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SOURSOP SEEDLINGS: EMERGENCE AND DEVELOPMENT UNDER DIFFERENT CULTIVATION ENVIRONMENTS AND SUBSTRATES – PART I

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ABSTRACT: Seedling quality is crucial to obtain vigorous plants in the field. This study aims to evaluate the emergence and development of soursop seedlings in different substrates in protected environments. The experiment was conducted at the Mato Grosso do Sul State University and carried out using five protected environments: greenhouse, greenhouse with thermo-reflective screen, nursery with monofilament screen, nursery with thermo-reflective screen, and nursery with palm thatch. The substrates (S) consisted of cattle manure (M), humus (H), cassava branches (C), and vermiculite (V) as in the following ratios: S1 = H + V (1:3), S2 = H + V (1:1), S3 = H + V (3:1), S4 = H + C (1:3), S5 = H + C (1:1), S6 = H + C (3:1), S7 = M + V (1:3), S8 = M + V (1:1), S9 = M + V (3:1), S10 = M + C (1:3), S11 = M + C (1:1), S12 = M + C (3:1), S13 = H + M + V (1:1:1), S14 = H + M + C (1:1:1), and S15 = H + M + V + C (1:1:1:1). For the statistical analysis, each of those environments was considered as an experiment in which was used the completely randomized design; subsequently, it was performed a combined analysis of them. In summary, the greenhouse with thermo-reflective screen and combined substrates with "M + V" promote greater development of the seedlings. High concentrations of "V" or "C" cause no beneficial effect on soursop seedlings.

KEYWORDS: *Annona muricata*; cattle manure; cassava branches; vegetable ambience.

MUDAS DE GRAVIOLEIRA: EMERGÊNCIA E DESENVOLVIMENTO SOB DIFERENTES AMBIENTES DE CULTIVO E SUBSTRATOS – PARTE I

RESUMO: A qualidade da muda é fundamental para obtenção de plantas vigorosas no campo. Desta forma, o presente trabalho teve como objetivo avaliar a emergência e o desenvolvimento de mudas de gravioleira, em diferentes substratos, no interior de ambientes protegidos. O experimento foi conduzido na Universidade Estadual de Mato Grosso do Sul, onde foram empregados cinco ambientes protegidos: estufa agrícola; estufa agrícola com tela termorrefletora sob o filme; viveiro agrícola coberto com tela de monofilamento; viveiro agrícola coberto com tela termorrefletora e viveiro coberto com palha. Os substratos foram constituídos por esterco bovino (E), húmus (H), ramas de mandioca (M) e vermiculita (V), nas seguintes proporções: S1 = H + V (1:3); S2 = H + V (1:1); S3 = H + V (3:1); S4 = H + M (1:3); S5 = H + M (1:1); S6 = H + M (3:1); S7 = E + V (1:3); S8 = E + V (1:1); S9 = E + V (3:1); S10 = E + M (1:3); S11 = E + M (1:1); S12 = E + M (3:1); S13 = H + E + V (1:1:1); S14 = H + E + M (1:1:1); e S15 = H + E + V + M (1:1:1:1). Cada ambiente foi considerado um experimento, sendo adotado o delineamento inteiramente casualizado e, posteriormente, análise conjunta dos mesmos. Em síntese, a estufa agrícola com tela termorrefletora sob filme e os substratos combinados com "E + V" promovem maior desenvolvimento de mudas. Altas concentrações de "V" ou "M" provocam efeito não benéfico às mudas de gravioleira.

PALAVRAS-CHAVE: *Annona muricata*; esterco bovino; ramas de mandioca; ambiência vegetal.

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INTRODUCTION

The soursop (*Annona muricata* L.) belongs to the Annonaceae family and has been showing great economic potential in the Brazilian fruit scenario, especially in Northeast Brazil. Its fruits are destined for consumption *in natura* and mostly for the agro-industry due to low-technology orchards (COSTA et al., 2005).

Similarly to the increasing fruit demand is the formation of orchards. In this case, the production of quality seedlings is an adopted practice to the production of soursop since seedlings with aggregate qualities reflect on growth and survival in the field, especially under adverse conditions, in addition to provide better performance and productivity (BARBOSA et al., 2003).

The propagation of fruit seedlings in protected environments is widely used as it promotes benefits related to the protection and internal microclimate. However, these environments may have different configurations, directly influencing the quality of seedlings (COSTA et al., 2010). When comparing different protected environments for jatoba, COSTA et al. (2011c) observed that the greenhouse with low-density polyethylene (LDPE) has provided more vigorous seedlings than those produced in environments covered with black or thermal-reflective screen, both with 50% shading. For passion fruit, COSTA et al. (2009) observed that environments with monofilament or thermal-reflective screens, both with 50% shading, promoted better seedlings when compared to those produced in greenhouse with LDPE. For araticum, CAVALCANTE et al. (2008) observed that the full sun environment provided larger seedlings than that produced in greenhouse with LDPE.

The substrate is another factor that influences seedlings; physical characteristics, such as porosity, moisture retention and density, as well as chemical characteristics, such as nutrient content, pH and cation exchange capacity, are important parameters for its choice (SILVA & FARNEZI, 2009). Thus, the choice of the elements that compose the substrates is a crucial factor for the formation of soursop seedlings. According to LIMA et al. (2009), soursop seedlings cultivated in substrate composed of soil, humus and carbonized rice husk (2:1:1) present greater vigor when compared with those cultivated in substrates with a mixture of soil, carbonized rice husk, and coconut shell powder. OKUMURA et al. (2008) describe that organic components fertilized with mineral components improve the development of grafted seedlings of soursop. OLIVEIRA et al. (2009b) shows that the isolated green husk powder causes depressive effect on soursop seedlings.

Considering that in the literature there is a lack of research about the influence of protected environments and substrates for the formation of soursop seedlings, studies should be conducted in this area. Thus, this study aimed to evaluate the effect of protected environments and substrates on the seedlings production of *Annona muricata* L. in Aquidauana, MS, Brazil.

