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Spent mushroom compost for germination and growth of *Ricinus communis* L.

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ABSTRACT
Objective: To determine the response of castor oil seeds and seedlings to six different substrata: regional soil (sandy-clay-loam), sand, spent mushroom compost (CAC), and mixes of CAC with 50% (A1:CAC1), 66% (A1:CAC2), and 33% (A2:CAC1) sand.
Design/Methodology/Approach: The experimental design consisted of two randomized complete blocks with 24 experimental units per substratum, monitored from their germination until 50 days later. Seed weight was correlated with the final seedling biomass.
Results: The best germination ($P<0.05$) was obtained with the A2:CAC1 and A1:CAC1 substrata, while regional soil had the lowest germination. CAC produced the highest growth and biomass in 50-day-old plants ($P<0.05$), followed by A1:CAC2, A1:CAC1, and A2:CAC1, while regional soil and sand had the lowest development. Seed weight had a statistically positive correlation ($P<0.05$) with seed biomass only in the sandy substratum.
Study Limitations/Implications: The experimental period and pod size limited the achievements of this research.
Findings/Conclusions: Independently of seed weight, CAC, and mixes with sand provide suitable texture and nutrient contents for castor oil seedling's germination and growth.

Keywords: seeds, seedlings, substrata, soil type.

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INTRODUCTION

The castor oil plant, or Ricinus (*Ricinus communis* L.), is an oily species with vast industrial applications, such as oils, lubricants, cosmetics, biofuels, and pharmacological products (Yeboah *et al.*, 2021). In Mexico, its cultivation was limited to only 1,044 ha in the entire territory in 2020 (SIAP, 2021); consequently, more than 6.6 million US dollars of castor oil was imported for industrial uses in the center of the country (DataMéxico, 2021). Therefore, its cultivation can generate significant income in the primary sector. An essential factor for its establishment in the places where it is intended to be cultivated is to determine its growing characteristics and its agricultural demands. In this sense, animal- and vegetable-waste-based composts are good sources of nutrients and contribute

Image by Alejandro Sánchez

to the sustainable improvement of soil structure since its nutrients are reintegrated into the system, avoiding their contribution to landfills (Adugna, 2016).

Spent mushroom compost (CAC) is the residual product of mushroom (*Agaricus bisporus* Lange) production. Although useless for the mushroom, it still has high levels of nutrients and enzymes (García-Mendívil *et al.*, 2014b). The agricultural use of CAC has been shown to improve the production of beans (*Phaseolus vulgaris* L.) and wheat (*Triticum* spp.) by increasing their productivity (García-Mendívil *et al.*, 2014a, 2014b). However, its use has not yet been tested in *R. communis*. In addition, *Ricinus* has not been commercially established yet in the central region of the state of Veracruz; consequently, its response to the site's soils is unknown.

Additionally, suitable substrata for its germination and growth have not been evaluated. Previous works have analyzed the germination and growth of castor oil plants in different substrata, such as sand, mixtures of sand with tezontle and compost (type of compost not specified), and regional soils. However, the substrate has never been evaluated, only plant provenances (Solís-Bonilla *et al.*, 2011, 2017; García-Herrera *et al.*, 2019). Meanwhile, although the importance of seed weight for better plant growth is known (Valdés-Rodríguez *et al.*, 2018), the response of seed biomass of *R. communis* to different substrata has not yet been researched. Therefore, further research is required to generate adequate recommendations for producers interested in venturing into this crop. Therefore, this research aimed to evaluate the response of *R. communis* seeds and seedlings to germination and development in the regional soil, sand, and sand-based mixtures and spent mushroom compost.

