Growing lettuce seedlings in different organic composts used as substrate

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Abstract

Lettuce is the most important vegetable in the world and the most consumed in Brazil. Seedling production with a good quality substrate is very important in the lettuce crop production process. The objective of this study was to evaluate the performance of organic compost substrates in the production of lettuce seedlings. The experiment was carried out in a greenhouse, using polyethylene trays containing 128 trapezoidal cells in a completely randomized design with a 5×2 factorial scheme consisting of 5 substrates (commercial substrate; compost I; compost II; compost II; compost IV) and P_2O_5 supplementation (absent and 0.525 kg m⁻³ P_2O_5) and 4 replications. Substrates were produced by composting agro-industrial waste: sawdust, wood shavings and residues from a restaurant, pig litter, cattle manure, residues from the shoot and tubers of sweet potato, soybean residue, and drained bovine ruminal residue. The emergence, plant height, number of leaves, fresh root and shoot biomass, and clod stability of the substrate were evaluated. The substrate formulated from pig litter, cattle manure, and sweet potato (compost II) and pig litter and drained bovine ruminal residue (compost IV) formed seedlings with a greater clod stability of the substrate, plant height, number of leaves, and fresh root and shoot biomass. Substrates based on composting pig litter, cattle manure, and sweet potato (compost II) and pig litter, cattle manure, and sweet potato (compost II) and pig litter and drained bovine ruminal residue (compost IV) were most suitable for producing lettuce seedlings.

Keywords: composting, phosphorus, Lactuca sativa L., organic waste

Introduction

Lettuce (*Lactuca sativa* L.) is an annual plant that is considered one of the most important vegetables in the world, and it is the most consumed vegetable in Brazil, especially as salads, *in natura*, and as a complement in sandwiches (Henz & Suniga, 2009; Sala & Costa, 2012; Echer et al., 2016). China is the world's largest producer with a production of 23.6 million tons. In Brazil, approximately 671.509 tons of lettuce are produced annually (Kist et al., 2018).

Field cultivation of lettuce in the conventional system is the most representative in terms of area and production (Henz & Suniga, 2009) and depends directly on transplanting seedlings. Seedling production is one of the most important steps in the production process, and the producer must obtain good quality seedlings, free from pests and diseases, and with uniform growth in the definitive field. According to Medeiros et al. (2008), the substrate is the most complete resource to produce seedlings, and it can come from a single source material or through mixtures.

A good substrate must have properties such as low density, high water retention capacity, good drainage, neutral, non-saline, non-alkaline, non-acidic, uniform, easily found, low cost to the producer, contain no phytopathogenic contaminants, weeds, or toxic substances, maintain stable properties or qualities when being sterilized, and capable of storage for a relatively long period while maintaining its quality (Schafer & Lerner, 2022). The quality of the compost reflects the proportions and materials that comprise the mixture.

In this sense, substrates from organic composts can have excellent physical, biological, and chemical properties and produce seedlings of excellent quality and with a good performance in the field (Schafer & Lerner, 2022). To obtain a substrate with the aforementioned

Eq. 1

Eq. 3

characteristics, the use of material available in the region, such as organic waste from other crops and/ or the consumption of food for composting and the formulation of substrates, is an alternative to the use of commercial substrates (Carvalho et al., 2012). Previous studies have suggested the feasibility of using alternative substrates made from decomposed swine manure and wood shavings, along with sugarcane bagasse and vermicompost, to produce lettuce seedlings (Monteiro et al., 2012; Freitas et al., 2013).

In this sense, the production of a substrate with alternative materials, low cost, and agro-industrial residues is of great importance. The objective of this study was to evaluate the performance of organic compost substrates in lettuce seedling production.

Materials and Methods

The experiment was conducted in a greenhouse covered by a shading screen with 50% light interception on the *campus* of the Federal Institute of Education, Science and Technology of Mato Grosso, Juína, Mato Grosso state, Brazil (11° 26' S and 58° 43' W at 320 m altitude), from April to May of 2020.

