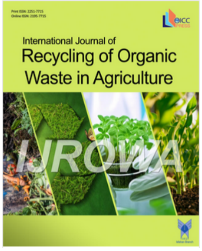

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Agronomic performance of curly lettuce seedlings from millicompost substrate under organic cropping system

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Original Research

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Abstract:

Purpose: Millicompost is an organic substrate generated by the activity of millipedes (diplopods) with high potential for seedling production. This work aimed to evaluate the performance of green curly lettuce (*Lactuca sativa* L.) using seedlings produced with millicompost, at different levels of nitrogen (N) fertilization at seedling transplant, in an organic production system.

Method: A field experiment was carried out in Seropédica, Rio de Janeiro State, Brazil, in a 2x4 factorial design with four replications, combining seedlings produced with two substrates (millicompost or Biomix®-used as reference) and four levels of N fertilization with castor cake (0, 50, 100 and 200 kg N ha^{-1}) at seedling transplant. Plants were sampled weekly, and lettuce production was evaluated at 42 days after transplant.

Results: Seedlings produced with millicompost had root dry mass, plant height, and number of leaves higher than those produced with commercial substrate. At levels 0 and 50 kg N ha^{-1}, lettuce plants originating from seedlings produced with millicompost showed a higher growth rate than those with the commercial substrates, between 35 and 42 days after transplant. Fresh commercial weight, head diameter, and number of leaves of lettuce plants increased linearly as applied N levels increased. These traits were superior in plants from seedlings produced with millicompost than with commercial substrate.

Conclusion: The millicompost provided seedlings of improved quality that increased the growth and yield of lettuce plants in the field at different levels of N fertilization.

Keywords: *Lactuca sativa*; Plant growth analysis; Organic fertilization; Horticulture; *Trigoniulus corallinus*

1. Introduction

Lettuce (*Lactuca sativa* L.) is one of the most important horticultural crops in the world and is produced in distinct environments such as production fields and protected systems. Lettuce includes several commercial varieties, of which the green curly type has great relevance (Stamford et al., 2019; Antunes et al., 2016). Lettuce production in Brazil was 671,509 Mg in the year 2020, with 64% of this amount yielded in the Southeast region (IBGE, 2020).

Seedling production affects the performance of horticultural crops (Borcioni et al., 2016), and the seedling substrate must guarantee the initial plant development after transplanting to the field. In organic production systems, some commercial substrates are restricted due to the presence of soluble chemical fertilizers or by comprising non-renewable resources such as peat (Santos et al., 2010; Luo et al., 2021), and organic substrates need registration by certifying entities (Sediyama et al., 2014). Moreover, the acquisition costs of these substrates by farmers should be taken into account (Ziech et al., 2014). Such costs can be reduced by using organic materials of quality and suitability for organic production, available in the farm or close by (Sediyama et al., 2014).

Millipedes are diplopod organisms whose popular names vary according to each region or country, such as gongolos in Brazil or rusty millipede in Asia (Kenny et al., 2015).

These diplopods act in litter fragmentation and decomposition via the association in their digestive tract with cellulose-digesting microorganisms (P Alagesan et al., 2003; Ashwini and Sridhar, 2005; Shelley et al., 2006; Kadamannaya and Sridhar, 2009; Alagesan, 2016). These millipedes excrete fecal pellets (coprolites) enriched with ammonia and uric acid, also increasing the soil contents of organic matter, phosphorus, calcium and magnesium (Souza and Resende, 2014; Alagesan, 2016). *Trigoniulus corallinus* is a species of pantropical distribution, occurring in different agricultural and natural environments (Kenny et al., 2015). Its individuals are about 5 cm long and have a reddish color (Antunes et al., 2019). These organisms are able to consume materials with a high C:N ratio and have been used in the process of composting of agricultural and urban residues, originating the so-called millicompost (Antunes et al., 2019, 2016).

Organic fertilization with animal manure and other organic sources has been used for cultivation of many vegetables, reducing the external inputs into the agricultural system and potentially decreasing nutrient leaching and its associated environmental impacts from chemical fertilizers (Ziech et al., 2014; Antunes et al., 2018). Organic fertilizers offer several benefits to the soil, including increasing soil water retention and percolation capacity, raising cation exchange capacity, and providing nutrients, thereby increasing the yields of several crops (Magro et al., 2010). Castor cake is an organic residue generated when the oil is extracted from the seeds of Ricinus communis. Due to its fast mineralization and high levels of nitrogen (N), phosphorus (P) and potassium (K), this cake has been widely used as a fertilizer in organic systems (Santos et al., 2012; McKeon, 2016; Lima et al., 2018).