MATERIAL AND METHODS

The experiments for the formation of soursop seedlings were conducted in the experimental area of the Mato Grosso do Sul State University (UEMS), campus Aquidauana, MS, Brazil (geographical coordinates: 20°20' S and 55°48' W; average altitude: 174 m), from January to April, 2012. The regional climate is classified as tropical warm sub-humid with rainy summers and dry winters, with mean annual precipitation of 1200 mm and temperature of 29 °C.

It was used different cultivation environments: (E1) a greenhouse with galvanized steel structure, zenithal opening in the ridge, covered with a 150-µm low-density polyethylene film (LDPE), light diffuser, closures in the front and side with black monofilament screen with 50% shading, arched ceiling format, 8 m wide, 18 m length, and 4 m of ceiling height; (E2) a greenhouse with galvanized steel structure, zenithal opening in the ridge, covered with a 150-µm low-density polyethylene film (LDPE), light diffuser, closures in the front and side with black monofilament screen with 50% shading, arched ceiling format, 8 m wide, 18 m length, and 4 m of ceiling height, and thermal-reflective screen with 50% shading under the LDPE; (E3) a screened nursery with galvanized steel structure, 8 m wide, 18 m length, and 3.5 m of ceiling height, coverage and closures in the front and side in 45º with black monofilament screen with 50% shading (Sombrite®); (E4) a screened nursery with galvanized steel structure, 8 m wide, 18 m length, and 3.5 m of ceiling height, coverage and closures in the front and side in 45º with aluminized thermal-reflective screen with 50% shading (Aluminet®); and (E5) a nursery with a wooden structure, 3 m wide, 1.2 m length, and 1.8 m of ceiling height, covered with a native palm thatch regionally known as bacuri, and without front and side closures.

The soursop fruits were collected from trees in the region of Aquidauana, MS, Brazil from November to December, 2011. The seeds were separated from the fruit pulp, washed in running water, and shade dried for three days. In order to release seed dormancy, the seeds were immersed in water at 25 °C for 24 hours. The cassava branches were triturated in a hammer mill (model TRF 650, TRAPP, Jaraguá do Sul, SC, Brazil) using an 8-mm mesh sieve; subsequently, it was placed in open-air on a canvas for composting during 60 days, being wet daily and turned completely inside out and upside down each 2 days. In addition, it was used vermiculite with medium texture, and both cattle manure (composted during 30 days) and earthworm humus were obtained in the region of Aquidauna and Dois Irmãos do Buriti, MS, respectively.

The sowing was carried out on January 6, 2012 in polyethylene bags in which were placed three seeds per container in the depth of 1–3 cm. After 42 days after sowing (DAS), it was performed the polish out when the seedlings presented two definitive leaves, keeping one plant per container.

Inside the protected environments, the plants were placed in polyethylene bags (15.0 × 21.5 cm) with 1.6 liter capacity and filled with substrates derived from combinations of earthworm humus (H), cattle manure (M), vermiculite (V) and cassava branches (C) (Table 1). The respective chemical analyses and density of the substrates (1970) are shown, respectively, in Tables 2 and 3.

TABLE 1. Substrates (S) derived from different mixture ratios of humus (H), cattle manure (M), vermiculite (V) and cassava branches (C).

Humus (H) + Vermiculite (V)	Humus (H) + Cassava Branches (C)
S1 = 25% H + 75% V;	S4 = 25% H + 75% C;
S2 = 50% H + 50% V;	S5 = 50% H + 50% C;
S3 = 75% H + 25% V.	S6 = 75% H + 25% C.
Cattle Manure (M) + Vermiculite (V)	Cattle Manure (M) + Cassava Branches (C)
S7 = 25% M + 75% V;	S10 = 25% M + 75% C;
S8 = 50% M + 50% V;	S11 = 50% M + 50% C;
S9 = 75% M + 25% V.	S12 = 75% M + 25% C.
Humus (H) + Cattle Manure (M) + Vermiculite (V)	
S13 = 33.3% H + 33.3% M + 33.3% V	
Humus (H) + Cattle Manure (M) + Cassava Branches (C)	
S14 = 33.3% H + 33.3% M + 33.3% C	
Humus (H) + Cattle Manure (M) + Vermiculite (V) + Cassava Branches (C)	
S15 = 25% H + 25% M + 25% de V + 25% C	

TABLE 2. Chemical analysis of the substrates organic materials used in the treatments.

	----- $g\ kg^{-1}$ -----							
*	N	P	K	Ca	Mg	S	C	OM
M	10.60	3.66	1.00	9.80	1.65	1.81	96.50	166.00
H	14.80	4.46	1.00	26.70	12.50	3.53	163.00	281.00
C	19.50	2.89	7.00	18.80	6.15	2.42	376.00	647.00
	-	-	-		----- $mg\ kg^{-1}$ -----			
	pH	U	C/N	Cu	Zn	Fe	Mn	B
M	6.50	2.86	9.10	17.50	75.00	7800.00	310.00	11.47
H	6.90	13.46	11.01	30.00	130.00	14800.00	370.00	14.40
C	7.20	11.23	19.28	20.50	87.50	3440.00	520.00	20.70

*Solanalise soil analysis laboratory, Cascavel, PR, Brazil. OM = organic matter; U = moisture, in % at 65 °C; M = cattle manure; H = earthworm humus; C = cassava branches; C/N = carbon to nitrogen ratio.

TABLE 3. Wet and dry densities of the substrates.