MATERIALS AND METHODS

Experimental site and plant material

The study was carried out in the city of Xalapa, Veracruz, Mexico (19° 30' 54" N and 96° 55' 05" W). It has a semi-warm humid climate, with average temperatures between 18 and 20 °C (INEGI, 2018). Seeds of *R. communis* were collected from a mother plant from the state of San Luis Potosí, in the Mexican Plateau (22° 36' 12" N, 100° 24' 47" W). The plants had the following characteristics: medium-sized plants (2-3 m), serrated purple leaves, glands on the margin, reddish brown veins, and red petioles; purple flower clusters with medium-sized fruits (2.5-3.0 cm) and greyish-purple seeds. Each seed was weighed individually with an analytical balance (Ohaus 310 g*0.1 g), obtaining an average weight of 491 mg per seed ± a standard deviation of 44 mg per seed. The following substrata were used: coarse sand (A) (1-2 mm) from the region, regional soil (soil extracted from the surface up to a depth of 30 cm) and spent mushroom compost (CAC). Additionally, the following ratio combinations of sand and compost were used: A1:CAC1 (50:50), A1:CAC2 (33:66), and A2:CAC1 (66:33). The silica sand was obtained in the suburbs of the municipality of Xalapa, Veracruz, Mexico. The CAC came from Altex Rioxal, a company that operates in the town of Las Vigas, Veracruz, Mexico, and which uses wheat straw enriched with wheat bran, gypsum as a structure improver, and sodium carbonate as an acidity or alkalinity stabilizer.

Experimental design and measuring

The experimental design consisted of a randomized complete block with 24 experimental units per substratum and two repetitions. The experiment was established in a 5 m×15 m×2 m (width×length×height) greenhouse covered with polyethylene. One seed per 15×20 cm black plastic bag was sown in their corresponding substratum. The first repetition (Rep1) began on November 27, 2014, and the second (Rep2) started on January 5, 2015. The number of sprouted seedlings was recorded daily. For each treatment, the germination percentage (the number of emerged plants divided by the number of seeds sown) and the mean germination time by substratum (the average number of days it took for all the sprouted seedlings) were estimated. The seedlings were watered every three days until the substratum was saturated. For each plant, the following variables were recorded every four days: plant height (metric tape), root collar diameter (digital vernier) at the base where it emerged, length and width of the largest leaf, and the number of leaves. The leaf area was estimated using the following equation: Area=0.55×length×width of the leaf (Jain & Misra, 1966). The observation period per plant was 50 days. Each plant was removed at 50 days, and the length and diameter of the taproot and primary lateral roots were measured with a measuring tape and a digital vernier, respectively. Also, the number of primary lateral roots (those sprouting directly from the stem base) was counted. Subsequently, the plant was separated into roots, stem, and leaves to obtain the fresh and dry weight of each one with an Ohaus digital scale (310 g*0.1g). During the experiment, the maximum and minimum temperatures and the environmental humidity were recorded daily with a digital thermo-hygrometer.

Substratum analysis

The pure substrata (sand, soil, and compost) were analyzed to determine texture (Bouyoucos method), pH (in aqueous solution, digital potentiometer), electrical conductivity (CE, conductivity meter), and organic matter (Walkley and Black). The textures of the substrata were divided as follows: sand (sandy), regional soil (sandy-clay-loam), and compost (sandy-loam-silty) (USDA, 2022) (Table 1). Organic matter was considered very low for sand, medium for soil, and high for CAC. The electrical conductivity had negligible salinity effects in the sand and regional soil, but it was slightly saline for CAC. The pH was considered neutral in the three substrata (SEMARNAT, 2002).

Table 1. Chemical analysis of the base substrata used to germinate *Ricinus communis* seeds.

Substratum	Textural composition (%)	Organic matter (%)	Electric conductivity (dS m^{-1})	pH
Sand	Sand: 91.3, clay: 0, silt: 8.7	0.32	0.04	7.10
Regional Soil	Sand: 59.3, clay: 22.4, silt: 18. 3	3.12	0.97	6.65
CAC	Sand: 62.6, clay: 16.0, silt: 21.4	27.17	2.83	6.85

CAC: spent mushroom compost.

Growth dynamics

The growth dynamics of the height, root collar diameter, leaf area, and the number of leaves were evaluated using a parameterized Gompertz model (Mora-Chacón *et al.*, 2022), represented by Equation 1.

$$V(d) = Is * \frac{e^{-e\beta_1(d_b-\beta_2)}}{e^{-e\beta_1(d-\beta_2)}} \quad (1)$$

Where: $V(d)$ is the value at time d, Is is the average value of the series at the base time, d is the time at which the function is evaluated, d_b is the base period, and 1 and 2 are constants that model the growth and shape of the curve. The increase in leaf area was represented by the straight-line equation (*area=slope*time+constant*) because this model obtained a higher coefficient of determination than the Gompertz model. The coefficient of determination (r^2) was obtained with the least squares method to determine the representation of each variable.