The process of composting organic residues is improved by using organisms such as earthworms and millipedes. The millicompost has been shown to have potential as an organic substrate for seedling production of horticultural crops (Antunes et al., 2021b; Antunes et al., 2022a; Antunes et al., 2022b). Furthermore, the efficacy of a substrate should be validated in terms of providing water and nutrients for producing seedlings of improved quality and high performance in the field. Thus, the present work aimed to evaluate the performance of curly lettuce using seedlings produced with millicompost, at different levels of N fertilization with castor cake at seedling transplant, in an organic production system.

2. Materials and methods

Production of lettuce seedlings

The curly lettuce cultivar Vera, with high resistance to early bolting, was used (Sakata, 2023). The seedlings were produced in Embrapa Agrobiologia, in Seropédica, Rio de Janeiro, Brazil, in expanded polystyrene trays with 200 cells filled with two different substrates, the millicompost or the Biomix®commercial substrate, with 4 trays for each substrate. The Biomix®, a substrate suitable for seedlings and certified for organic production system, is composed of coconut powder or fiber, crushed and composted pine bark and Bokashi. The millicompost was produced by the mixture of leaves of the tree Bauhinia sp., leaves of *Paspalum notatum*, banana leaves (*Musa* sp.), and chopped cardboard (pieces $\leq$ 5 cm), in the proportion of 200, 150, 100 and 50 L for each residue, respectively. Adult individuals of *Trigoniulus corallinus* were collected manually in earthworm beds, compost pots and on the ground, and 2.2 L of animals (approximately 4000 individuals) were placed in concrete rings (0.5 m height and 1.0 m width) containing the mixture of waste. The composting proceeded for 180 days, as described by (Antunes et al., 2021a), and the compost was sieved into 2 mm mesh and stored in plastic bags.

The substrates were analyzed for physical properties such as macroporosity, microporosity, total porosity and volumetric density, using the tension table at 60 cm tension, and for water holding capacity at 10 cm tension (Brasil, 2008; Teixeira et al., 2017). The pH was measured with distilled water (5:1 v:v) and the electrical conductivity was determined in the same aqueous extract, as described by Brasil (Brasil, 2007). The N and C concentrations were determined in a CHN elemental analyzer (Nelson and Sommers, 2015). The total concentrations of P, K, Ca and Mg of the substrates were evaluated by digestion of the samples (Teixeira et al., 2017). The results of substrate analysis are shown in Table 1.

At 25 days after sowing, at transplanting to the field, ten lettuce seedlings were removed from each tray, totaling four samples of each substrate. The number of true leaves and the height of the plants (from the insertion of the root to the leaf apex) were evaluated. Shoots and roots were weighed, oven dried at 65°C for 72 h and weighed.

When the seedlings were removed, the clod stability was classified by notes varying from 1 to 5, as follows (Antunes et al., 2018): 1 low stability (more of 50% of the clod was retained in the container and the clod did not remain cohesive); 2 average stability (between 30% and 50% of the clod was retained in the container and the clod did not remain cohesive); 3 regular stability (between 15% and 30% of the clod was retained in the container and the clod did not remain cohesive); 4 good stability (the clod was completely detached from the container and up to 90% of its volume remains cohesive); 5 excellent stability (the clod was completely detached from the container and more than 90% of its volume remains cohesive).

Experimental conditions

A field experiment was carried out in the Integrated System of Agroecological Production (Fazendinha Agroecológica Km 47), at coordinates 22°45′21″ S, 43°40′28″ W, 33 m of altitude, in Sero-pédica, Rio de Janeiro, Brazil, between August and September 2018. The regional climate is Aw by Köppen. Climatic data collected at the Ecology Agricultural meteorological station, near the experimental area, showed an average maximum temperature of 21.2 and 23.3°C, and a minimum temperature of 20.1 and 22.2°C, in August and September, respectively, with total precipitation of 18 mm. The experiment had a randomized block design in a 2 × 4 factorial scheme with four replications, combining two substrates of lettuce seedlings (millicompost or Biomix®) and 4 levels of N fertilization with castor cake (0, 50, 100 and 200 kg N ha^{-1}).

Table 1. Values of pH, electrical conductivity, concentration of macronutrients, macro and microporosity, water retention capacity at 10 cm tension (CRA10 cm) and volumetric density, of the substrates Biomix®and millicompost used in the production of lettuce seedlings.