	Density ($kg.m^{-3}$)							
	S1	S2	S3	S4	S5	S6	S7	S8
W	791.11	958.74	1104.50	931.50	1146.17	1229.06	648.14	741.17
D	279.83	479.71	500.62	273.65	502.11	648.20	209.47	270.36
	S9	S10	S11	S12	S13	S14	S15	-
W	838.14	652.16	700.27	816.03	956.67	766.94	988.09	-
D	399.47	216.58	244.08	376.33	442.97	289.88	457.02	-

W = wet density; D = dry density; S1 = 25% H + 75% V; S2 = 50% H + 50% V; S3 = 75% H + 25% V; S4 = 25% H + 75% C; S5 = 50% H + 50% C; S6 = 75% H + 25% C; S7 = 25% M + 75% V; S8 = 50% M + 50% V; S9 = 75% M + 25% V; S10 = 25% M + 75% C; S11 = 50% M + 50% C; S12 = 75% M + 25% C; S13 = 33,3% H + 33,3% M + 33,3% V; S14 = 33,3% H + 33,3% M + 33,3% C; S15 = 25% H + 25% M + 25% V + 25% C.

Each cultivation environment was considered as an experiment because there was no repetition of them. In each environment was adopted a completely randomized design with six replications of five plants each. Initially, the data were submitted to the analysis of individual variances of the substrate; then the evaluation of the mean square error and the combined analysis of the experiments were performed (BANZATTO & KRONKA, 2006). Statistical calculation was carried out by using the software Sisvar 5.3 (FERREIRA, 2010) and the averages submitted to the F-test and compared by the Scott-Knott test at 5% probability.

After sowing and the beginning of seedling emergence, it was evaluated the emergence speed index (ESI) and the mean germination time (MGT). The data were collected daily from 29 January, 2012 (23 DAS) to 26 February, 2012 (51 DAS) and subsequently transformed according to the equation $\sqrt{x+0.5}$. Seedling height was measured by using a millimetric ruler every fifteen days from the polish out; the number of leaves was evaluated at 55, 70, 85 and 100 DAS. In addition, the temperatures of dry and wet bulb were measured daily at 9:00, 12:00 and 15:00 h in each cultivation environment during all the experiment time, and the relative humidity was determined with the software Psychrometric Function Demo (Table 4).

TABLE 4. Average temperature (°C) and relative humidity (%) at 9:00, 12:00 and 15:00 h for each cultivation environment (E) during the experiment, from January 7 to April 4, 2012.

*	DBT	WBT	DBT	WBT	DBT	WBT	RH		
			°C					%	
	9 h		12 h		15 h		9 h	12 h	15 h
E1	30.2	25.5	33.3	26.4	33.3	26.1	68.9	58.5	57.0
E2	28.6	24.4	32.8	25.8	33.2	25.9	71.1	57.6	56.4
E3	29.5	24.7	34.1	26.2	34.6	26.3	67.9	54.0	52.4
E4	28.1	24.5	32.7	26.2	32.7	26.2	74.7	60.2	60.2
E5	27.8	24.6	31.6	26.0	31.9	25.9	77.2	64.5	62.4

*DBT = dry bulb temperature (ºC); WBT = wet bulb temperature (ºC); RH = relative humidity (%); E1 = greenhouse covered with a low-density polyethylene (LDPE) film; E2 = greenhouse covered with a LDPE and aluminized thermal-reflective screen with 50% shading under the film; E3 = screened nursery covered with black monofilament screen with 50% shading; E4 = screened nursery covered with aluminized thermal-reflective screen with 50% shading; E5 = cultivation environment covered with a native palm thatch regionally known as bacuri.

RESULTS AND DISCUSSION

In order to carry out the combined analysis of experiments and compare the cultivation environments, the division between the highest and lowest mean square error of the individual analyses of variance of treatments (substrates) within the environments cannot exceed the approximate ratio of 7:1 (BANZATTO & KRONKA, 2006). For the studied variables, this ratio was lower than 7:1, allowing carrying out the combined analysis of the experiments and comparing the cultivation environments (Table 5).

TABLE 5. Analysis of variance with calculated F, coefficient of variation, and the relation between the minimum and maximum mean square error for the emergence speed index (ESI), mean germination time (MGT), seedling height (SH) and number of leaves (NL) at 55 (SH1; NL1), 70 (SH2; NL2), 85 (SH3; NL3) and 100 (SH4; NL4) days after the soursop sowing.

	ESI	MGT	SH1	SH2	SH3
Environment	162.3**	15.0**	199.6**	253.5**	220.3**
Substrate	32.9**	2.9**	5.9**	8.8**	11.6**
Interaction	3.9**	1.8**	2.0**	1.9**	2.0**
RMSE	1.89	5.15	2.63	1.99	2.83
CV (%)	15.3	1.57	9.0	10.8	10.7
	SH4	NL1	NL2	NL3	NL4
Environment	146.4**	22.8**	56.8**	44.6**	13.7**
Substrate	11.1**	3.2**	6.5**	6.8**	3.8**
Interaction	2.1**	1.5*	1.9**	1.9**	1.7**
RMSE	3.03	2.72	3.34	5.58	4.47
CV (%)	10.45	13.5	12.9	11.6	11.9

^{NS} = Not significant; * = Significant at 5%; ** = Significant at 1%; CV = Coefficient of variation; RMSE = Relation between the minimum and maximum mean square error of the different cultivation environment.

It was observed interaction between the environments and substrates for all variables by means of the F test (Table 5). In the interaction between substrates within the environments for ESI, the substrates that showed higher performance were those that contained at least 50% of vermiculite in their composition (S1, S7 and S8), promoting greater ESI in all cultivation environments (Table 6). Although the seedlings of the substrate S1 have showed higher ESI, in most of the measurements the plants exhibited the lowest heights (Tables 9 and 10).

It is possible that the substrates S1 and S7 may have contributed to the increasing ESI by promoting better physicochemical conditions and less resistance to emergence due to their weight since they were constituted with large proportions of vermiculite (75%). In fact, FERREIRA et al. (2010) referred to vermiculite as the responsible for the greater ESI observed in *Rollinia mucosa* due to the greater balance between moisture and aeration of the substrate. In this sense, SILVA et al. (2009) showed that lightweight materials, such as carbonized rice husk, provided greater ESI in mangabeira. The proportion of vermiculite present in the substrate S2 (50%) also provided appropriate physical conditions for the soursop emergence, as well as the substrate S9 with a high percentage of cattle manure (75%) since the organic matter has the property of promoting greater water adsorption.

