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RESEARCH ARTICLE

AGRO INDUSTRIAL WASTE: ALTERNATIVE SOURCES OF SUBSTRATES FOR THE PRODUCTION OF LETTUCE SEEDLINGS

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ABSTRACT

The agro industrial sector produces large amounts of waste which often are not properly disposed. The objective of this study was to evaluate the potential use of burnt rice husks and sugarcane bagasse as a substrate for the production of iceberg lettuce seedlings. The study was conducted in the city of Frederico Westphalen, RS, in 2015. The experimental design was completely randomized and consisted of six treatments and four replications with 100 plants / replication. The treatments were composed of organic waste, a commercial substrate, and mixtures of these components resulting in the following treatments: T_1 = 100% commercial substrate (SC); T_2 = 100% burnt rice husks (CA); T_3 = 100% sugarcane bagasse (BC); T_4 = 25% CA + 75% BC; T_5 = 50% CA+ 50% BC and T_6= 75% CA + 25% BC. Responses were evaluated for germination, emergence speed index, dry weight of the shoot and root system. From the results it can be concluded that the best performance of the seedlings was found in the commercial substrate; however, costs can be reduced with the use of burnt rice husks, even though the chemical characteristics may influence the development of the seedlings. The use of sugarcane bagasse resulted in low levels of seedling development, this was likely related to the physical characteristics of these mixtures and the possible presence of allelopathic substances.

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INTRODUCTION

Currently, lettuce (*Lactuca sativa* L.) is one of the most widespread leafy vegetables, and itis cultivated in almost every country in the world. In Brazil, it is among the most important crops in terms of economics and daily consumption (Medeiros *et al*, 2011). Improving the developmental conditions of plants can increase the success and rigor of plant growth after propagation. The production of plants for propagation can be seen as one of the most important steps in the production cycle of lettuce. Plant substrates are one of the factors which has a direct influence on the initial development of plants. Substrates can deliver not only water, but oxygen and nutrients for plants, this is especially important as the growth of roots can be limited when plants are grown within rigid containers (Fermino, 2002; Medeiros *et al.,* 2011).

For the production of seedlings, it is important to use high quality substrates considering that, during germination and early stages of development, because the plants are especially sensitive to negative factors of the environment such as temperature, solar radiation and pathogen attack (Carmona *et al.,* 2012.; Medeiros *et al.,* 2011). A good substrate should have ideal characteristics for water economy, provide decent aeration, permeability, function as a buffer to pH, have a high nutrient retention capability, resistance to decomposition, and generally promote plant health thus avoiding the spread of pests and diseases (Kämpf, 2005). For the production of plants, the most widely used commercial substrates are those based on vermiculite or peat; however, the high price of these materials has spurred interest in discovering alternative sources of substrates. These substrates would need to present ideal physical and chemical characteristics for providing adequate growth and early development of seedlings. Studies have been published on the potential use of different waste of agricultural origin as substrates for seedlings (Carmona *et al.,* 2012; Freitas *et al.,* 2013; Medeiros *et al.,* 2011; Andreani Junior *et al.,* 2011). In order to reduce production costs of lettuce

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propagation, this study aimed to evaluate the potential use of agro-industrial waste for the production of iceberg lettuce seedlings.

MATERIALS AND METHODS

Experimental Site and Plant Material

The experiment was conducted at the Federal University of Santa Maria, Campus Frederico Westphalen– RS in the period from June to August 2015; the university is located in the state's northwest region, whose geographic coordinates are 27º 23 '48' 'south latitude, 53 25' 45 " west longitude. The climate is Cfa, humid temperate with hot summers, according to Köppen. The experiment was conducted in an arched greenhouse, 10 X 20 m, covered with transparent polyethylene film of low density and a thickness of 150 microns. For the composition of the substrates used in the experiment, residue from sugarcane processing which washeld in a dry composting system for eight months in the city of Frederico Westphalen – RS was obtained. The commercial substrate Carolina® was used in this study. The husks of burnt rice were obtained in a fuel industry in the southern region of the state. After creating various mixtures of the sample substrates, the chemical characteristics of each substrate were analyzed, in the period June to August 2015. Substrates and materials were used to grow the American lettuce variant 'iceberg', which had a germination rate of 85%. Seeding was performed manually by placing three seeds per cell, in a seedling tray. After emergence, thinning was conducted and one seedling was left per cell. Irrigation management consisted of daily manual watering according to crop needs.