Attribute	Biomix	Millicompost
pH	6.69	7.34
Electrical conductivity (dS m^{-1})	0.49	0.69
C concentration (g kg^{-1})	325	354
N concentration (g kg^{-1})	7.76	23.25
P concentration (g kg^{-1})	1.53	2.96
K concentration (g kg^{-1})	2.32	4.78
Ca concentration (g kg^{-1})	9.03	31.69
Mg concentration (g kg^{-1})	2.73	4.48
C:N ratio	41.93	15.24
Macro porosity (%)	12.77	31.24
Micro porosity (%)	62.07	60.45
Total porosity (%)	74.85	91.70
CRA $_1 0cm$ (mL 50 cm^{-1})	31.04	30.23
Volumetric density (g cm^{-1})	0.45	0.20

The experimental area has a Red Yellow Argisol (Typic Hapludult) of low natural fertility. Soil chemical analysis at 0-20 cm depth (Teixeira et al., 2017) showed: C_{org} 7.0 g kg^{-1}; pH 6.7; N_{total} 1.0 g kg^{-1}; P available 161 mg L^{-1}; K available 143 mg L^{-1}; Ca 3.6 $cmol_c$ L^{-1}; Mg 0.9 $cmol_c$ L^{-1}; Al 0.0 $cmol_c$ L^{-1}; H+Al 1.5 $cmol_c$ L^{-1}. Soil was plowed and harrowed, and beds were raised with a laying device coupled to a tractor, with 0.20 m high, 1 m wide and 4.8 m long. Four days before seedling transplant, castor cake was applied at each respective N level and incorporated into the soil. This was the sole fertilizer applied during lettuce cultivation; therefore, the treatment with 0 kg N ha^{-1} did not receive any fertilization in the field. The analysis of castor cake, performed according to (Teixeira et al., 2017), presented the total levels of (in g kg^{-1}): N 58.6; P 6.4; K 11.25; Ca 17.8 and Mg 3.7.

The seedlings were transplanted to the field 25 days after sowing in trays. Each plot consisted of four rows spaced by 30 cm, with 24 plants per row spaced by 20 cm. Irrigation was performed by four lines of drips per bed with drippers spaced by 0.20 m, with a flow rate of 1.8 L h^{-1}. Water demand was estimated by evapotranspiration of reference by the Penman-Monteith method (Allan et al., 1998), corrected by crop coefficients, using meteorological data obtained at the Ecology Agricultural station.

Assays

Two plants were sampled per plot every seven days, between 7 and 42 days after transplanting (DAT). One plant was sampled from each of the two central rows, and a subsequent plant was kept alive to act as an inner border for the next sample. The plants were removed from the root system by shovel and taken to laboratory. The root system was washed, leaf area was determined by a photoelectric meter (Li-Cor, LI-3100), and shoots and roots were oven dried at 65°C for 72 h and weighed. Specific leaf area was estimated as the ratio between leaf area and leaf dry mass at each time of sampling. The final lettuce harvest occurred at 42 DAT, when six plants were removed from the two central rows of each plot by cutting the plants at ground level. The commercial fresh weight, the diameter and height of the plant, and the number of leaves over 5 cm were evaluated. Commercial productivity was estimated with the useful area index of the cultivated hectare (76%), with a plant density of 152,000 plant ha^{-1} (Antunes et al., 2018).

The dry samples of shoots collected at 42 DAT were ground. The concentrations of N, P, K, Ca and Mg were determined according to Teixeira et al. (2017). The amount of nutrient accumulated was obtained by multiplying the nutrient concentration value versus the dry mass of the shoot.

Statistical analysis

Data obtained from lettuce seedlings at transplant to the field were subjected to analysis of variance as a single factor comparing the two substrates by the F test. Data of total dry mass and leaf area per plant at different growth stages were adjusted by linear regression to the second-degree exponential polynomial model, with the sampling dates as independent variable. The values of R^2 were higher than 0.84 for biomass and 0.86 for leaf area. By differentiating the adjusted models, the absolute growth rate (AGR) and the net assimilation rate (NAR) were estimated. The data were tested for variance homogeneity by the Bartlett test and for normality by the Shapiro-Wilk test. Data on plant biomass and leaf area were transformed into a natural logarithm to homogenize the variance between harvests (Araújo, 2003). The analysis of variance was conducted as a triple factorial between substrate, N levels and harvests. The significance of the interaction between treatments and time of sampling for natural logarithm transformed data on plant biomass was interpreted as significant differences in plant growth rates (as discussed by Araújo (2003)). Data on final lettuce production and nutrient accumulation were subjected to analysis of variance as a double factorial between substrate and N levels. Response to N levels was evaluated through regression analysis considering N levels as independent variable. All analysis was performed with SISVAR software (Ferreira, 2014).