The experimental design was completely randomized in a 5 × 2 factorial scheme, with the factors consisting of 5 substrates (commercial; compost I; compost II; compost III; compost IV) and P_2O_5 (absent and present) and 4 replications. The characterization of the production method and chemical composition of the composts are shown in **Tables 1** and **2**. Phosphorus was applied at a dose of 0.525 kg m⁻³ of P_2O_5 , and the nutrient source was simple superphosphate (21% P_2O_5). The phosphorus dose was defined based on David et al. (2008). Each experimental plot consisted of 32 cells, in which only 10 central plants in each plot were evaluated, thus disregarding the border plants.

The lettuce cultivar used was the pelleted crispy cv. Amanda (Seminis Company, Bayer, with 90% germination). The seeds were deposited at a depth of approximately 1.0 cm in polyethylene trays containing 128 trapezoidal cells and 40 mL of the substrate treatment. Then, the trays were placed in the greenhouse under supports at a height of 0.80 m. Irrigation was performed using a microsprinkler system for 1 min, 10 times between 06:00 am and 18:00 pm. The evaluations started 3 days after sowing and lasted for another 24 days, when the seedlings presented more than 3 definitive leaves and the study ended.

In the initial phase, the percentages of initial and final emergence, average time, speed index, and emergence speed were calculated according to the following formulas:

- Emergence speed index (ESI):

ESI =∑ ^ĸ_{i=1} ni ÷ ti

where ESI = emergence speed index; ni = number of emerged seeds per day;

ti = sowing time (days); i = interval (3–9 days). Unit: dimensionless.

- Mean emergence time (MET):

$$MET = \underbrace{\left(\sum_{i=1}^{K} nix ti\right)}_{\sum_{i=1}^{K} ni} Eq. 2$$

where MET = mean emergence time; ni = number of emerged seeds per day;

i = sowing time (days); i = range 3–9 days. Unit: days.

- Average germination speed (AGS): AGS = $\frac{1}{TME}$

where AGS = mean emergence speed; TME = mean time to emergence.

The relative total chlorophyll content was evaluated using a chlorophyllometer (ClorofiLOG, model CFL 1030, Falker®) at the central point of the last fully developed leaf. The stem diameter (SD) was obtained at the height of the plant's neck with readings taken on a digital caliper with a precision of 0.03 mm. Plant height (H) was determined from the soil surface to the shoot apex using a millimeter ruler. The number of true leaves was obtained by directly counting the fully developed leaves on the plant.

Substrate clod stability was evaluated at the end of seedling production, at the time of individualization of the seedlings, and dispatch for transplanting, in relation to the permanence of the clod in the container, with assigned grades that ranged from 1 to 5 (**Figure 1**), where grade 1 corresponds to the substrate whose clod had the lowest stability and grade 5 to that of best stability (Freitas et al., 2010), as described below:

- Grade scale 1: Low stability, more than 50% of the clod is retained in the container, and the clod does not remain cohesive;

- Grade scale 2: Between 10 and 30% of the clod is retained in the container, and the clod does not remain cohesive;

- Grade scale 3: The clod detaches from the container but does not remain cohesive;

- Grade scale 4: The clod detaches from the container, but there is a loss of up to 10% of the substrate;

- Grade scale 5: The entire clod is detached from the container, and more than 90% of it remains cohesive.

The plants were then sectioned and separated

Table 1. Composition and method of obtaining substrates.

Substrate	Composition	Method of obtaining		
Commercial	Poultry manure, limestone, and processed and decomposed bark and vermiculite.	Industrial process.		
Compost I	Sawdust (30%), wood shavings (30%), and residues from the preparation and food of a restaurant (40%).	Composting for 120 days, with the windrow being turned over every 30 days. Irrigation of the piles took place when the humidity was below 50%.		
Compost II	Pig litter composed of feces and wood shavings (40%), cattle manure (40%), and residues from sweet potato shoots and tubers (20%).	Composting for 120 days, with the windrow being turned over every 30 days. Irrigation of the piles took place when the humidity was below 50%.		
Compost III	Soybean residue composed of the shoot (50%) and swine litter composed of feces and wood shavings (50%).	Composting for 120 days, with the windrow being turned over every 30 days. Irrigation of the piles took place when the humidity was below 50%.		
Compost IV	Pig litter composed of feces and wood shavings (50%) and drained bovine ruminal residue (50%).	Composting for 120 days, with the windrow being turned over every 30 days. Irrigation of the piles took place when the humidity was below 50%.		