Experimental Design and Statistical Analysis

A complete randomized design (DIC) consisting of six treatments and four repetitions was used, each repetition being composed of 100 plants / repetitions. The responses were evaluated after 54 days of germination. The treatments were composed of organic waste and mixtures of commercial substrates, resulting in treatments: T_1 = 100% commercial substrate (SC); T_2 = 100% burnt rice husks (CA); T_3 = 100% sugarcane bagasse (BC); T_4 = 25% CA + 75% BC; T_5 = 50% CA+ 50% BC and T_6= 75% CA + 25% BC.

The following variables were evaluated:

germination percentage (PG) and speed index of germination (IVG).The variable speed of germination index (IVG)determined by counting emerged seedlings daily, according to the formula proposed by Maguire (1962).

$$\mathrm{IVG} = (\Sigma\, \mathrm{G})/(\Sigma\, \mathrm{N})$$

The variable germination percentage (GP) was calculated according Laboriau and Valadares (1976), using the following formula:

$$PG = (N/A) \quad 100$$

Where: PG = Number of germinated seeds per day;
N = Number of days.

After 54 days the cultures were observed, and it was found that the seedlings of treatments T3; T4; T5 and T6 did not reach necessary development for transplantation (four to five leaves).Thus, for the variables of shoot dry mass, and root system mass (MSPA and MSSR, respectively), we only performed statistical analyses for treatments T1 (SC) and T2 (CA). The experimental design for these two variables was the DIC with two treatments and four replications, with each repetition consisting of 100 plants. The experiment was conducted in 200 cell trays, with 100 plants / repetition, totaling 400 plants per treatment. The data were submitted to analysis of variance and the treatment means were compared by using a Tukey test at 5% probability, using the statistical program GENES (Cruz, 2013).

Physical and Chemical Analysis of Substrates

For analysis of the physical substrate, approximately five liters of substrate were sieved in a 20-mm sieve, as according to the European Standard EN 13040 (Cen, 1999). The material retained in the sieve measured less than 10% of the total mass, and thus the following procedure was adopted for substrate density analysis (DS), and analysis of the water retention curve. For the analysis of DS analysis a self-compaction method described by Hoffmann (1970) was used, which consists in supplementing a transparent plastic beaker with 250 ml of moist substrate at 0.50 ± 0.03 g g^{-1}. After this step, the same sample was dropped into a test tube from a height of about 10 cm, for 10 consecutive times. With help of a spatula, the surface was leveled off, values were recorded, and then the wet material was weighed and taken to the drying oven at65ºC, for 48 hours, before being weighed again. For the analysis of the water retention curve, a method for analyzing table tension was used, described by Kiehl (1979), in which a cylinder was filled (5 cm diameter x 5 cm height) with a substrate, at the density given above. Next, the cylinders were placed in plastic containers and every 30 minutes distilled water was added at approximately 0.5 cm to approximately 4.5 cm height. 24 hours after the last addition of water, the cylinder containing the substrate was removed and the saturated substrate was weighed and then submitted to a regimen of different pressure values: -1; -3; -5; -6 and -10 kPa, until the sample presented a constant weight in the range of 12 hours between a weighing and another. Between each applied tension change, the samples were weighed. After the final pressure application, the samples were placed in a drying oven at 105ºC, until a constant weight was achieved. The volume of water retained in the substrate in the tension 0 kPa (saturated substrate) defines the total porosity (PT). The -1 kPa pressure determines the volume of air present in the substrate after free drainage ceases. The difference between the PT of the substrate and the volume of water retained -1 kPa corresponds to the aeration space (AS) of the substrate (Corá and Fernandes, 2008).The readily available water (AFDWas determined by the difference between the amount of water found between points -1 and -5 kPa. The buffering water (AT) was defined as the volumetric water released between tensions of -5 and -10 kPa. Available water (AD) was determined by the difference between the volume of water -1 and -10 kPa. The remaining water (AR) was determined by the amount of water remaining in the sample after being subjected to the suction pressure -10kPa.