3. Results and discussion

The shoot dry mass of the lettuce seedlings was similar for the substrates millicompost or Biomix (Table 2). Seedlings produced with millicompost had a greater root dry matter, plant height and number of leaves than those produced with commercial substrate. The stability of the clod was also greater with millicompost (Table 2). Seedlings showed high vigor, good appearance, and absence of diseases or nutritional deficiencies. Analysis of variance of lntransformed data of biomass and leaf area indicated significant triple interactions between substrate, N levels and dates of harvest, indicating that the treatments affected differently the growth rates of lettuce plants in the field. The total dry mass and leaf area of lettuce plants (Fig. 1 and Fig. 2) were similar in all treatments between 7 and 21 days after transplant (DAT). At 42 DAT, at the level of 0 kg N ha^{-1}, lettuce plants originating from the millicompost accumulated more biomass than those from the commercial substrate; at the other N levels, plant biomass was similar for both substrates (Fig. 1). At 42 DAT, at the levels of 50 and 200 kg N ha^{-1}, plants raised in the millicompost had a greater leaf area than those grown in the commercial substrate (Fig. 2).

After seedling transplant to the field, the initial development of lettuces was slow, with low absolute growth rate (AGR) until 21 DAT. After this adaptation period, lettuce plants increased their AGR in every level of fertilization with castor cake (Fig. 3). At 0 and 50 kg N ha^{-1}, plants raised in millicompost showed a greater AGR than those with commercial substrate between 35 and 42 DAT (Fig. 3). The net assimilation rate (NAR) declined between 7 and 28 DAT but increased after 28 DAT except at 200 kg N ha^{-1} (Fig. 4). Plants at 0 kg N ha^{-1} presented lower initial values of NAR than at other N levels. At 0 kg N ha^{-1}, plants originating from millicompost showed lower NAR than plants with commercial substrate but had an intense increase in NAR overcoming plants from commercial substrate after 28 DAT (Fig. 4).

Plants originating from seedlings with millicompost had higher specific leaf area than plants with commercial substrate, at 0 kg N ha^{-1} at 14, 21 and 42 DAT, and at 50 kg N ha^{-1} at 35 DAT (Table 3). At 14 and 21 and 42 DAT, plants raised in millicompost had a higher specific leaf area at the lowest N level, whereas at 28 and 35 DAT plants with commercial substrate had a lower specific leaf area at the lowest N level (Table 3). Traits evaluated at lettuce harvest at 42 DAT showed a linear response to increased N levels applied to the crop (Fig. 5). Commercial fresh mass varied from 119 to 212 g per head for plants with commercial substrate, and from 120 to 363 g for plants raised in millicompost. Plants raised in millicompost had commercial fresh weight 34, 25 and 71% higher, respectively, at the levels 50, 100 and 200 kg N ha^{-1}, than plants grown in commercial substrate. The height and diameter of lettuces were higher with millicompost but only at 200 kg N ha^{-1}. Plants raised in millicompost had a greater number of leaves than plants grown in commercial substrate at 50 and 200 kg N ha^{-1} (Fig. 5). Converting the commercial fresh mass per lettuce head to the land area, plants originating from seedlings with commercial substrate had yields ranging from 18.1 to 32.2 Mg ha^{-1}, whereas plants raised in millicompost from had yields ranging from 18.2 to 55.2 Mg ha^{-1}, at the levels of 0 and 200 kg N ha^{-1}, respectively. The amount of macronutrients accumulated in shoots of lettuce plants at 42 DAT increased linearly with the level of N applied (Fig. 6). The amount of nutrients followed the order K > N > Ca > P > Mg. The accumulation of nutrients was higher in lettuce plants from seedlings produced with the millicompost, particularly at lower applied N levels. At 200 kg N ha^{-1}, only P and Ca accumulation was higher in plants raised in millicompost, as compared with plants grown with commercial substrate (Fig. 6).

The stage of seedling production is very important for the performance of lettuce crop, since weak seedlings delay to establish in the field thereby compromising plant growth. The millicompost provided lettuce seedlings with enhanced growth than the commercial substrate (Table 2), thereby influencing the subsequent performance of lettuces in the field. The millicompost had nutrient concentrations much higher than the commercial substrate and better physical properties such as lower density and higher clod stability, which facilitates the root system development (Table 1 and Table 2). Values of volumetric density between 100 and 300 kg m^{-3} and porosity higher than 85% are con-sidered as reference for substrates used in trays (Fermino, 2014). The millicompost met these benchmarks, while the commercial substrate showed higher density (Table 1), maybe due to the pine bark with high density in its composition. Antunes et al. (2016) also found that the millicompost was an efficient substrate for the production of lettuce seedlings. Ramanathan and Alagesan (2012) obtained improved growth and pepper production with millicompost as compared to vermicompost. The total lettuce growth cycle ranges from 120 to 170 days until seed production (Franco et al., 2018). The commercial