Table 2. Chemical characterization of the commercial substrate and composts used in the experiment.

Parameter	Units	Commercial Substrate	Compost I	Compost II	Compost III	Compost IV
pH (H ₂ O)	-	6.69	7.19	7.90	7.50	7.69
pH (CaCl ₂)	-	6.00	6.40	7.30	6.80	7.09
Ν	g kg-1	5.60	11.20	16.80	15.40	15.40
Р	g kg ⁻¹	1.50	2.10	6.60	9.70	6.40
К	g kg ⁻¹	2.90	4.50	4.90	0.80	2.10
Ca	g kg-1	13.80	11.70	38.50	55.80	40.60
Mg	g kg-1	3.20	2.10	8.30	9.60	7.30
S	g kg ⁻¹	0.70	1.20	2.10	2.20	1.30
В	mg kg⁻¹	33.00	11.40	16.40	23.20	14.80
Cu	mg kg⁻¹	62.00	22.00	105.0	213.0	100.0
Fe	mg kg⁻¹	31703	8425	10935	9876	8219
Mn	mg kg⁻¹	874.6	177.0	318.4	385.0	283.4
Zn	mg kg⁻¹	68.00	44.00	197.0	201.0	132.0

pH – Hydrogen potential



Figure 1. Substrate clod stability rating scale.

between the shoot and root systems. The roots were washed in running water to remove excess substrate, and on an analytical balance with a precision of 0.0001 g, the fresh root biomass (FRB) and the fresh shoot biomass (FSB) were determined. The roots and shoots were placed in paper bags and placed in a forced circulation oven for 72 hours at 65°C. They were then weighed to determine the dry root biomass (DRB) and dry shoot biomass (DSB).

From the previously measured parameters, the

total dry biomass (TDB) was determined, and the following morphological indices were calculated: height (cm)/ stem diameter (mm) ratio (H/SD); dry shoot biomass/dry root biomass (DSB/DRB); and Dickson quality index (DQI) (Dickson et al., 1960) using the equation DQI = total dry biomass (g)/((height (cm))/diameter (mm)) + (dry shoot biomass (g)/dry root biomass (g)).

In all considered datasets, data normality was analyzed using the Anderson–Darling test, and the

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homoscedasticity of the data was verified using the variance equation test (or Levene's test). The data obtained for each variable were submitted to variance analysis ($p \le 0.05$) and the factors were compared using the Scott-Knott test in the statistical program R version 3.2.3 with the ExpDes package. For the substrate clod stability and initial emergence variables, the raw data were transformed by $\sqrt{x} + 0.5$ to standardize the variance. However, the values presented are the original means.

Results and Discussion

Different substrates significantly influenced the parameters evaluated, except for IE, ESI, MET, and AGS. Phosphorus application was not significant for the evaluated parameters, except for FRB. There was a significant interaction between the substrates and phosphorus application for DSB, TDB, and DQI (**Table 3**). Watthier et al. (2017) verified significant effects in lettuce seedling production when using substrates made from tung compost, carbonized rice husk, and earthworm humus. Fonseca et al. (2019) found positive effects of using compost and silage made with fish waste as a substrate in lettuce germination and growth.

Composts II and IV showed greater substrate clod stability compared to the other composts and to the commercial substrate (**Figure 2**A). Both substrates had grades higher than four, indicating that the clod detached itself from the container, but there was a loss of up to 10% of the substrate (Figure 1). The better stability of these substrates may be related to a balance between larger and smaller particles, which caused a better substrate porosity, such as wood shavings and swine manure. Wood shavings after composting contained some particles that conditioned the substrate to have a better porosity, reflecting better seedling root development and consequent aggregation of material when the seedlings were removed. According to Regan (2014), the addition of agro-industrial residues, such as wood shavings, when used in the substrate, should ensure structural stability, allow gas exchange through the roots, and provide water to the plants.

The highest emergence percentage was verified in compost II (86%), although there was no significant difference in relation to the commercial substrate (72%) and compost IV (67%) (Figure 2B). According to Santos et al. (2010), the use of organic waste in the substrate's composition contributes to aeration, water retention capacity, and the formation of an adequate physical structure for the development of the radicle and epicotyl, corroborating the effects of the composted materials in this study.

