Water availability and substrate in the emergency and initial development of *Bauhinia scandens* L.[1]

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ABSTRACT

Bauhinia scandens is an ornamental plant and it has anti-tumor properties. The demand for seedlings of this species makes it necessary to know about the germinative behavior of seeds and the initial growth of seedlings. The objective of the present work was to determine the type of substrate and moistening conditions that are more favorable to the emergence and development of seedlings of *B. scandens*. *B. scandens* seeds were disposed on three substrates: sand, vermiculite, and commercial substrate, maintained at four levels of moistening 25, 50, 75, and 100% of the water holding capacity of the substrate and kept in a greenhouse for 60 days. The following parameters were evaluated: emergence, speed index, average time, number of leaves, length of aerial part and root, diameter of neck, fresh and dry mass of aerial part and root. The analysis of variance was performed in a completely randomized design, in a 3 X 4 factorial scheme, with four replications of 25 seeds. The sowing of *B. scandens* should be carried out initially in vermiculite with a holding capacity of 100% until the establishment of the seedling emergence and then transplanted to commercial substrate keeping between 50 and 75% holding capacity.

Keywords: seedlings; wetting; vine-bauhinia; ornamental plant; production.

INTRODUCTION

Bauhinia scandens L. Fabaceae is a climbing plant with ornamental value, nonetheless, still little known in Brazil (Bacher, 2018). However, the plant also gained prominence due to the medicinal properties of its leaves, from the isolation and identification of glycerol 1-O-alkyl, Hazra & Chatterjee (2008) established the antitumor property of this chemical compound through a bioassay, accepted internationally, called the Brine Shrimp toxicity test. In other species of the same genus such as *B. longifolia* L. and *B. acuruana Moric*. anticancer substances have also been identified (Góis *et al*., 2017; Aquino *et al*., 2019).

For the domestication and rational exploitation of the economic potential of plants, it is necessary to develop techniques for the commercial production of seedlings (Jellani *et al*., 2016). In the nursery, the substrate is one of the most important external factors from germination to seedling development (Dutra *et al*., 2012). An ideal substrate has desirable chemical, physical, and biological characteristics for the growth of the root system, ensuring the supply of water, nutrients and root support during the entire period of seedling formation (Alves *et al*., 2017).

Among the most used substrates for Fabaceae seedlings, the following stand out: organic compound added to the soil for *B. forficata* Link. and *B. variegata* L. (Duarte & Nunes, 2012; Krefta *et al.*, 2017); commercial substrate for *Copaifera langsdorffii* Desf. and *Plukenetia volubilis* L. (Jeromini *et al.*, 2017; 2018), and vermiculite for *Dalbergia miscolobium* Benth and *Plukenetia volubilis* L. (Moura *et al*., 2016; Jeromini *et al*., 2018).

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In addition to the substrate, water availability is considered one of the most important abiotic factors for seedling growth and production (Portes *et al.*, 2006). Water is essential for seed germination and for metabolic activities of plants (Padilha *et al*., 2016). Each plant species has a water requirement for its growth in nurseries, due to its morphological characteristics and adaptation to environments (Dresch *et al.*, 2016; Mota *et al.*, 2017).

The adaptation of species to environments with less water availability was reported by Scalon *et al.* (2014, 2011) for seedlings of *Eugenia pyriformis* Cambess, and *Guazuma ulmifolia* Lamarck showed greater growth when maintained at 50% of the water holding capacity of the substrate, probably because they are species of Cerrado. For *Caesalpinia ferrea* Benth. and *Alibertia edulis* (Rich.) A.Rich., the substrate should be maintained with 70% and 100% water holding capacity, respectively, probably due to the greater water needs of these species, due to the occurrence sites that vary from the Cerrado to the Atlantic Forest (Lenhard *et al*., 2010; Jeromini *et al*., 2019).