Table 2. Shoot dry weight (SW), root dry weight (RW), plant height (PH), number of leaves (NL) and clod stability (CS), evaluated in seedlings of curly lettuce cultivar Vera produced with two substrates at 25 days after sowing.

	mg plant^{-1}				
Substrate	SW	RW	PH(cm)	NL(unit)	CS
Biomix	80 a	10 b	10.8 b	6.0 b	3.5 b
Millicompost	80 a	20 a	11.6 a	6.4 a	4.9 a
CV (%)	22.64	37.59	9.18	6.17	16.25

Means followed by the same letter within a column do not differ by F test at 5% level.

Table 3. Specific leaf area of lettuce plants originating from seedlings produced with two substrates (millicompost or Biomix), at different levels of nitrogen fertilization (0, 50, 100 and 200 kg N ha-1) with castor bean cake, in Seropédica, Rio de Janeiro, Brazil.

	Specific leaf area (cm2 g-1) Millicompost			
Days after transplant	0 kg N ha-1	50 kg N ha-1	100 kg N ha-1	200 kg N ha-1
7	211	187	184	196
14	406*a	333 b	327 b	309 b
21	408*a	336 b	354 b	285 b
28	186	198	210	238
35	207	270*	244	264
42	262*	244	221	258
	Specific leaf area (cm2 g-1) Biomix			
Days after transplant	0 kg N ha-1	50 kg N ha-1	100 kg N ha-1	200 kg N ha-1
7	184	175	172	150
14	276	286	324	277
21	282 b	279 b	310 a	232 b
28	157	177	193	197
35	181 b	193 a	218 a	208 a
42	174	211	209	206

Means followed by the same letter within a row do not differ by Scott-Knott at 5% level.
* Significant difference between substrates in the same fertilizer level and date by the F test at 5% level.

harvest of leaves of curly lettuce cultivar Vera usually begins at 60 days of total growth cycle (Sakata, 2023). In the present work, the lettuce cycle comprised 67 days (25 days in trays more 42 days in the field). Lettuce plants were harvested at full vegetative development without displaying signals of the reproductive stage, such as stem elongation and a reduction in leaf number (Aquino et al., 2014). Therefore, the growth rates of lettuces increased throughout the experimental period. After 35 DAT, lettuce plants from seedlings with millicompost showed higher growth rates than those with commercial substrate, particularly at low levels of N fertilization (Fig. 3 and Fig. 4), which confirms the beneficial effects of the seedlings produced with millicompost. The analysis of the microbiota during the millicomposting process revealed the presence of the Alphapro-teobacteria class of the Rhizobiales order, taxa which comprise several diazotrophic bacteria (Antunes, 2021). The presumed presence of diazotrophics and growth-promoting microorganisms in the millicompost could have contributed to the improved performance of lettuce plants, as compared to

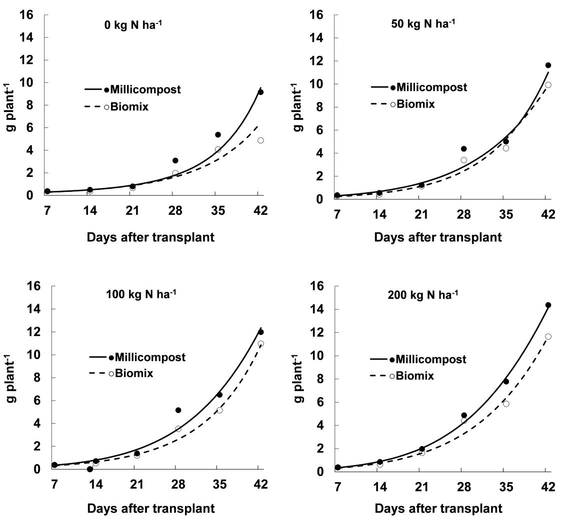

Figure 1. Total dry mass of lettuce plants originating from seedlings produced with two different sub-strates (millicompost or Biomix), at different levels of nitrogen fertilizer (0, 50, 100 and 200 kg N ha-1) with castor cake, in Seropédica, Rio de Janeiro, Brazil.