Fonseca et al. (2019) found a higher percentage of seedling emergence of lettuce cv. Monica when using fish waste compost as a substrate. Monteiro et al. (2012) verified the greatest emergence of lettuce seedlings when the seeds were sown in the alternative substrate composed of 100% decomposed swine manure + wood shavings and in the commercial substrate, corroborating the results obtained in this study.

The largest stem diameter was observed in lettuce seedlings grown in compost II (**Figure 3**A), being 29% higher than those produced in the commercial substrate. Stem diameter best predicts the seedling's performance after planting (Ritchie et al., 2010), and thus, higher values of this variable are generally related to an abundant root system, as verified in this study. According to Cargnelutti Filho et al. (2012), this favors the establishment and growth of plants when transplanted to the field. When composting waste from the chicken production chain

SV	SCS	IE	E	ESI	MET	AGS
Substrate(S)	31.97**	2.191 ^{ns}	3.484*	1.582 ^{ns}	2.050 ^{ns}	1.355 ^{ns}
Phosphor(P)	0.228 ^{ns}	2.480 ^{ns}	3.617 ^{ns}	2.056 ^{ns}	0.897 ^{ns}	0.180 ^{ns}
S × P	0.086 ^{ns}	0.585 ^{ns}	0.514 ^{ns}	1.234 ^{ns}	1.647 ^{ns}	2.245 ^{ns}
CV	9.41	41.18	35.09	37.27	11.87	13.42
FV	Н	SD	RCC	NL	FRB	FSB
Substrate(S)	15.55**	3.703*	6.085**	8.090**	5.950**	12.610**
Phosphor(P)	0.150 ^{ns}	0.142 ^{ns}	3.629 ^{ns}	1.823 ^{ns}	6.346*	0.013 ^{ns}
S × P	2.005 ^{ns}	1.355 ^{ns}	0.195 ^{ns}	2.276 ^{ns}	2.133 ^{ns}	2.614 ^{ns}
CV	14.36	20.32	6.75	6.57	26.88	29.50
FV	DRB	DSB	TDB	H/S	DSB/DRB	DQI
Substrate(S)	6.623**	5.969**	7.320**	5.848**	4.021**	3.984**
Phosphor(P)	0.667 ^{ns}	1.985 ^{ns}	1.736 ^{ns}	0.047 ^{ns}	0.029 ^{ns}	1.133 ^{ns}
S × P	2.366 ^{ns}	4.556**	4.577**	0.204 ^{ns}	0.144 ^{ns}	2.884*
CV	33,44	27.64	26.08	22.01	25.71	32.81

Table 3. Analysis of variance (F values) for different effects on the agronomic development of lettuce seedlings.

* and **: significant at 1 and 5%, respectively; ns: not significant; SV: sources of variation; CV: coefficient of variation (%); SCS: substrate clod stability; IE: initial emergence; E: emergence; ESI: emergence speed index; MET: mean emergence time; AGS: average germination speed; H: plant height; SD: stem diameter; RCC: relative chlorophyll content; NL: number of leaves; FRB: fresh root biomass; FSB: fresh shoot biomass; DRB: dry root biomass; DSB: dry shoot biomass; TDB; total dry biomass; H/S: height/stem diameter ratio; DSB/ DRB: dry shoot biomass/dry root biomass ratio; DQI: Dickson quality index.



Figure 2. Substrate clod stability (A) and percentage of lettuce seed emergence due to different substrates (B). Means followed by the same letter do not differ by the Scott–Knott test (5% probability). The vertical bar indicates the standard error of the mean (n = 4).



Figure 3. Growth and relative chlorophyll content of lettuce plants due to different substrates. Means followed by the same letter do not differ by the Scott–Knott test (5% probability). The vertical bar indicates the standard error of the mean (n = 4).

and using this compost as a substrate in the production of lettuce seedlings, Ripp et al. (2020) verified that the diameter obtained in seedlings produced in this type of substrate was 27% higher than those produced in the commercial substrate. These results are very close to those observed in this study.

