et al. (2018), evaluating the effect of different substrates on the quality of seedlings in five forest species. They point out that plants with a higher IL index have greater development of the root system and aerial part, so they have a greater capacity to adapt in the final field; in addition, it provides support to the plants against water stress and climatic variations. Orozco et al. (2010) suggest that the plants produced should have between 0.17 and 0.26 IL. In this study, the IL (0.19) of the plants produced in the CRH substrate + chicken manure (1:1) in the bag type container is within the recommendations of the authors.

Moreira and Moreira (1996) indicate that the IR is recognized as one of the indices that best indicates the survival capacity of plants in the field. Thus, IR values between 5 and

TABLE 7 Tukey's test for lignification index (LI), robustness index (RI), and Dickson quality index (DQI) of *Calycophyllum spruceanum* plants by effect of different containers and substrates in conventional nursery.

	Container				
		RI		DQI	
Substrates	LI	Bag	Tube	Bag	Tube
Control: agricultural land + fine sand (1:1)	0.16b	3.3Ab	3.2Ab	0.9Ab	0.6Ab
CRH + chicken manure + sawdust (2:1:1)	0.17b	5.0Ab	4.1Ab	1.1Ab	1.0Ab
CRH + chicken manure (1:1)	0.19a	5.6Aa	4.9Ba	3.3Aa	1.8Ba
Average	0.17	4.6	4.1	1.7	1.1

Note: Equal uppercase letters in the row and lowercase letters in the column do not show statistically significant differences according to Tukey's test at 1% probability.
Abbreviation: CRH, carbonized rice husk.

10 indicate better plant quality, values over 10 indicate a very tall plant with respect to diameter, while values <5 indicate a plant of short height with respect to diameter. Therefore, the IR value (5.56) of the plants produced in the bag-type container with CRH + chicken manure (1:1) was within the recommendations of the authors.

However, the highest DQI (3.26) was obtained in the plants produced with the substrate CRH + chicken manure (1:1) in the bag container. Similar results were reported by Faria et al. (2016) working with *Mimosa setosa* on substrates composed of chicken manure. Gomes and Paiva (2004) report that the DQI is considered one of the most complete indexes for the evaluation of plant quality since its calculation includes the relationships between the morphological variables of the aerial part and BAP, BRP, and TDB. Therefore, the higher the value, the better the quality of the plants. Dickson et al. (1960) report that this index avoids selecting disproportionate plants and discarding plants of lower height, but with greater vigor.

On the other hand, the minimum value of DQI for Gomes and Paiva (2004) is 0.2. In this study, the lowest values were produced in tubes with the control substrate—agricultural soil + fine sand (1:1). Similar results were reported by Gonzaga et al. (2016) working with *Hymenaea courbaril* L plants; the authors determined that the plants produced in bags presented better morphological characteristics than those produced in tubes, a fact that they attributed to the greater volume of the substrate and consequently to the greater amount of nutrient available.

4 | CONCLUSIONS

The substrate based on CRH + chicken manure + decomposed sawdust (1:1:1) generated a higher percentage of emergence, emergency speed index, and mean emergence time in *C. spruceanum* plants.

The use of a bag type container and the substrate of CRH + chicken manure (1:1) allowed us to obtain *C. spruceanum* plants with the best lignification, robustness, and DQI at 90 days after planting.

AUTHOR CONTRIBUTIONS

Wilson Francisco Guerra-Arévalo: Conceptualization; methodology; project administration; resources; writing—original draft; writing—review and editing. **José Roy Cercado-Delgado**: Methodology; writing—original draft. **Héctor Francisco Espinoza-García**: Resources. **Tatiana Mildred Ucañay-Ayllon**: Validation. **Diego Gonzalo García-Soria**: Data curation. **Carlos Abanto-Rodríguez**: Formal analysis; writing—review and editing. **Dennis del Castillo-Torres**: Supervision; acquisition; supervision; validation. **Luis Ernesto Freitas-Alvarado**: Methodology. **Rossana Díaz-Soria**: Supervision. **Héctor Guerra-Arévalo**: Formal analysis; investigation; writing—review and editing.

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CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

ORCID

Wilson Francisco Guerra-Arévalo https://orcid.org/0000-0002-2887-1436

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