Statistical analysis

The Log Rank test ($P \leq 0.05$) was used to evaluate germination and survival between treatments. This test is considered appropriate to analyze events that change over time with probability distributions of the germination curve type (Romano and Stevanato, 2020). Final biomass and morphological variables were subject to an analysis of variance for a randomized complete blocks model with two replicates, using substratum and replicates as factors. In all cases, mean comparisons were performed using Tukey's test ($P \leq 0.05$). Correlations between seed weight and the final seedling biomass were performed using a Pearson linear correlation for each substratum. The SigmaPlot 10.0 software was used for all tests.

RESULTS AND DISCUSSION

Germination and survival

The Log Rank test showed significant differences between treatments (Rep1, $P<0.001$; Rep2, $P=0.046$). The sand and compost substrata (A1:CAC1 and A2:CAC1) obtained the highest germination rates and the shortest germination times, while the sandy-clay-loam soil recorded the lowest and slowest germination (Table 2). Consequently, sand-compost mixtures were the most suitable substrata for germination because they were light and nutritious, with a pH close to neutral. Since the CAC substratum was treated against mushroom pathogens, the mixtures allowed good aeration, stimulated germination, and prevented diseases (Sun *et al.*, 2013; García-Mendívil *et al.*, 2014). Improvements in germination with substrata that combine sand and compost are also reported for other oily seeds, such as *Jatropha curcas* (Díaz-Chuquizuta *et al.*, 2017), highlighting the ability of the compost to degrade seed coat and improve the imbibition of the seed. The average germination times (Table 2) were like those reported by Valdés-Rodríguez and Pérez-Vázquez (2019) at 22-28 °C temperatures, 71-89% humidity levels, and 4-10 days

Table 2. Average environmental conditions during germination of *R. communis* in different substrata.

Substratum and environmental conditions	Germination (%)		Mean germination time (days)	
	Rep1	Rep2	Rep1	Rep2
Sand	95.8 b	91.7 b	8.9	9.2
Regional soil	66.7 c	91.7 b	9.6	9.3
CAC	91.7 b	91.7 b	9.3	9.3
A1:CAC2	100.0 a	95.8 ab	7.2	9.0
A1:CAC1	100.0 a	100.0 a	7.8	8.9
A2:CAC1	100.0 a	100.0 a	7.8	9.0
Maximun temperature (°C)	26.9	23.7		
Minimun temperature (°C)	19.8	15.9		
Humedad ambiental (%)	80.1	79.6		

Rep1: November 27, 2014; Rep2: January 5, 2015. CAC: spent mushroom compost, A1:CAC1: 50 % sand y 50 % CAC, A2:CAC1: 66 % sand y 33 % CAC, A1:CAC2: 33 % sand y 66 % CAC. Different letters in the same column indicate statistical differences (Tukey, $P \leq 0.05$).

on average. In this regard, *R. communis*' great plasticity allows it to adapt to different temperatures but germinates better at ranges from 20 to 30 °C (Ribeiro *et al.*, 2014). In this work, the minimum temperatures recorded in Rep2 (3.9 °C less than in Rep1) could have caused a slight delay in the mean germination time, which on average, lasted 1.4 days more in mixtures with compost.

Plant growth

The analysis of variance showed significant differences between treatments for all variables and between repetitions for plant height and leaf area ($P<0.001$). The CAC substratum recorded the largest sizes in all the variables, followed by the A1:CAC2 substratum, which was similar to the CAC in plant height and number of leaves (Table 3). The sand substratum showed the lowest growth, while height growth and diameter thickness increased by 38% and 18%, respectively, in the region's soil compared to the sand substratum. The minimum addition of CAC (A2:CAC1) resulted in significant increases in all variables, with plant height values 2.2 times higher, stems 1.4 times thicker, and 3.3 times greater leaf area than in the sandy substratum. Leaf area and stem thickness showed the most significant growth responses to increments in the percentage of CAC. These results indicate that the CAC substratum or its mixtures with sand provide the nutrients, the pH, and a suitable texture to achieve better seedling development. In this regard, the texture and pH of the CAC were ideal for the growth of *R. communis* since the roots of this species are better suited to penetrate light-texture soils with 6-7 pH levels, which facilitates nutrient absorption (Souza *et al.*, 2014; Wang *et al.*, 2019). Concerning the alkalinity of the CAC, Soares de Lima *et al.* (2015) found that *R. communis* grows well with CE values of 2.1 dS m^{-1} and the results in this work confirm that slightly higher levels do not affect plant development. The environmental conditions explain the differences between repetitions: in Rep1, the average temperature was 16.1 °C, while Rep2 recorded 16.9 °C and 38 more minutes of sun exposure. These conditions favor a more significant development of the stem

Table 3. *R. communis* plant measurements in different substrata 50 days after germination.