RESULTS AND DISCUSSION

Regarding the chemical composition of substrates, treatments showed differences, mainly pH (Table 1), and this is one of the factors which likely influenced the growth and development of produced seedlings. It can be observed that the commercial substrate had the greatest accumulation of analyzed nutrients (Table 2). Physical and chemical properties are essential for the stability of a substrate, providing greater amount of nitrogen available to plants, as well as moisture retention capacity (Martinez, 2002). An ideal substrate must be able to supply water, oxygen and nutrients for good plant growth (Fermino, 2002). The most common encountered problems regarding substrates are with regards to low and high water retention capacity, because they must be able to retain and also drain water so that the roots of the seedlings are aerated. This will depend on the size and shape of the particles, as well as the container height (Carmona *et al,* 2012). It can be observed that the physical characteristics in different mixtures of substrate did not vary greatly (Table 2). The air space was lower in all treatments containing pure BC or as a mixture, resulting in lower drainage of the substrate for plants. The substrates with CA and SC likely allowed for better drainage of water due to its lower absorption capacity, and value of water retention thus creating space for help and improving root aeration and the activity of microorganisms (Pagliarini *et al.,* 2012).

The use of sugarcane bagasse can result in issues with the retention of available water for a culture, and in short periods of time this could result a small fraction of extracted water, making it necessary to more efficiently control irrigation when using this substrate. Although the rice husks showed the lowest total porosity values, it was seen that this substrate presented the highest and lowest values for AFD and AR; this indicates a low degree of water adsorption. These results are important because they reduce the need for frequent irrigations and also the risks of deficient aeration. When a substrate has low aeration, it may become more limited by this than low water retention, a problem which can be solved by more frequent watering (Carmona *et al.* 2012). For the evaluated substrates, the lowest value was found for CA (0.25 m^{-3} m^{-3}). It was observed that the density of the substrates was higher in treatment CA. This is due to the increase in the micro porosity of the substrate, which provides more easily extractable water through a greater retention capacity (Trigueiro and Guerrini, 2014), this can be observed in the substrates' water retention curve (Figure 1). It also presents higher porosity, a lower value of unavailable water, and greater values for water retention at -1 and -3 kPa; these are desirable factors, as they relate to total water availability. In relation to sugar cane bagasse, it appears that the majority of water present in the substrate results in direct drainage; the water is essentially wasted. The low degree of water availability for this substrate is very small, requiring more frequent irrigation.

Table 1. Chemical composition of the substrates used for the production of iceberg lettuce seedlings, Frederico Westphalen, UFSM, 2015

substrate	Nutrients % (mm^{-1})						
	Nitrogen (N)	Phosphorus (P)	Potassium (K)	Calcium (Ca)	Magnesium (Mg)	Carbon org. C	pH
SC*	0.58	0.06	0.33	1.53	2.2	22.98	5.60
CA	0.12	0.12	0.44	0.12	0.02	2.61	9.30
BC	0.24	0.04	0.15	0.08	0.02	28.72	5.6
50% CA + 50% BC	0.26	0.08	0.27	0.09	0.02	6.22	7.00
75% CA+ 25% BC	0.23	0.07	0.23	0.09	0.02	7.66	7.60
25% CA + 75% BC	0.24	0.04	0.22	0.08	0.04	11.5	7.30