In view of this, we can assume that there is a difference in the initial growth of the seedlings of *B. scandens* when subjected to different conditions of moistening and substrate. The objective of the present work was to determine the type of substrate and conditions for its moistening that are more favorable to the emergence and development of seedlings of *B. scandens.*

MATERIAL AND METHODS

B. scandens fruits were harvested from 10 parent plants in Botucatu-SP and sent to the Seed Analysis Laboratory of the Plant Production Department at UNESP, Jaboticabal, SP. The seeds were processed and homogenized, then packed in Kraft paper bags and stored in a controlled environment (9 ºC), during 20 days, until the moment of the experiment installation, when the water content of 6 ± 1% was determined, through two repetitions of 10 seeds, using the greenhouse method at 105 ± 3 ºC for 24 h (Brasil, 2013).

Transparent plastic boxes (22 x 15 x 5 cm) were filled with the following substrates (still dry): fine sand (particles between 0.10 and 0.25 mm), medium vermiculite (particles between 1.19 and 0.50 mm), and commercial substrate. The commercial substrate used was Basaplant Florestais® formulated based on pine bark, fibrous peat, coconut fiber, vermiculite, NPK, and micronutrients (Baseagro, 2016).

Four subsamples of 25 seeds were sown in each type of substrate at 2.0 cm depth. After sowing, the substrates were moistened with four levels of water holding capacity, these being 25, 50, 75 and 100%. The water holding capacity was determined by adopting the methodology described by Souza *et al*. (2001) and Brasil (2013).

The boxes were kept for 60 days in a greenhouse with shading, temperature, and relative humidity of 50%, 28 ± 3 °C and 68 ± 3%, respectively. The maintenance of the substrate moistening in the different water holding capacities was obtained by daily weighing the boxes on a scale and replacing the water when necessary. The following quality parameters were evaluated:

Seedling emergence - normal seedlings emerged on the 26th day after sowing. It was considered as emerged normal seedling those that show itselves healthy, and with part of the hypocotyl visible outside the substrate (Jeromini *et al.*, 2015). The results were expressed as percentage (Brasil, 2013).

Speed index and average emergency time - conducted together with the emergency test, daily counting the number of normal seedlings emerged and applying the methodology and formula proposed by Maguire (1962) and Labouriau & Valadares (1976), respectively. For the mean time of emergence, the results were expressed in days.

The following parameters were evaluated 60 days after sowing:

Number of leaves - obtained by counting real and expanded leaves per plant.

Length of aerial part and root - evaluated by measuring the distance between the neck to the point of insertion of the last real leaf and the neck to the root cap, respectively, with the help of a millimeter ruler. The results were expressed in centimeters.

Stem diameter – it was measured close to the substrate with the aid of a digital caliper and the results were expressed in centimeters.

Fresh mass of aerial part and root - the plants were removed from the substrate, washed in running water and kept on the laboratory table until the surface water dried. The aerial part and the root were sectioned, separated, and weighed on a precision scale (0.0001 mg).

Dry mass of aerial part and root - the parts obtained in the previous test were packed separately in Kraft paper bags and placed to dry in an oven with forced air circulation at 65 ºC until constant weight. Then the materials were weighed on a precision scale (0.0001 mg).

The analysis of variance was performed in a completely randomized design, in a 3 X 4 factorial scheme (substrates x water levels), with four replications of 25 seeds. The experimental data were submitted to the normality test. After that they were submitted to the analysis of variance, when a significant effect was identified, the qualitative means of the treatments were compared using the Tukey test and when quantitative, they were evaluated using regression analysis ($p \leq 0.05$).

RESULTS AND DISCUSSION

In the summary of the analysis of variance, there was only no interaction for the length of aerial part and root, as well as fresh root mass, for which only the isolated factor substrate was significant (Table 1).

It was found that the moistening of the substrate in association with vermiculite for the variables percentage of emergence and speed of emergence of seedlings of *B. scandens* caused similar behaviors for both variables, with a linear increase in the values of these variables concomitantly with the increase of water availability, with a maximum percentage of emergence and a higher rate of seedling emergence speed, 72% and 1.10 at 100% holding capacity, respectively (Table 2). In contrast, the substrate sand caused linear decrease of these variables with the increase of the used holding capacity.