Substratum	Root collar diameter (mm)	Plant height (cm)	Number of leaves	Leaf area (cm^2)
Sand	5.29 e	10.04 d	5.00 c	21.10 c
Regional soil	6.59 d	16.54 c	5.47 bc	50.59 d
A2:CAC1	7.26 c	22.33 b	5.81 ab	69.84 c
A1:CAC1	7.44 bc	22.46 b	6.12 ab	84.82 b
A1:CAC2	7.77 b	25.44 ab	6.00 ab	96.04 b
CAC	8.55 a	26.09 a	6.19 a	116.53 a

CAC: spent mushroom compost, A1:CAC1: 50% sand y 50% CAC, A2:CAC1: 66% sand y 33% CAC, A1:-CAC2: 33% sand y 66% CAC. Different letters in the same column indicate statistical differences (Tukey, P≤0.05).

and leaf area since the growth response of *R. communis* increases in higher temperatures and solar radiation due to its high photosynthetic capacity (Ribeiro *et al.*, 2014).

Concerning the growth dynamics (Figure 1), the Gompertz model explained 95 to 99% of the average variation in heights and number of leaves and 85 to 96% in root

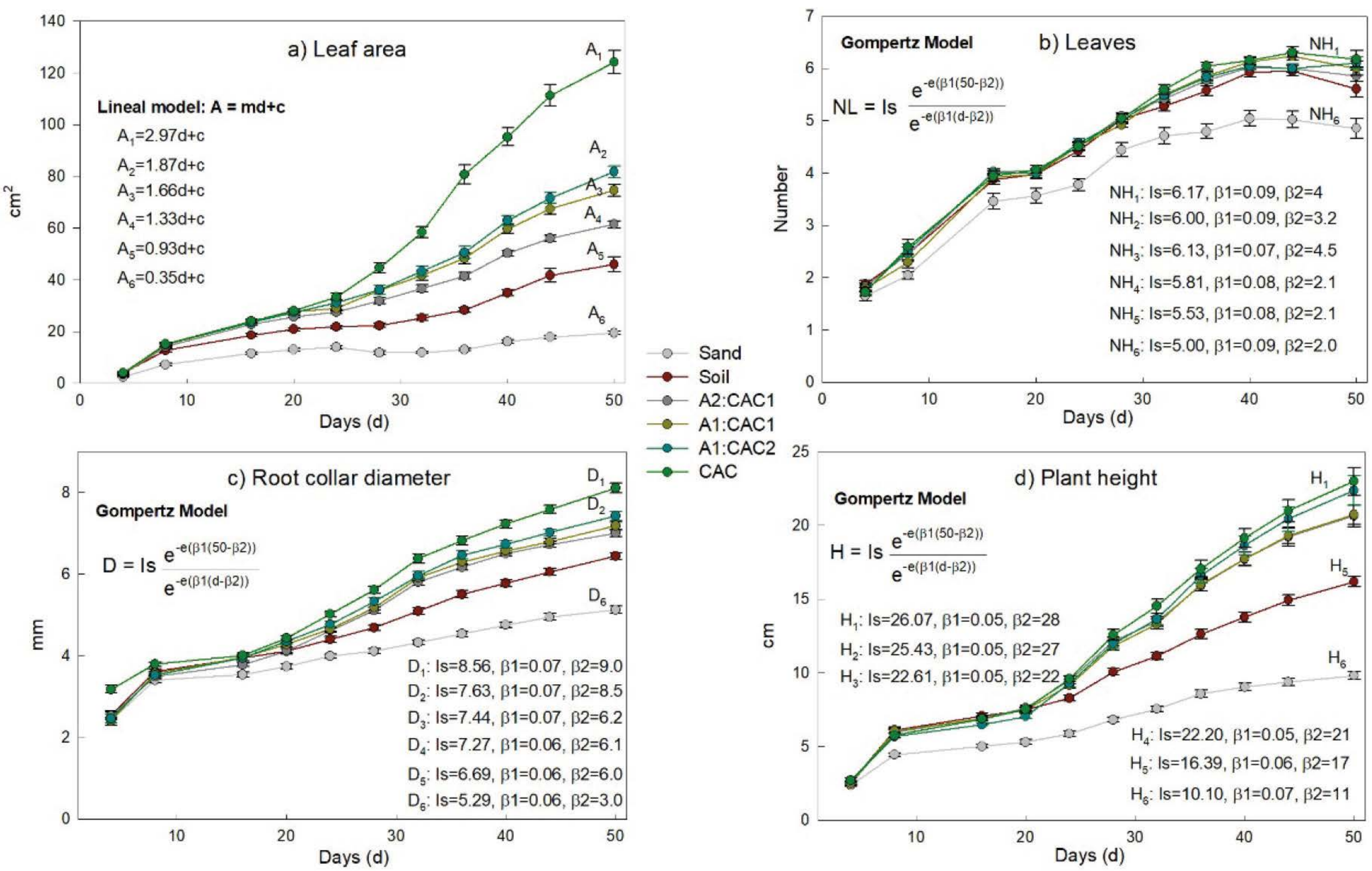

Figure 1. Aerial growth of *Ricinus communis* on different substrata. Each point represents the mean of 48 replicates ± standard error. A: leaf area, NH: number of leaves, D: root collar diameter, AP: plant height, CAC: spent mushroom compost, A1:CAC1: 50% sand and 50% CAC, A2:CAC1: 66% sand and 33% CAC, A1:CAC2: 33% sand and 66% CAC.

collar diameter. Therefore, this model is considered appropriate to represent the growth dynamics of *R. communis* seedlings (Mora-Chacón *et al.*, 2022). The 2 coefficient —related to the growth rate of the curve— indicates an increase of up to 2.5 times in seedling stems in CAC compared to those in the sand substratum. Regarding the leaf area, linear growth had rates eight times higher in CAC than in sand. These increases in foliage growth rates are mainly attributed to the high content and availability of N that CAC possesses, which allows an accelerated development of seedlings, especially in the foliar area (Nahar and Pan, 2015). On the other hand, the progressive decrease in growth (*e.g.*, the number of leaves) can be explained by the size of the container bags that limited the development of the plants because this depends on the size of the pod (SEB, 2012).