*SC = commercial substrate; CA = burnt rice husks; BC = sugarcane bagasse

Table 2. Physical composition of the substrates used for the production of iceberg lettuce seedlings. DS (density), PT (total porosity), EA (aeration space), AFD (water readily available), AT (total water), AD (available water) and AR (remaining water). Frederico Westphalen, UFSM, 2015

Substrates	DS mg m^{-3}	PT $m^{-3}m^{-3}$	EA $m^{-3}m^{-3}$	AFD $m^{-3}m^{-3}$	AT $m^{-3}m^{-3}$	AD $m^{-3}m^{-3}$	AR $m^{-3}m^{-3}$
SC	0.10	0.69	0.32	0.14	0.03	0.16	0.22
CA	0.13	0.60	0.25	0.22	0.04	0.26	0.09
BC	0.08	0.68	0.45	0.02	0.00	0.02	0.21
50% CA + 50% BC	0.11	0.72	0.43	0.13	0.03	0.15	0.13
75% CA+ 25% BC	0.11	0.72	0.43	0.13	0.03	0.15	0.13
25% CA + 75% BC	0.11	0.73	0.43	0.11	0.02	0.14	0.16

SC = commercial substrate; CA = burnt rice husks; BC = sugarcane bagasse.

Table 3. Shoot dry mass (MSPA) androot system dry mass(MSSR)of iceberg lettuce seedlings. Frederico Westphalen, UFSM, 2015.

Treatment	MSPA (g)	MSSR (g)
Commerical Substrate	3.550 a	1.950 a
Burnt Rice Husks	2.520 b	1.070 b
CV (%)	13.80	15.60

*Means followed by the same letter do not differ by test Tukey at 5 % of significance.

For the variable of germination percentage (PG), the SC treatment was only significantly different from the burnt rice husk treatment (CA) (Figure 2) where SC was seen to promote a greater degree of seed germination. This performance can be explained by the higher nutritional intake of other treatments, when compared to treatments with the addition only of CA. For the variable, germination speed index (IVG), the analysis of variance was not significant (p> 0.05) (data not shown).

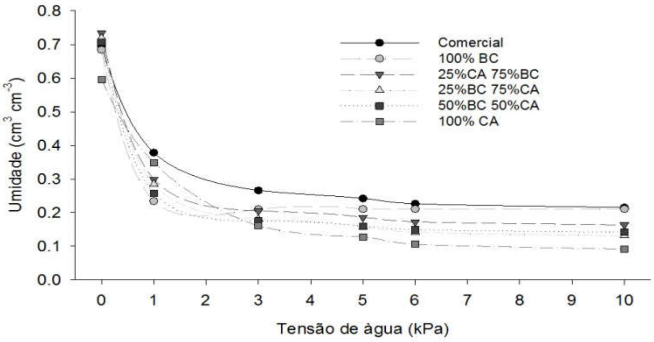

Figure 1. Retention curve of substrates used in this experiment. CA = burnt rice husks; BC = sugarcane bagasse.Frederico Westphalen, UFSM, 2015

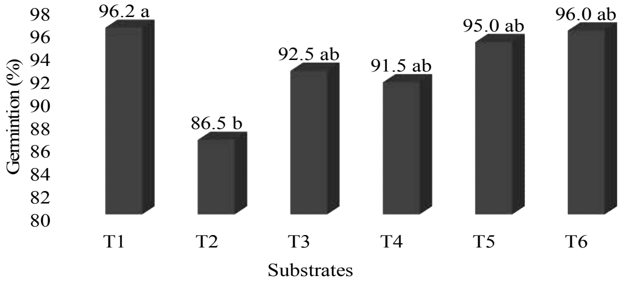

Figure 2. Percent of lettuce seed germinationin different substrates T_1 = 100% commercial substrate (SC); T_2 = 100% burnt rice husks (CA); T_3 = 100% sugarcane bagasse (BC); T_4 = 25% CA + 75% BC; T_5 = 50% CA+ 50% BC and T_6= 75% CA + 25% BC. Means followed by the same letter do not differ by the Tukey testat5 % significance.Frederico Westphalen, UFSM, 2015