For emergence and emergence index of seedling from commercial substrate, they did not fit any regression model, as well as for the average seedling emergence time in sand (Table 2).

In the comparison between substrates, greater emergence and speed index were obtained when seeds kept in sand at 25 and 50% of the water holding capacity, not differing from those kept in commercial substrate at 25%. At 75% capacity, it was lower only in the emergency compared to other substrates; and at higher capacity, this substrate was inferior to the others, not differing from the commercial one.

Table 1: Analysis of variance of emergence (E), emergence speed index (ESI), average emergence time (AET), number of leaves (NL), aerial part length (APL) and root length (RL), neck diameter (ND), fresh weight of aerial part (FWAP), fresh weight of root (FWR), and total fresh weight (TFW), dry weight of aerial part (DWAP) and dry weight of root (DWR) of *Bauhinia scandens* seedlings submitted to different substrates (S) and water holding capabilities of substrate (WHCS)

Evaluated parameters	Variation factor			Means	m.s.d	CV (%)
	S	WHCS	S x WHCS			
	Mean square					
E (%)	176.31^{ns}	390.07**	1,394.70**	40.00	12.82	18.23
ESI	0.07^{ns}	0.09^{ns}	0.36**	0.52	0.32	35.61
AET (days)	81.08**	29.90**	12.97*	17.02	3.87	13.18
NL	19.59**	0.70*	0.90**	4.25	0.76	10.33
APL (cm)	5.83**	0.05^{ns}	0.14^{ns}	3.58	0.36	11.90
RL (cm)	8.50**	3.76^{ns}	3.66^{ns}	5.92	1.09	21.29
ND (cm)	0.24**	0.03*	0.03**	1.09	0.17	9.08
FWAP (g)	0.059**	0.004**	0.002*	0.123	0.047	22.34
FWR (g)	0.004**	0.0002^{ns}	0.0004^{ns}	0.045	0.012	32.54
DWAP(g)	0.001**	0.0001*	0.0001*	0.026	0.010	22.75
DWR (g)	1.5x10^{-5*}	0.2x10^{-5ns}	1.6x10^{-5**}	0.006	0.003	32.37

*, ** and ^{ns}= significant value at pd"0.05, pd"0.01 and not significant by the "F" test; CV = coefficient of variation.

Table 2: Emergence, average emergence time and emergence speed index of *Bauhinia scandens* seedlings as a function of different substrates and water holding capabilities

Substrates	Water holding capacity (%)				Equation	R^2
	25	50	75	100		
Seedling emergence (%)						
Sand	52 a	56 a	29 b	40 b	$\hat{Y}$ = -0.2630x + 60.8750	0.46
Vermiculite	20 b	21 b	45 a	72 a	$\hat{Y}$ = 0.7160x – 5.2500	0.90
Commercial	40 a	31 b	46 a	35 b	$\hat{Y}$ = did not adjust	-
Seedling emergence index						
Sand	0.64 a	0.77 a	0.41 a	0.39 b	$\hat{Y}$ = -0.0044x + 0.8263	0.61
Vermiculite	0.25 b	0.27 b	0.71 a	1.10 a	$\hat{Y}$ = 0.0119x – 0.1647	0.91
Commercial	0.41 ab	0.44 b	0.58 a	0.40 b	$\hat{Y}$ = did not adjust	-
Average emergence time of seedlings (days)						
Sand	17 a	18 b	19 b	17 a	$\hat{Y}$ = did not adjust	-
Vermiculite	16 a	14 a	13 a	15 a	$\hat{Y}$ = -0.0390x + 16.8750	0.79
Commercial	22 b	18 b	19 b	14 a	$\hat{Y}$ = -0.0930x + 24.3750	0.84

Means followed by the same lowercase letter in the column do not differ by Tukey's test at 5% probability.

When the average emergence time was evaluated, the vermiculite and commercial substrates provided significant effect in relation to the tested moistening, with a decrease in the time to emergence as there was an increase in water availability.