Root systems

The analysis of variance showed statistical differences between substrata in all variables but only found differences between repetitions in the number of roots, with an average of only 4% more lateral roots in Rep2 than in Rep1 (Table 4). The CAC substratum recorded the thickest taproots, while the total length and diameter of lateral roots were similar in all treatments with some CAC ratio but were higher than those obtained in soil and sand. The light textural composition and nutrient concentration in CAC resulted in thicker roots, allowing good aeration and facilitating their development in length and thickness (Nahar and Pan, 2015). Nevertheless, roots are usually thinner in a nutrient-poor substrate (Helliwell *et al.*, 2019), as can be seen in the case of the roots in the sand, which were thinner than in substrata with some compost ratio.

Seedlings fresh and dry weight

The analysis of the variance of fresh weights found highly significant differences between the substrata and the repetitions in all plant parts ($P<0.001$). Highly significant differences were also found in plant dry weight, although there were no differences in the stem between repetitions (Table 5). The fresh and dry weights of stems, leaves, and roots were statistically higher in the CAC than in the other substrata. A1:CAC2 and A1:CAC1

Cuadro 4. Average root measurements of 50-day-old *R. communis* seedlings in different substrata.

Substratum	Taproot length (cm)	Lateral roots length (cm)	Taproot diameter (mm)	Lateral roots diameter (mm)	Number of lateral roots
Sand	31.80 c	31.14 c	3.38 d	0.87 b	32.77 a
Suelo de la región	30.03 c	30.53 c	3.95 c	0.87 b	34.85 a
A2:CAC1	34.28 bc	33.52 bc	4.78 b	1.00 a	34.33 a
A1:CAC1	37.07 ab	34.74 ab	5.05 b	1.04 a	35.18 a
A1:CAC2	37.18 ab	35.46 ab	5.22 b	1.09 a	33.35 a
CAC	39.77 a	36.91 a	6.09 a	1.10 a	34.73 a

CAC: spent mushroom compost, A1:CAC1: 50% sand y 50% CAC, A2:CAC1: 66% sand y 33% CAC, A1:CAC2: 33% sand y 66% CAC. Different letters in the same column indicate statistical differences (Tukey, $P\leq0.05$).