Among the environments, the greenhouse without thermal-reflective screen (E1) provided low ESI for all substrates (Table 6), which differs from the results obtained by SASSAQUI et al. (2013) in tamarind seedlings. On the other hand, the greenhouse with thermal-reflective screen (E2) promoted higher ESI (Table 6). SANTOS et al. (2011) compared a greenhouse with thermal-reflective screen (50% shading) under the polyethylene film (I) and a screened nursery with monofilament screen with 50% shading (II), and observed a higher ESI in the environment I to *Hymenaea stigonocarpa* Mart. The environment E2 led to lower temperatures and higher relative humidity when compared to the environment E1 (Table 2); these conditions favored the emergence of soursop in the environment E2 as observed by OLIVEIRA et al. (2009a) in seedlings of *Copernicia hospita* Martius, reporting also that the greater seedling emergences may be related to lower temperatures and higher relative humidity.

TABLE 6. Interaction between environments and substrates (E × S) for the emergence speed index (ESI) and mean germination time (MGT) of soursop.

**	E1	E2	E3	E4	E5
			IVE		
S1	1.13 Ac*	1.88 Ba	1.70 Ab	1.65 Ab	2.09 Aa
S2	0.90 Bc	1.48 Cb	1.77 Aa	1.88 Aa	1.84 Ba
S3	0.75 Bb	1.07 Db	1.02 Db	0.91 Cb	1.32 Ca
S4	0.68 Bb	1.80 Ba	1.71 Aa	1.70 Aa	1.80 Ba
S5	0.72 Bc	1.74 Ba	1.48 Bb	1.38 Bb	1.54 Cb
S6	0.80 Bb	1.42 Ca	1.05 Db	1.21 Ba	1.28 Ca
S7	0.98 Ad	2.27Aa	1.88 Ab	1.61 Ac	2.05 Aa
S8	1.33 Ad	2.21 Aa	1.88 Ab	1.67 Ac	2.01 Ab
S9	1.22 Ac	2.04 Aa	1.61 Bb	1.77Ab	1.84 Bb
S10	1.11 Ab	1.71 Ba	0.97 Dc	0.86 Cc	1.29 Cb
S11	0.76 Bc	1.57 Ca	1.70 Aa	1.30 Bb	1.52 Ca
S12	1.05 Ac	1.67 Ba	1.38 Cb	1.29 Bb	1.42 Cb
S13	1.00 Ad	1.88 Ba	1.45 Bc	1.63 Ab	2.03 Aa
S14	1.17 Ab	1.79 Ba	1.75 Aa	1.75 Aa	1.69 Ba
S15	1.14 Ac	1.87 Ba	1.21 Cc	1.39 Bb	1.52 Cb
*	E1	E2	E3	E4	E5
			TME		
S1	39.03 Ba*	39.69 Aa	39.15 Aa	38.91 Ba	39.43 Aa
S2	39.12 Ba	39.08 Aa	39.62 Aa	39.05 Ba	39.55 Aa
S3	38.47 Cb	39.28 Aa	39.20 Aa	38.66 Bb	39.60 Aa
S4	37.89 Cb	39.26 Aa	39.30 Aa	39.78 Aa	39.65 Aa
S5	38.56 Cb	39.43 Aa	39.50 Aa	39.93 Aa	39.74 Aa
S6	39.02 Bb	39.01 Ab	38.99 Ab	39.49 Aa	39.89 Aa
S7	39.51 Aa	39.17 Aa	39.87 Aa	39.66 Aa	39.81 Aa
S8	40.02 Aa	39.38 Aa	39.67 Aa	39.81 Aa	39.72 Aa
S9	39.65 Ab	39.39 Ab	40.10 Aa	39.22 Bb	40.19 Aa
S10	39.89 Aa	39.52 Ab	39.19 Ab	38.94 Bb	40.13 Aa
S11	38.78 Cb	39.55 Aa	39.41 Aa	39.08 Bb	39.95 Aa
S12	39.57 Aa	39.15 Aa	39.17 Aa	39.46 Aa	39.97 Aa
S13	39.13 Ba	39.56 Aa	39.57 Aa	39.41 Aa	39.84 Aa
S14	39.42 Ba	39.14 Aa	39.52 Aa	39.49 Aa	39.87 Aa
S15	39.23 Bb	39.04 Ab	39.64 Aa	39.26 Bb	40.02 Aa

*Means followed by the same uppercase letters on columns and lowercase letters on rows do not differ by the Scott-Knott test at 5% probability; **E1 = greenhouse; E2 = greenhouse with a thermal-reflective screen under the film; E3 = screened nursery with monofilament screen; E4 = screened nursery with thermal-reflective screen; E5 = cultivation environment with native palm thatch. S1 = 25% H + 75% V; S2 = 50% H + 50% V; S3 = 75% H + 25% V; S4 = 25% H + 75% C; S5 = 50% H + 50% C; S6 = 75% H + 25% C; S7 = 25% M + 75% V; S8 = 50% M + 50% V; S9 = 75% M + 25% V; S10 = 25% M + 75% C; S11 = 50% M + 50% C; S12 = 75% M + 25% C; S13 = 33,3% H + 33,3% M + 33,3% V; S14 = 33,3% H + 33,3% M + 33,3% C; S15 = 25% H + 25% M + 25% V + 25% C.

The ESI varied from 37.89 (environment E1, substrate S4) to 40.19 days (environment E5, substrate S9), with an overall average of 39.42 days, showing small difference between substrates and environments. OLIVEIRA et al. (2005) observed that the temperature causes effect both on the percentage of germination and germination speed index in *Annona montana*; also, the optimum temperature found for germination is around 30 °C, as can be observed in this study (Table 4).