Minor rates of germination were found in the burnt rice husk treatment, these results are consistent with Martins *et al*. (2001), whom, working with lettuce seedlings in commercial substrates and humus with the addition of vermiculite, found the best results in commercial substrate Plantmax®. Já Pereira *et al*. (2011), evaluating production of vegetable seedlings in different substrates, observed that lettuce and broccoli showed no statistically significant differences for the variables analyzed between a commercial substrate (Plantmax®) and an alternative (humus and burnt risk husks), indicating the adaptation of these species to different substrates. Seedlings of lettuce produced with organic and biofertilizer substrates showed higher values for almost all variables (Medeiros *et al.,* 2007). There was a significant difference by the test F ($p \leq 0.05$) for the two variables, MSPA and MSSR. For these variables, the commercial substrate was significantly higher when compared to the burn rice husk treatments (CA) (table3). The commercial substrate and burnt rice husk treatments resulted in the greatest shoot accumulations when compared to treatments carried out with sugarcane bagasse. It was seen that increased proportions of sugarcane bagasse significantly reduced the growth of seedlings. The burnt rice husks showed a higher dry mass of lettuce seedlings after 54 days, even though the substrate had a high pH and relatively low nutrient composition. Compared to other substrates, sugarcane bagasse showed, in no capacity, a valuable use for lettuce seedlings. Disregarding the chemical characteristics of this plant, which were not greatly different from the other treatments, the physical properties were observed to be unideal for the lettuce seedlings given that the high porosity resulted in a lower water capacity for water retention. The water supply was equal for all treatments; those which were in part composed of BC, may have had a greater water deficit, resulting in the observed growth differences. This deficit can be addressed by increasing the frequency of irrigation in these treatments. Freitas *et al.* (2013) testing different substrate compositions also noted that increasing the ratio of burnt rice husks in substrates caused a significant reduction in the growth of leaf area, this factor may be related to higher intake of nutrients in the alternative substrates. The burnt rice husks are different from the burned bark because the latter has less aeration space, providing greater retention of water for the plants. Besides the chemical and physical effects of sugarcane bagasse in the production of lettuce seedlings, it has been hypothesized that this substrate has allelopathic compounds capable of inhibiting the growth of plants or indirectly affect the subsequent germination and development (Oliveira and Simões, 2014). For example, sugarcane stalks have'direct action' organic compounds which have known allelopathic effects, among these we would like to highlight both hydroxamic acid 2,4-diidroxi–1,4-benzoxazin-3-ona (diboa), and its degradation product, 2-benzoxazolinona (BOA) (Sampietro *et al.,* 2007; Singh *et al.,* 2003). Other compounds are also reported in the literature, such as trans-ferulic acid, cis-ferulic acid, vanillic acidand syringic acid which are considered to be growth inhibitors of weeds and lettuce (Sampietro *et al*, 2006). In addition, lettuce is highly sensitive to allelopathic compounds and is considered an indicator plant allelopathy (Alves *et al.,* 2004). The results of this study demonstrated that the chemical and physical characteristics of the evaluated substrates directly influence the production of lettuce seedlings. Moreover, the substrates used may have allelopathic effects capable of inhibiting plant growth; further studies are required in order to elucidate these characteristics.

Conclusions

The commercial substrate demonstrated a greater efficiency for the production of iceberg lettuce seedlings than the alternative substrates; it has balanced physical and chemical properties. Burnt rice husks can also be used for production of lettuce seedlings, but it is necessary to increase the frequency of irrigation for this substrate. Mixtures with sugarcane bagasse showed low plant development likely due to low water holding capacity and overall values of available water, and possibly related to the release of allelopathic compounds.

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