The highest plant height was observed in lettuce plants grown on substrates based on composts II and

IV (Figure 3B). The higher plant height obtained in the compost-based formulated substrates is related to the greater availability of macronutrients for the plants (Table 1). The use of substrates made from organic residues in lettuce (Fonseca et al., 2019; Ripp et al., 2020), cabbage, and beetroot (Costa et al., 2014) also favored the increase in plant height, reflecting the best seedling vigor and development (**Figure 4**).



Figure 4. Visual appearance and growth of lettuce plants due to different substrates.

The highest number of leaves was verified in seedlings produced in composts II, III, and IV, producing 15% more leaves, on average, compared to the commercial substrate (Figure 3C). This result differs from that found by Gonçalves et al. (2014), who found no significant differences between the number of leaves of lettuce plants that were produced using the compost of agro-industrial residues as a substrate and those produced using the commercial substrate.

The highest relative chlorophyll content was observed in plants produced in the substrate formed from composts III and IV (Figure 3D). The quantification of chlorophyll in lettuce leaves is important, as it is an indicator of the level of nitrogen in plants, and this element is absorbed from the substrates produced from these composts, as shown in Table 1, where these composts show a higher amount of N and Mg, which are closely linked to the photosynthesis process.

The highest FRB was observed in lettuce seedlings produced in compost II (**Figure 5**A). Similarly, fertilization with P promoted changes in the root biomass of lettuce plants regardless of the substrate, and the addition of this element to the substrate resulted in a reduction in the root biomass (Figure 5B).

Greater root biomass production is related to greater structural stability given by the arrangement of particles that make up this substrate, which is reflected in better root development. Good root production (Figure 5A), which is associated with good clod stability (Figure 2A), provides an excellent condition for transplanting the seedling to the bed. The reduction in seedling growth with the addition of P may be related to the amount of this element already achieved by the compost (Table 1). Fonseca et al. (2019) found that the substrate produced from fish waste promoted better root system development in lettuce seedlings. In contrast, Costa et al. (2014) found no differences between the use of commercial substrate and substrates based on compost made from vegetable waste and cattle manure.

The highest FSB was obtained in seedlings produced in composts II and IV (Figure 5C). The commercial substrate produced lettuce seedlings with 41% less fresh biomass compared to the best composts. According to Fonseca et al. (2019), the increase in fresh shoot biomass in lettuce seedlings is of fundamental importance, as it reflects the gain of water and nutrients by the seedlings, thus reflecting the vigor, and therefore, the substrate elaborated from pig litter compost made from feces and wood shavings + cattle manure + shoot residues and sweet potato tubers and swine litter composed of feces and wood shavings + drained bovine ruminal residues are excellent alternatives for the composition of substrates.

The highest DRB was obtained when the substrate used was formed by composts II, III, and IV (Figure 5D), with the biomass obtained from the commercial substrate being about 51% lower than the average obtained for these substrates. According to Filgueira (2013), good rooting and the restart of plant development after transplanting are favored by tissues rich in dry mass. When formulating substrates from regional residues, Brito et al. (2017) showed that the substrate formed from carnauba residue and rice husk promoted a dry root biomass in lettuce seedlings that was superior to that which used a commercial substrate, similar to the results obtained in this study.

For DSB (Figure 5E) and TDB (Figure 5F), an interaction was observed between P fertilization and the substrates. For compost II, P addition reduced the biomass, while for compost III, P addition improved the biomass production of the seedlings. In relation to substrates where P was not added, the substrate formulated from compost II produced higher DSB and TDB, with biomass production being approximately twice as high as that



Figure 5. Fresh and dry root and shoot biomass of lettuce plants as a function of different substrates and phosphate fertilization. Means followed by the same letter do not differ by the Scott–Knott test (5% probability). The vertical bar indicates the standard error of the mean (n = 4).

of the commercial substrate. For the substrates where P was added, the DSB and TDB obtained from seedlings produced in substrates based on composts II, III, and IV were superior to those produced in the commercial substrate.