As regards the number of leaves, there were practically no great variations of the substrates within the environments E1 and E2 in the four countings (NL1, NL2, NL3 and NL4); in general, the substrates containing humus in their composition, especially S1, S2 and S3, provided the lowest average of leaves per plant (Tables 7 and 8). Inside the nurseries (E3 and E4), there was difference among the substrates only in the intermediate countings (NL2 and NL3) in which, in general, plants cultivated on substrates containing proportions of vermiculite and manure (S7, S8 and S9) presented a higher number of leaves. Low concentrations of cassava branches associated with humus + cattle manure (S14) or only with manure (S11) favored the seedlings in those nurseries (Tables 7 and 8).

Perhaps the low percentage of cassava branches (33.3% and 25%) in the substrates S14 and S15 has not interfered with the availability of nutrients for the seedlings, particularly potassium, which can be found in the concentration of 7 g kg^{-1} in cassava branches (Table 2). According to Barbosa et al. (2003), potassium is one of the nutrients required in higher proportion to the aerial part of soursop seedlings up to 195 days after the polish out.

For the nursery with palm thatch (E5), the substrates S1, S2 and S3 promoted the lowest average number of leaves at 100 DAS (NL4) (Table 8). Combinations of cassava branches and cattle manure (S10, S11 and S12) provided a negative effect since, in general, seedlings cultivated in these substrates had lower average number of leaves for the four countings (Tables 7 and 8).

TABLE 7. Interaction between environments and substrates (E × S) for the number of leaves (NL) of soursop at 55 and 70 DAS.

**	E1	E2	E3	E4	E5
			NL1 (55 DAS)		
S1	3.06 Aa*	3.50 Aa	3.24 Aa	3.27 Aa	3.33 Ba
S2	2.99 Ab	3.63 Aa	3.52 Aa	3.88 Aa	3.76 Aa
S3	3.35 Aa	3.17 Aa	3.00 Aa	3.08 Aa	3.14 Ba
S4	2.63 Bb	3.81 Aa	3.49 Aa	3.26 Aa	3.48 Aa
S5	2.21 Bb	3.60 Aa	2.99 Aa	3.26 Aa	3.18 Ba
S6	3.11 Ab	3.83 Aa	3.11 Ab	3.14 Ab	3.38 Ab
S7	3.17 Aa	3.47 Aa	3.47 Aa	3.26 Aa	3.45 Aa
S8	3.22 Ab	4.17 Aa	3.26 Ab	3.53 Ab	3.83 Aa
S9	3.25 Aa	3.60 Aa	3.04 Aa	3.43 Aa	3.42 Aa
S10	3.15 Aa	3.42 Aa	2.97 Aa	3.03 Aa	3.16 Ba
S11	3.00 Ab	3.79 Aa	3.32 Ab	3.33 Ab	2.97 Bb
S12	3.00 Ab	3.72 Aa	3.44 Aa	3.08 Ab	3.24 Bb
S13	2.83 Ab	4.05 Aa	3.34 Ab	3.31 Ab	3.30 Bb
S14	3.19 Ab	3.67 Aa	3.73 Aa	3.53 Aa	2.97 Bb
S15	3.39 Aa	3.64 Aa	3.18 Aa	3.45 Aa	3.55 Aa
			NL2 (70 DAS)		
S1	3.43 Bb	5.13 Aa	4.59 Ba	4.88 Aa	5.33 Aa
S2	4.11 Ab	5.12 Aa	4.70 Bb	5.68 Aa	5.20 Aa
S3	2.79 Bb	4.49 Aa	4.13 Ba	4.44 Ba	4.79 Ba
S4	3.80 Ab	5.15 Aa	4.67 Ba	5.38 Aa	5.18 Aa
S5	3.35 Bb	5.41 Aa	4.63 Ba	5.31 Aa	5.02 Aa
S6	4.14 Ab	4.98 Aa	4.28 Bb	5.16 Aa	4.44 Bb
S7	4.04 Ab	5.44 Aa	5.13 Aa	5.06 Aa	5.43 Aa
S8	4.45 Ab	5.50 Aa	5.28 Aa	4.80 Ab	5.47 Aa
S9	4.28 Ab	5.37 Aa	4.97 Aa	5.10 Aa	5.54 Aa
S10	4.07 Ab	5.01 Aa	4.32 Bb	3.39 Cc	4.38 Bb
S11	4.08 Ab	5.27 Aa	5.11 Aa	4.65 Aa	4.77 Ba
S12	4.18 Ab	5.29 Aa	5.60 Aa	4.29 Bb	4.98 Aa
S13	3.81 Ab	5.35 Aa	5.03 Aa	4.97 Aa	4.87 Ba
S14	4.18 Ab	5.43 Aa	5.49 Aa	5.15 Aa	5.10 Aa
S15	4.56 Aa	5.48 Aa	4.76 Ba	4.75 Aa	5.14 Aa

*Means followed by the same uppercase letters on columns and lowercase letters on rows do not differ by the Scott-Knott test at 5% probability; **E1 = greenhouse; E2 = greenhouse with a thermal-reflective screen under the film; E3 = screened nursery with monofilament screen; E4 = screened nursery with thermal-reflective screen; E5 = cultivation environment with native palm thatch. S1 = 25% H + 75% V; S2 = 50% H + 50% V; S3 = 75% H + 25% V; S4 = 25% H + 75% C; S5 = 50% H + 50% C; S6 = 75% H + 25% C; S7 = 25% M + 75% V; S8 = 50% M + 50% V; S9 = 75% M + 25% V; S10 = 25% M + 75% C; S11 = 50% M + 50% C; S12 = 75% M + 25% C; S13 = 33,3% H + 33,3% M + 33,3% V; S14 = 33,3% H + 33,3% M + 33,3% C; S15 = 25% H + 25% M + 25% V + 25% C.