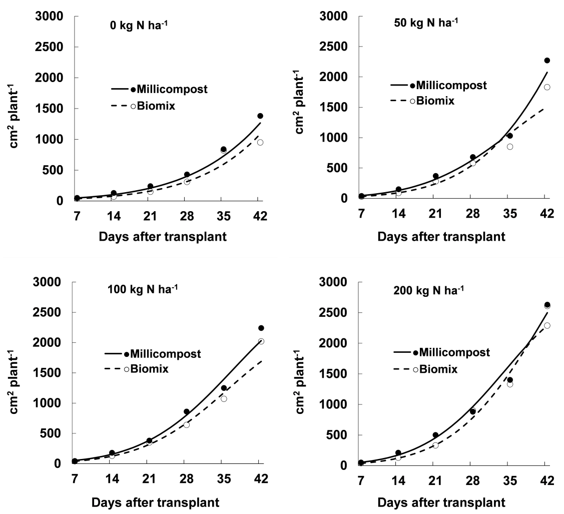

Figure 2. Leaf area of lettuce plants originating from seedlings produced with two different substrates (millicompost or Biomix), at different levels of nitrogen fertilizer (0, 50, 100 and 200 kg N ha-1) with castor cake, in Seropédica, Rio de Janeiro, Brazil.

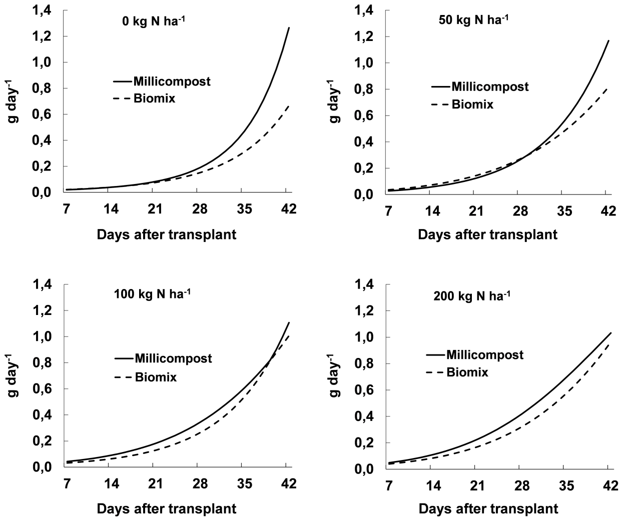

Figure 3. Absolute growth rate of lettuce plants originating from seedlings produced with two different substrates (millicompost or Biomix), at different levels of nitrogen fertilizer (0, 50, 100 and 200 kg N ha^{-1}) with castor cake, in Seropédica, Rio de Janeiro, Brazil.

plants grown with commercial substrates, regardless of the levels of N fertilization.

Under conditions of high solar irradiance, plants of C3 species tend to increase the specific leaf area (i.e. by producing thinner leaves) under restricted N availability (Evans and Poorter, 2001). Lettuces produced with millicompost grown without N fertilizer increased their specific leaf area between 7 and 14 DAT, as compared to plants produced with commercial substrate (Table 3). Concomitantly, the net assimilation rate was lower in plants raised in millicompost during the initial growth in the field (Fig. 4), since thinner

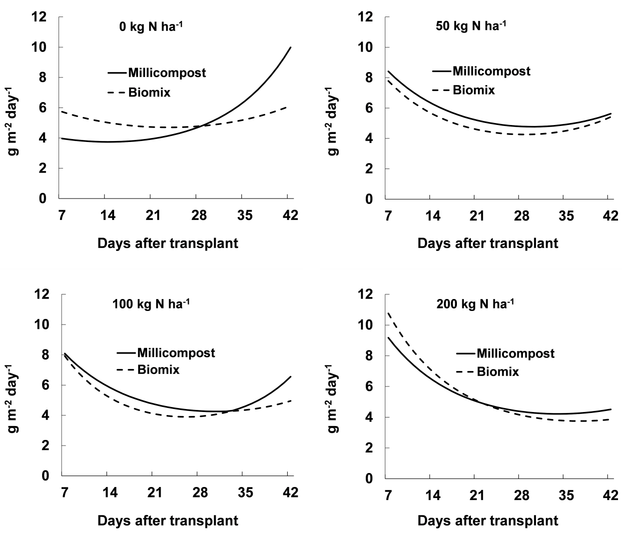

Figure 4. Net assimilation rate of lettuce plants originating from seedlings produced with two different substrates (millicompost or Biomix), at different levels of nitrogen fertilizer (0, 50, 100 and 200 kg N ha^{-1}) with castor cake, in Seropédica, Rio de Janeiro, Brazil.