Cuadro 5. Average fresh and dry weights of *R. communis* in different substrata 50 days after germination.

Substratum	Fresh weight (g)			Dry weight (g)		
	Leaves	Stem	Roots	Leaves	Stem	Roots
Sand	1695.50 f	1601.5 e	4242.9 e	307.29 e	271.88 e	428.96 e
Regional soil	3902.30 e	4090.7 d	5916.4 d	646.15 d	555.29 d	559.02 d
A2:CAC1	5371.50 d	7255.6 c	11187.1 c	819.85 c	767.00 c	787.42 c
A1:CAC1	6545.30 c	8206.0 bc	12448.6 b	1024.35 b	888.27 b	877.60 b
A1:CAC2	8319.30 b	8834.3 b	12814.0 b	1067.73 b	898.79 b	900.94 b
CAC	11387.10 a	11396.5 a	16001.8 a	1660.60 a	1189.92 a	1160.59 a

CAC: spent mushroom compost, A1:CAC1: 50% sand y 50% CAC, A2:CAC1: 66% sand y 33% CAC, A1:CAC2: 33% sand y 66% CAC. Different letters in the same column indicate statistical differences (Tukey, $P \leq 0.05$).

recorded similar weights, and they outperformed A2:CAC1, which was superior to the plants in regional soil, while they were more developed than the ones in the sand. This behavior implies that the higher the nutrient concentration in the substratum, the greater the development of all the plant organs. In this regard, studies with CAC and beans (*Phaseolus vulgaris* L.) showed that compost increases the photosynthetic capacity of plants (García Mendívil *et al.*, 2014), enhancing their ability to assimilate and process nutrients. For their part, Nahar and Pan (2015) found that the response of 28-day-old seedlings of *R. communis* to different nitrogen levels is directly proportional to the increase in their biomass, indicating that high levels of nitrogen favor the initial growth of *R. communis*. It is also noteworthy that the root-biomass ratio increased in the substrata with fewer nutrients: 31% in compost and 43% in the sand. This performance indicates that the roots of the plants in substrata with few nutrients had a more significant development than their foliage, which allowed them to explore their medium better and obtain more nutrients (Azcón-Bieto and Talón, 2013).

Correlation of seed weight with final dry weight per substratum

The correlations between seed weight and seedlings' dry weight show that seed weight only has a positive and significant correlation ($P \leq 0.05$) with plant biomass in the sandy substratum (the least nutritious substratum). Meanwhile, there is only a positive correlation, but not significant, between the regional soil and the substrata with the lowest compost content (Table 6). These values indicate that nutrient content in CAC and their mixes produce such development in the plants that seed biomass does not play a predominant role in the growth of the *R. communis* seedlings. Meanwhile, seed weight is essential in the less nutritious substratum to obtain additional energy for the plant's better development and survival under unfavorable conditions (Kolodziejek, 2017). These results also match the findings of Valdés-Rodríguez and Pérez-Vázquez (2018), who found significant positive correlations between *R. communis* seedlings biomass and their seed weight in a sandy and nutrient-poor substratum 59 days after germination.

Table 6. Correlation coefficients between seed weight and final dry weight of *R. communis* seedlings at 50 days.

Substratum	Leaf	Stem	Root
Sand	0.56*	0.65*	0.57*
soil of the region	0.18	0.27	0.37
A2:CAC1	0.21	0.32	0.29
A1:CAC1	0.00	0.10	0.05
A1:CAC2	0.07	0.28	0.06
CAC	−0.31	−0.35	−0.31

*Significative to $P \leq 0.05$.

CONCLUSIONS

R. communis seeds can germinate in less time and with a higher percentage in light substrata mixed with spent mushroom compost (CAC) and sand. When CAC is the only element of the substratum, it enables the best development of stems, leaves, and roots compared to its mixtures with sand in ratios of up to 66%. Besides, root and aerial development of *R. communis* seedlings is significantly improved in soils with up to 33% CAC mixed with sand compared with sandy substrata with a low content of organic matter or a sandy-clay-loam soil moderately rich in organic matter. Seed weights positively impacts the growth of *R. communis* seedlings only in nutrient-poor sandy substrata, but its effect cannot be determined in medium- or high-nutrient substrata.

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