The lowest number of leaves observed for the substrates S1, S2 and S3 may be correlated to the earthworm humus in their composition since the chemical analysis showed high zinc content compared to the other components (Table 2). According to SILVA & FARNEZI (2009), zinc at concentrations above those required by soursop can promote toxicity or nutritional imbalance.

Considering the environments within the substrates, there was no difference for the substrates S1, S3, S7, S9, S10 and S15 at 55 DAS, S15 at 70 DAS, S4, S8 and S14 at 85 DAS and S2, S4, S8, S10, S13, S14 and S15 at 100 DAS (Tables 7 and 8). In the other substrates, the greenhouse with thermal-reflective screen under the polyethylene film (E2) promoted plants with higher number of leaves in the four samples (55, 70, 85 and 100 DAS). The greenhouse without the thermal-reflective screen under the polyethylene film (E1) did not favor leaves production and in generally promoted the lowest counts in the four samples. The seedlings cultivated in the nurseries E3, E4 and E5 showed similar results to that found for the environment E2 (Tables 7 and 8).

It is possible that the different responses to the number of leaves are due to changes in physiological processes, respiration and photosynthesis, which are due to changes in the microclimate of the environment caused by variations in the configuration of their coverage (Costa et al., 2011b).

TABLE 8. Interaction between environments and substrates (E × S) for the number of leaves (NL) of soursop at 85 and 100 DAS.

**	E1	E2	E3	E4	E5
			NL3 (85 DAS)		
S1	4.31 Cb*	6.17 Ba	5.66 Ba	5.69 Ba	6.58 Aa
S2	5.12 Bb	6.26 Ba	6.16 Aa	6.93 Aa	6.64 Aa
S3	4.19 Cc	5.42 Bb	5.44 Bb	5.34 Bb	6.39 Aa
S4	5.75 Aa	6.51 Aa	6.33 Aa	6.26 Aa	6.39 Aa
S5	5.44 Ab	7.04 Aa	6.25 Aa	6.29 Aa	6.39 Aa
S6	5.90 Ab	6.96 Aa	6.06 Ab	5.97 Bb	5.85 Bb
S7	5.38 Ab	6.83 Aa	6.80 Aa	6.31 Aa	6.57 Aa
S8	6.49 Aa	7.17 Aa	6.57 Aa	6.73 Aa	7.13 Aa
S9	5.50 Ab	7.13 Aa	5.97 Ab	6.30 Aa	6.49 Aa
S10	5.26 Ab	6.93 Aa	5.53 Bb	5.81 Bb	5.28 Bb
S11	4.92 Bc	7.10 Aa	6.02 Ab	6.28 Ab	5.87 Bb
S12	5.58 Ab	6.80 Aa	6.35 Aa	5.87 Bb	5.88 Bb
S13	5.11 Bb	7.19 Aa	5.88 Bb	6.49 Aa	6.53 Aa
S14	5.85 Aa	6.67 Aa	6.16 Aa	6.53 Aa	6.58 Aa
S15	5.75 Ab	7.02 Aa	5.51 Bb	6.13 Ab	6.33 Ab
			NL4 (100 DAS)		
S1	5.43 Bb	6.57 Aa	6.71 Aa	6.94 Aa	7.30 Ba
S2	6.94 Aa	7.20 Aa	6.87 Aa	7.68 Aa	7.28 Ba
S3	5.47 Bb	7.07 Aa	6.90 Aa	6.60 Aa	7.38 Ba
S4	7.08 Aa	8.03 Aa	7.37 Aa	6.97 Aa	7.67 Aa
S5	7.03 Ab	7.33 Ab	7.19 Ab	6.79 Ab	8.21 Aa
S6	7.80 Aa	7.74 Aa	7.11 Ab	6.62 Ab	7.74 Aa
S7	6.05 Bb	7.51 Aa	8.00 Aa	7.33 Aa	7.83 Aa
S8	7.78 Aa	8.40 Aa	7.53 Aa	7.43 Aa	8.10 Aa
S9	6.85 Ab	7.40 Ab	7.55 Ab	6.83 Ab	8.34 Aa
S10	7.06 Aa	7.79 Aa	7.19 Aa	7.25 Aa	6.62 Ba
S11	6.83 Ab	8.09 Aa	7.62 Aa	6.98 Ab	7.03 Bb
S12	6.73 Ab	8.16 Aa	8.46 Aa	7.12 Ab	6.86 Bb
S13	6.69 Aa	7.47 Aa	7.28 Aa	7.19 Aa	6.97 Ba
S14	6.95 Aa	7.60 Aa	7.49 Aa	7.07 Aa	8.16 Aa
S15	7.49 Aa	7.48 Aa	7.17 Aa	6.48 Aa	7.55 Aa

*Means followed by the same uppercase letters on columns and lowercase letters on rows do not differ by the Scott-Knott test at 5% probability; **E1 = greenhouse; E2 = greenhouse with a thermal-reflective screen under the film; E3 = screened nursery with monofilament screen; E4 = screened nursery with thermal-reflective screen; E5 = cultivation environment with native palm thatch. S1 = 25% H + 75% V; S2 = 50% H + 50% V; S3 = 75% H + 25% V; S4 = 25% H + 75% C; S5 = 50% H + 50% C; S6 = 75% H + 25% C; S7 = 25% M + 75% V; S8 = 50% M + 50% V; S9 = 75% M + 25% V; S10 = 25% M + 75% C; S11 = 50% M + 50% C; S12 = 75% M + 25% C; S13 = 33,3% H + 33,3% M + 33,3% V; S14 = 33,3% H + 33,3% M + 33,3% C; S15 = 25% H + 25% M + 25% V + 25% C.

For the variable seedling height, seedlings cultivated on substrates composed of materials with a high percentage of vermiculite or cassava branches (S1, S4 and S7) showed overall lower average at 55, 70, 85 and 100 DAS within the environment E1. In the environment E2, in general, only the combinations of humus and vermiculite (S1, S2 and S3) provided lower average height (Tables 9 and 10).