An increase in DSB and TDB is of fundamental importance, as they reflect greater nutrient accumulation in the lettuce seedlings, resulting in greater vigor and survival of these seedlings when transplanted to the soil. In this sense, composts II, III (with P supplementation), and IV proved to be an excellent alternative, as they favored the increase in total dry biomass and dry shoot biomass. According to Ripp et al. (2020), the organic substrate is one of the most significant factors related to the seedling quality, as it directly influences root development, which is reflected in shoot growth. Unlike the results achieved in this study, Watthier et al. (2017) did not verify efficiency in the use of substrates formulated with tung compost, carbonized rice husk, and earthworm humus.

The highest H/SD ratio was found when using composts II, III, and IV (**Figure 6**A). The H/SD ratio is a characteristic that expresses the morphological profile of the shoot, so that if the value of H/SD is high, it can be an indication that, in the shoot, the dry matter partition favors longitudinal growth to the detriment of lateral growth. If this does not occuran increase in the diameter of the



Figure 6. Effect of different substrates and fertilization on the morphological indices of lettuce plants. Means followed by the same letter do not differ by the Scott–Knott test (5% probability). The vertical bar indicates the standard error of the mean (n = 4).

stem compatible with the growth in height can cause the seedling to topple over. However, despite representing a lower toppling risk, a very low H/SD value may reflect the formation of seedlings in which height growth is slow, compromising field development with weeds and can even affect the productivity of the culture.

Based on visual observation of the seedlings (Figure 4), the seedlings were not etiolated, and in those with a lower H/SD ratio, seedlings had less capacity to grow in the field. In this sense, the high values of H/SD ratio cannot be analyzed individually and characterized as a negative aspect in seedling production, especially in the case of lettuce plants since the growth in height is more intense in the initial phase of the plant than in the growth in the stem diameter (Zuffo et al., 2016).

The lowest values of the DSB/DRB ratio were found in plants produced on the commercial substrate and in compost II (Figure 6B). Seedlings with a higher DSB/ DRB ratio are totally undesirable for transplanting, as they are more susceptible to death by dehydration, as their leaf area is exposed to solar radiation and wind and the small root volume is not sufficient for water and nutrient absorption, resulting in lower survival rates of plants and slow initial development and leading to a greater need for replanting and higher production costs (Schwertner et al., 2013). When evaluating alternative substrates in cucumber seedling production, Medeiros et al. (2018) verified lower DSB/DRB values when using a commercial substrate, thus disagreeing with the results obtained in the present study.

For the DQI, an interaction was observed between P fertilization and the substrates. For the substrates formulated from compost II, P addition reduced the DQI, while for the other composts, it did not matter (Figure 6C). In relation to substrates in which P was not added, the substrate formulated from compost II had the highest DQI; this value was approximately twice as high as the commercial substrate. For the substrates in which P was added, there was no significant difference (Figure 6C).

DQI is a good indicator of seedling quality, and the highest values for this parameter are classified as having the best quality (Petter et al., 2012; Araújo et al., 2016). This makes it an important tool because its calculation considers parameters that assess the robustness and distribution of biomass in the plant. The higher DQI in lettuce seedlings obtained with the substrate formulated from compost II reflects the good performance of this substrate in the parameters of height, diameter, and biomass (Figures 3 and 4). Similar to the results obtained in this study, Ripp et al. (2020) found a higher DQI in lettuce seedlings cultivated in substrates made from organic composts compared to those cultivated in commercial substrates.

The assessment of seedling quality should not

be based on the use of an isolated parameter, since if this assessment considers only the height, for example, it could indicate that the best quality seedlings would be those with increased height growth. However, when taken to the field, they tend to topple. In contrast, smaller seedlings would be more resistant to toppling, but they would be neglected. Therefore, the relationships based on biomass production, height, and stem diameter, such as the DQI, may better represent the balance between the shoot and root system, making it possible to obtain similar values for seedlings with greater growth and seedlings underdeveloped in height.

Conclusions

Substrates formulated by composting pig litter containing feces, wood shavings, cattle manure, and residues from the shoot and sweet potato tubers (compost II) and swine litter composed of feces, wood shavings, and drained bovine ruminal residue (compost IV) were those that produced the best lettuce seedlings. The production of organic composts formulated from agro-industrial residues and, subsequently, their use as a substrate to produce lettuce seedlings proved to be a good alternative to the reuse of residues.

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