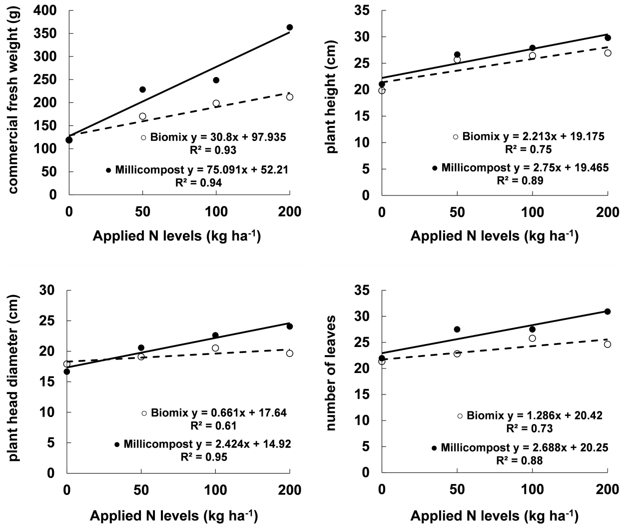

Figure 5. Commercial fresh weight per plant (A), plant height (B), plant head diameter (C) and number of leaves per plant (D) of lettuce plants originating from seedlings produced with two substrates (millicompost or Biomix), at different levels of nitrogen fertilization (0, 50, 100 and 200 kg N ha^{-1}) with castor cake, in Seropédica, Rio de Janeiro, Brazil.

leaves usually have a lower photosynthetic activity per unit leaf area (Evans and Poorter, 2001). This indicates an improved ability of adaptation to low N availability in plants produced with millicompost.

Lettuce plants responded to increased N fertilization up to the maximum level of 200 kg N ha^{-1} with castor cake (Fig. 5), regarding traits of commercial fresh mass, plant height, head diameter and number of leaves. Insufficient N supply reduces the leaf area of lettuces and renders the leaves less attractive to consumers (Santos et al., 2018). According to Araújo et al. (2011), recommended fertilization for lettuce is around 100 to 130 kg ha^{-1} of N or 40 to 60 Mg ha^{-1} of cattle manure. Vaz et al. (2019) observed that curly lettuce responded to levels up to 150 kg N ha^{-1} while levels above 200 kg N ha^{-1} reduced plant growth. Santos et al. (2018) also found a linear response of lettuces to increased N levels, but the highest yield occurred at the level of 260 kg N ha^{-1}. Mantovani et al. (2017) verified that the fresh weight of lettuces increased linearly as the amount of applied cattle manure was increased up to the level of 160 kg N ha^{-1}. Castor cake is an N-rich material broadly used as organic fertilizer, presenting a fast mineralization of N and P, although elevated levels can be detrimental to plant growth (Lima et al., 2011). Assuming that the castor cake was not entirely mineralized during the short growth cycle of lettuces in the field, high levels of N such as 200 kg N ha^{-1} would be required for maximum growth and yield (Fig. 5).

Kano et al. (2012) observed the order K > N > Ca > P > Mg for nutrient uptake in lettuce, similar to this work. Plants originating from seedlings produced with millicompost accumulated more nutrients, mainly at the level of 0 kg N ha^{-1}, where nutrient uptake was twice that of plants

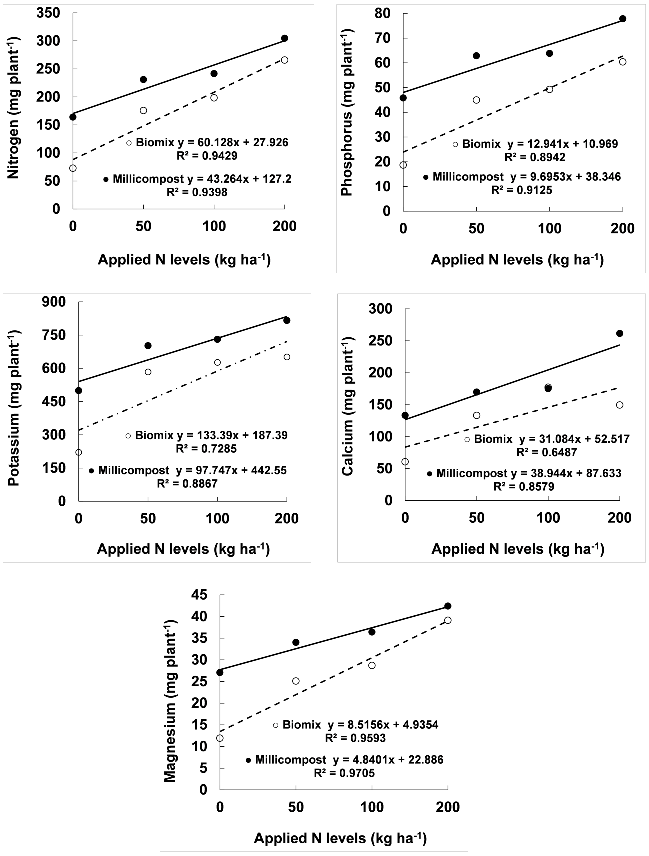

Figure 6. Accumulation of nitrogen, phosphorus, potassium, calcium and magnesium in shoots of lettuce plants originating from seedlings produced with two substrates (millicompost or Biomix), at different levels of nitrogen fertilization (0, 50, 100 and 200 kg N ha^{-1}) with castor cake, in Seropédica, Rio de Janeiro, Brazil.