The lower heights due to the substrates S1, S4 and S7 may be associated with nutritional insufficiency caused by high concentration of vermiculite (S1 and S7) and cassava branches (S7), all with the proportion of 75%. COSTA et al. (2005) observed that the substrates containing vermiculite or vermiculite + carbonized rice husk provided soursop rootstock with low development since it is likely that these components are exempt from appropriate nutritional content for the plant development.

In the environments E3 and E4, the substrates formulated with cattle manure (from S7 to S15), in general, promoted higher average plant height, indicating that this material may be suitable for the formation of seedlings. Soursop propagated in substrates with cattle manure and vermiculite (S7, S8 and S9) presented higher average height in the environment E5 (Tables 9 and 10).

TABLE 9. Interaction between environments and substrates (E × S) for the seedling height (SH) of soursop at 55 and 70 DAS.

**	E1	E2	E3	E4	E5
			SH1 (55 DAS)		
S1	7.53 Bb*	11.71 Aa	11.34 Aa	10.65 Ba	10.54 Aa
S2	8.71 Ac	12.25 Aa	12.07 Aa	11.10 Ab	11.03 Ab
S3	7.51 Bb	10.69 Aa	10.97 Ba	10.55 Ba	9.83 Aa
S4	7.69 Bb	11.83 Aa	11.65 Aa	10.93 Ba	10.83 Aa
S5	7.55 Bc	12.95 Aa	11.94 Aa	10.66 Bb	10.84 Ab
S6	9.04 Ab	12.06 Aa	10.96 Ba	11.84 Aa	10.98 Aa
S7	8.16 Bc	12.11 Aa	11.57 Aa	10.54 Bb	11.08 Ab
S8	9.39 Ac	12.96 Aa	11.57 Ab	11.50 Ab	11.46 Ab
S9	8.94 Ab	12.08 Aa	11.55 Aa	11.77 Aa	11.15 Aa
S10	8.41 Ac	12.25 Aa	9.90 Bb	9.49 Cb	10.11 Ab
S11	7.28 Bc	12.69 Aa	11.50 Ab	10.94 Bb	10.87 Ab
S12	8.79 Ac	11.83 Aa	12.03 Aa	9.47 Cc	10.24 Ab
S13	8.17 Bc	12.21 Aa	11.71 Aa	11.72 Aa	10.37 Ab
S14	8.57 Ab	12.05 Aa	12.39 Aa	11.92 Aa	11.33 Aa
S15	9.87 Ab	12.78 Aa	10.88 Bb	11.69 Aa	10.49 Ab
			SH2 (70 DAS)		
S1	9.37 Bc	16.77 Aa	14.40 Bb	13.64 Cb	13.78 Bb
S2	9.88 Bc	17.13 Aa	16.18 Aa	14.82 Bb	14.56 Ab
S3	9.39 Bc	14.45 Ba	15.53 Aa	12.98 Cb	12.87 Bb
S4	8.59 Bc	17.46 Aa	16.70 Aa	14.82 Bb	13.93 Bb
S5	9.77 Bc	18.81 Aa	16.54 Ab	14.63 Bb	14.98 Ab
S6	11.40 Ac	17.75 Aa	15.10 Bb	15.17 Bb	14.84 Ab
S7	10.18 Bc	17.63 Aa	16.62 Aa	14.72 Bb	15.33 Ab
S8	12.13 Ac	19.36 Aa	17.12 Ab	16.43 Ab	16.64 Ab
S9	10.80 Bc	18.04 Aa	15.64 Ab	16.66 Aa	14.55 Ab
S10	10.56 Bc	17.39 Aa	13.63 Bb	13.33 Cb	12.95 Bb
S11	8.70 Bc	17.96 Aa	15.70 Ab	14.51 Bb	14.43 Ab
S12	10.44 Bc	17.14 Aa	16.44 Aa	12.66 Cb	13.41 Bb
S13	10.60 Bc	18.39 Aa	15.84 Ab	15.90 Ab	15.03 Ab
S14	11.50 Ac	17.49 Aa	16.75 Aa	16.85 Aa	15.05 Ab
S15	13.13 Ac	18.92 Aa	14.19 Bc	16.34 Ab	13.77 Bc

*Means followed by the same uppercase letters on columns and lowercase letters on rows do not differ by the Scott-Knott test at 5% probability; **E1 = greenhouse; E2 = greenhouse with a thermal-reflective screen under the film; E3 = screened nursery with monofilament screen; E4 = screened nursery with thermal-reflective screen; E5 = cultivation environment with native palm thatch. S1 = 25% H + 75% V; S2 = 50% H + 50% V; S3 = 75% H + 25% V; S4 = 25% H + 75% C; S5 = 50% H + 50% C; S6 = 75% H + 25% C; S7 = 25% M + 75% V; S8 = 50% M + 50% V; S9 = 75% M + 25% V; S10 = 25% M + 75% C; S11 = 50% M + 50% C; S12 = 75% M + 25% C; S13 = 33,3% H + 33,3% M + 33,3% V; S14 = 33,3% H + 33,3% M + 33,3% C; S15 = 25% H + 25% M + 25% V + 25% C.

The high concentration of cassava branches (75%) in the substrates S4 and S10 may have reduced the moisture retention capacity due to the higher percentage of macropores, causing low size seedlings. The increase of macropores may be related to the particle size of the branches; in fact, COSTA et al. (2011a) observed that more than 50% of milled cassava branches were retained in sieves with mesh of 2 mm.

According to BARBOSA et al. (2003), the soursop has low initial growth, with average fortnightly growth rates of 3.3 cm. In this study, from the beginning of the biweekly height measurements (55 DAS), the average increase of the seedlings were similar to those described by BARBOSA et al. (2003) in the substrates S6 and S15, with 3.02 and 3.09 cm in the environment E1. The substrate S8 provided average increase above 4.00 cm in the environments E2, E3, E4 and E5.