Regarding the emergence speed characterized by the speed index and average emergence time of seedlings, it was observed that the substrates sand and vermiculite in general favored the increase in speed in relation to the commercial substrate. There was no significant difference between the substrates in the moistening of 75% for the emergence speed index, and in 100% in the average germination time, with an average of 0.5666 and 15 days, respectively (Table 2).

For the aforementioned variables, which evaluate the initial development of seedlings, it was found that with the increase in water supply, there was reduction in the time of emergence, probably due to the high water requirement for the initial stages of the germination process. It was also observed for *Alibertia edulis* (Jeromini *et al.*, 2019) which, like *B. Scandens*, develops in tropical forests.

The greater water requirement associated with the lighter substrate, such as vermiculite, may have enabled greater gas exchange and better drainage, reducing the physical barrier for the initial seedling development (Maggioni *et al*., 2014; Silva *et al.*, 2016) as also observed for *Plukenetia volubilis* L. (Silva *et al*., 2016; Jeromini *et al*., 2018).

The commercial substrate provided superior performance for aerial part length, root length, and fresh root mass with maximum of 4.2 cm, 6.5 cm, and 0.064 g, respectively; it only did not differ from vermiculite in root length (Table 3).

Therefore, the commercial substrate does not provide ideal conditions for starting seedling development. However, it was responsible for the maximum development of the seedlings already emerged, possibly by means of the supply of macro and micro nutrients after the emergence of the seedlings and their mobilization to the aerial part and roots, as observed in *Eugenia uniflora* (Antunes *et al*., 2012) and *Plukenetia volubilis* (Jeromini *et al*., 2018).

The highest leaf production was observed when *B. scandens* seedlings were developed in a commercial substrate, not fitting a regression model, probably due to the similarity between holding capacities. While for sand and vermiculite, they adjusted to the linear model, however, with decreasing behavior for those kept in sand and increasing for those with vermiculite, with increase of the holding capacity (Table 4).

The moistening of 25 and 50% of water holding capacity of the substrate reduced leaf development. Gordin *et al*. (2016) observed in their review that since leaf expansion is a process controlled by cell turgor, under conditions of low water availability, there may be reduction in the quantity, expansion, and size of leaves.

Higher diameter values were obtained from seedlings from the commercial substrate, not differing from the other substrates in 50 and 75% of the holding capacity. In this variable, the sand substrate did not fit the regression models, while for the vermiculite substrate the maximum diameter values were observed in 51%; seedlings from the commercial substrate, on the other hand, it obtained a larger diameter at lower holding capacities with decrease in these values with increase in holding capacity.

The commercial substrate showed a different result to the factor diameter, providing greater development in the stem diameter in a condition of lower wetting. This phenomenon was observed when there is water reduction and, as a form of protection, there is a thickening of the stem to accumulate reserves (Taiz & Zeiger, 2013), as also observed in *Amburana cearenses* (Pimentel & Guerra, 2011) and *Parapiptadenia rigida* (Benth .) Brenan (Dutra *et al.*, 2016).

The accumulation of fresh mass of seedling was greater when kept in commercial substrate in all evaluated capacities. When the adjustment to the regression models for the sand substrate was evaluated, the maximum mass accumulation was observed in 55%; while the seedlings from the commercial substrate obtained greater accumulation of mass in the lower holding capacities with decrease in these values with the increase in the holding capacity, and the reverse for those maintained in vermiculite.

For dry matter of the aerial part, the commercial substrate caused the greatest mass accumulation,

Table 3: Aerial part length, root length, and fresh weight of root of *Bauhinia scanden*s seedlings in initial development submitted to different substrates

Substrate	Aerial part length	Root length	Fresh weight of root
	(cm)		(g)
Sand	3.6 b	5.1 b	0.032 b
Vermiculite	3.0 c	5.9 ab	0.036 b
Commercial	4.2 a	6.5 a	0.064 a

Means followed by the same lowercase letter in the column do not differ by Tukey’s test at 5% probability.