from commercial substrate (Fig. 6). At levels of 50 and 100 kg N ha^{-1}, nutrient uptake was very close in both substrates, indicating that the effect of the millicompost was reduced as N availability in the field increased.

The number of leaves and the head diameter affect the market value of lettuces. Lettuces harvested at 42 DAT had number of leaves and head diameter greater in plants originating from millicompost than from commercial substrate, even at higher levels of N fertilization (Fig. 5). Curly lettuces usually have from 17 to 25 leaves per plant and head diameter between 20 and 21 cm (Antunes et al., 2016; Meneses et al., 2016; Sousa et al., 2018; Goulart et al., 2018). At the level of 200 kg N ha^{-1}, the lettuce originating from seedlings with millicompost had a fresh weight of 363 g and 30 leaves per plant, above the values recorded by Antunes et al. (2018)(308 g and 19 leaves per plant) for the cultivar Vera originating from seedlings with millicompost treated with Bokashi organic fertilizer. The estimated lettuce yield per land area was 34, 25 and 72% higher from seedlings with millicompost than with commercial substrate, at the levels of 50, 100 and 200 kg N ha^{-1}, respectively (Fig. 5). Yields of plants that received castor cake are within the range foreseen in the organic production system, between 20 and 40 Mg ha^{-1} (Souza and Resende, 2014), but plants raised in millicompost at the level of 200 kg N ha^{-1} yielded even more, with 55 Mg. Antunes et al. (2018) obtained yields of 38 Mg ha^{-1} in the curly lettuce cultivar Vera fertilized with Bokashi at a level equivalent to 130 kg N ha^{-1}.

The potential of composts produced with earthworms as substrates for seedling production is widely recognized (Steffen et al., 2010; Oliveira, 2011), but substrates processed by diplopods are little known. Antunes et al. (2018) demonstrated that millicompost can be produced within 90 days, but longer processing may be necessary to achieve a better substrate since the organic material becomes more stable and richer in nutrients over time. Millipedes are easily found for capture in humid areas with litter accumulation, where they act as detritivores (Bugni et al., 2019). Under conditions of low humidity and temperature, especially in winter, these animals reduce their activity and become more difficult to find. Unlike earthworms, whose reproduction is well known and easy to conduct, the management of millipedes requires further study. During the composting process, these animals produce abundant offspring, but few reach adulthood. This can be associated with the type "r" strategy of arthropods, with a high population at the beginning of colonization of the environment (Souza and Resende, 2014).

Lettuces harvested after 42 days of growth in the field showed improved yields when plants were originating from seedlings produced with millicompost. Millicompost is an organic substrate of easy production that presents physical and chemical characteristics suitable for plant development during the seedling stage. Furthermore, it is a ready-to-use organic compost that spares the combination with other feedstock, reducing the costs for farmers. The potential of millicompost for enhancing plant growth by producing humic substances and stimulating growth-promoting microorganisms deserves further studies.

4. Conclusion

Millicompost can be used as a substrate for the production of lettuce seedlings, providing superior plant growth compared to commercial substrate.

The lettuces that were developed on both substrates responded to the increase in nitrogen fertilization up to the level of 200 kg N ha^{-1} with castor bean cake, increasing the fresh mass of the plants and the amounts of accumulated macronutrients.

Lettuce plants from millicompost showed a higher number of leaves, stem diameter and commercial yield than those from commercial substrates, at different levels of nitrogen fertilization.

Author contribution

Luiz Fernando de Sousa Antunes: Conceptualization, Methodology, Investigation, Formal analysis, data curation, Supervision, Writing – original draft, Writing – review & editing. Adelson Paulo Araújo: Investigation, Supervision, Writing – review & editing. Maria Elizabeth Fernandes Correia: Writing – review & editing, Resources, Funding acquisition. Norma Gouvêa Rumjanek: Supervision, Writing – review & editing, Resources, Funding acquisition. José Guilherme Marinho Guerra: Writ-ing – review & editing.

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Conflict of interest statement

The authors declare that they are no conflict of interest associated with this study.

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