TABLE 10. Interaction between environments and substrates (E × S) for the seedling height (SH) of soursop at 85 and 100 DAS.

**	E1	E2	E3	E4	E5
			SH3 (85 DAS)		
S1	11.79 Bb*	18.87 Ba	17.73 Ba	17.09 Cb	16.77 Ba
S2	12.75 Bb	19.87 Ba	19.43 Ba	19.12 Ba	18.35 Ba
S3	11.48 Bb	16.34 Ca	18.13 Ba	16.05 Ca	16.58 Ba
S4	11.69 Bc	20.78 Aa	20.42 Aa	18.53 Bb	18.11 Bb
S5	12.56 Bc	22.08 Aa	20.94 Aa	18.74 Bb	18.18 Bb
S6	14.36 Ac	21.17 Aa	18.91 Bb	18.52 Bb	17.64 Bb
S7	12.44 Bb	20.83 Aa	21.70 Aa	19.58 Ba	20.08 Aa
S8	15.76 Ab	23.48 Aa	22.27 Aa	21.22 Aa	21.23 Aa
S9	12.94 Bb	21.38 Aa	20.53 Aa	21.39 Aa	19.58 Aa
S10	14.11 Ac	21.37 Aa	17.55 Bb	16.16 Cb	15.70 Bb
S11	11.98 Bd	22.83 Aa	20.34 Ab	18.83 Bc	17.45 Bc
S12	13.34 Bc	21.76 Aa	20.72 Aa	16.62 Cb	16.78 Bb
S13	12.14 Bc	22.69 Aa	19.93 Ab	20.02 Ab	18.54 Bb
S14	13.99 Ab	20.73 Aa	20.83 Aa	20.93 Aa	18.62 Ba
S15	15.73 Ac	22.80 Aa	18.28 Bc	19.98 Ab	17.56 Bc
			SH4 (100 DAS)		
S1	13.42 Bb	19.97 Ba	20.39 Ba	20.49 Ba	18.91 Ba
S2	16.74 Ab	21.16 Ba	20.92 Ba	22.29 Ba	20.37 Ba
S3	14.39 Bc	18.49 Bb	21.73 Ba	20.29 Ba	18.88 Bb
S4	14.64 Bb	23.19 Aa	22.58 Ba	21.76 Ba	20.89 Ba
S5	16.33 Ab	23.14 Aa	23.45 Aa	22.06 Ba	21.43 Ba
S6	18.10 Ac	23.96 Aa	21.39 Bb	20.56 Bb	21.28 Bb
S7	15.31 Bb	23.11Aa	25.55 Aa	23.07 Aa	23.49 Aa
S8	19.52 Ab	26.55 Aa	24.68 Aa	24.40 Aa	24.34 Aa
S9	15.86 Bb	23.66 Aa	24.03 Aa	24.03 Aa	23.34 Aa
S10	17.21 Ab	22.85 Aa	20.90 Ba	20.06 Ba	18.51 Bb
S11	14.48 Bc	25.53 Aa	23.17 Aa	23.45 Aa	20.41 Bb
S12	16.62 Ad	25.15 Aa	24.62 Aa	21.54 Bb	19.44 Bc
S13	15.04 Bc	24.24 Aa	22.19 Bb	23.52 Aa	20.81 Bb
S14	17.42 Ab	23.54 Aa	23.26 Aa	24.05 Aa	21.84 Aa
S15	19.16 Ab	24.34 Aa	20.09 Bb	22.88 Aa	20.33 Bb

*Means followed by the same uppercase letters on columns and lowercase letters on rows do not differ by the Scott-Knott test at 5% probability; **E1 = greenhouse; E2 = greenhouse with a thermal-reflective screen under the film; E3 = screened nursery with monofilament screen; E4 = screened nursery with thermal-reflective screen; E5 = cultivation environment with native palm thatch. S1 = 25% H + 75% V; S2 = 50% H + 50% V; S3 = 75% H + 25% V; S4 = 25% H + 75% C; S5 = 50% H + 50% C; S6 = 75% H + 25% C; S7 = 25% M + 75% V; S8 = 50% M + 50% V; S9 = 75% M + 25% V; S10 = 25% M + 75% C; S11 = 50% M + 50% C; S12 = 75% M + 25% C; S13 = 33,3% H + 33,3% M + 33,3% V; S14 = 33,3% H + 33,3% M + 33,3% C; S15 = 25% H + 25% M + 25% V + 25% C.

For environments within the substrates, in general, seedlings cultivated in the greenhouse with thermal-reflective screen under the polyethylene film (E2) exhibited greater heights in all substrates in the four measurements (SH1, SH2, SH3 and SH4). The nursery covered with monofilament screen (E3) provided results similar to those observed in the environment E2 at 100 DAS (SH4). The greenhouse without thermal-reflective screen (E1) promoted lower height within all substrates in all measurements (Tables 9 and 10).

Seedlings cultivated in environments which provided greater shading (E2, E3, E4 and E5) presented, in general, greater height when compared to those cultivated in the environment covered only with polyethylene (E1). The attenuation of light stimulates adaptive mechanisms and causes increased growth of the seedlings.

It is possible that the low average in the height of seedlings observed in the greenhouse covered only with polyethylene film (E1) is associated with fewer leaves and consequently lower leaf area and lower photosynthetic activity. As observed by CAMPOS et al. (2008), soursop plants containing more leaves showed greater height probably caused by more intense photosynthetic activity.

CONCLUSIONS

The greenhouse with thermal-reflective screen under the film provides proper development to the seedlings and it is the most appropriate for soursop.

Combinations of cattle manure and vermiculite favor the development of soursop seedlings.

High concentrations of vermiculite and cassava branches are not indicated for the development of soursop seedlings.

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