Table 4: Number of leaves, leaf width, fresh weight of aerial part, dry weight of aerial part, dry weight of root and total weight, and neck diameter of *Bauhinia scandens* seedlings in initial development submitted to different substrates and water holding capacities

Substrates	Water holding capacity (%)				Equation	R^2
	25	50	75	100		
Number of leaves						
Sand	4 b	3 b	3 c	3 c	$\hat{Y}$ = -0.0039x + 3.5668	0.93
Vermiculite	3 b	4 b	4 b	5 b	$\hat{Y}$ = 0.0186x + 2.8288	0.88
Commercial	5 a	6 a	6 a	6 a	$\hat{Y}$ = did not adjust	-
Stem diameter (mm)						
Sand	1.06 b	1.11 a	1.10 a	0.87 b	$\hat{Y}$ = did not adjust	-
Vermiculite	0.91 b	1.15 a	1.04 a	0.99 b	$\hat{Y} = -0.0002x^2 + 0.0204x+0.5188$	0.93
Commercial	1.36 a	1.16 a	1.22 a	1.19 a	$\hat{Y}$ = -0.0019x + 1.3488	0.45
Fresh weight of aerial part (mg)						
Sand	0.094 b	0.132 b	0.078 b	0.062 b	$\hat{Y} = -0.00002x^2+0.0022x+0.0601$	0.68
Vermiculite	0.054 b	0.095 b	0.086 b	0.106 b	$\hat{Y}$ = 0.0006x + 0.0483	0.71
Commercial	0.152 a	0.206 a	0.206 a	0.201 a	$\hat{Y}$ = -0.0007x + 0.1499x	0.66
Dry weight of aerial part (mg)						
Sand	0.023 a	0.033 a	0.019 b	0.019 b	$\hat{Y}$ = did not adjust	-
Vermiculite	0.012 b	0.017 b	0.022 b	0.023 b	$\hat{Y}$ = 0.0002x + 0.0089	0.93
Commercial	0.031 a	0.039 a	0.038 a	0.038 a	$\hat{Y}$ = did not adjust	-
Dry weight of root (mg)						
Sand	0.007 a	0.008 a	0.004 b	0.004 b	$\hat{Y}$ = -0.0001x + 0.0087	0.69
Vermiculite	0.004 a	0.004 a	0.007ab	0.008 a	$\hat{Y}$ = 0.0001x + 0.0022	0.89
Commercial	0.006 a	0.008 a	0.008 a	0.008 a	$\hat{Y}$ = did not adjust	-

Means followed by the same lowercase letter in the column do not differ by Tukey's test at 5% probability.

however, it did not differ from the sand in the moistening of 25 and 50% of the water holding capacity. However, these substrates did not obtain adjustment in the regression analysis of this variable, whereas for seedlings obtained in the vermiculite substrate, accumulation was verified with the increase of the holding capacity.

In the results of dry mass of roots, it was verified that when the seedlings were maintained at 25 and 50% of the water holding capacity, there was no difference between the tested substrates; whereas for 75 and 100%, the commercial substrate did not differ from vermiculite. In the adjustment to the regression models for the commercial substrate, probably due to similar values, there was no adjustment; however, the seedlings from the sand substrate obtained greater accumulation of dry mass of root in the lower holding capacities with decrease of these values with the increase of the holding capacity, the opposite being verified for the seedlings maintained in vermiculite.

The accumulation of dry mass of root in the lowest holding capacities is probably due to the need for root expansion by the plant in the capture of water and nutrients when these are deficient or even absent (Silva & Delatorre, 2009), as in the case of sand.

The accumulation of seedling mass represented by fresh and dry mass of the aerial part and dry mass of root showed maximum of 0.206 g; 0.039 g and 0.008 g, respectively, when kept in the commercial substrate, and with water availability between 50 and 75% of the water holding capacity of the substrate.

CONCLUSION

The sowing of *B. scandens* should be carried out initially in vermiculite with a holding capacity of 100% until the establishment of the seedling emergence and then transplanted to commercial substrate keeping between 50 and 75% holding capacity.

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