UNIVERSIDADE FEDERAL RURAL DO RIO DE JANEIRO
INSTITUTO DE FLORESTAS
CURSO DE GRADUAÇÃO EM ENGENHARIA FLORESTAL

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BIOSOLID IN THE COMPOSITION OF SUBSTRATE FOR THREE URBAN AFFORESTATION SPECIES SEEEDLINGS PRODUCTION

Prof. Dr. PAULO SÉRGIO DOS SANTOS LELES
Orientador

SEROPÉDICA, RJ
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Monografia apresentada ao Curso de Engenharia Florestal, como requisito parcial para a obtenção do Título de Engenheiro Florestal, Instituto de Florestas da Universidade Federal Rural do Rio de Janeiro.

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Monografia aprovada em 06 de junho de 2018

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DEDICATÓRIA

Se você buscar a verdade, encontrará o conforto no final;
se buscar o conforto, não terá nem o conforto nem a verdade.
C.S. Lewis

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ABSTRACT

The use of urban sewage sludge in agriculture as a source of organic matter has been shown as a sustainable alternative for final disposal of this residue. After treatment and stabilization sewage sludge is known as biosolid and can offer organic matter and nutrients for plants growth. This study aimed to evaluate the feasibility of biosolid used as a substrate component for *Schinus molle*, *Licania tomentosa* and *Peltophorum dubium* seedlings production. The design was completely randomized with four treatments and six replications: three different proportions of biosolid in the mixture (25, 50 e 100%) compared to substrate containing 50% of cattle manure as control, all treatments were associated with sand and subsoil. Each seedling represented a replication. Firstly, seedlings were produced in 280 cm³ tubes and 120 days after sowing they were transplanted to 18 L vases. Plants shoot height and collar diameter were determined to evaluate seedlings growth at the time of 0, 6, 12 and 18 months after transplantation. Biosolid presented a high potential for the production of seedlings for urban afforestation promoting the best growth results in both evaluated morphological aspects. The results demonstrate that biosolid may be considered suitable for the production of these species and its usage as seedlings substrate is a feasible alternative for final disposal.

Keywords: Sewage sludge and cattle manure.

RESUMO

O uso de resíduos sólidos urbanos na agricultura como fonte de matéria orgânica tem se apresentado como uma alternativa sustentável para a sua disposição final. O lodo de esgoto depois de estabilizado é conhecido como biossólido e constitui em fonte de matéria orgânica e de nutrientes para as plantas. Objetivou-se neste trabalho verificar o potencial do biossólido como componente de substrato para produção de mudas de *Schinus molle*, *Licania tomentosa*. e *Peltophorum dubium*. Foi utilizado o delineamento inteiramente casualizado, constituído por quatro tratamentos e seis repetições: três diferentes composições contendo biossólido em proporções crescentes (25, 50 e 100%) e um tratamento sem uso de biossólido, contendo 50% de esterco bovino, todos associados com terra de subsolo e areia. Cada repetição foi composta por uma muda. Inicialmente, as mudas foram produzidas em tubetes plásticos de 280 cm^3 e 120 dias após a semeadura foram transplantadas para vasos de 18 litros. Mensurou-se a altura da parte aérea e o diâmetro do coleto na época do transplantio e, posteriormente, aos 6, 12 e 18 meses após o transplantio. O biossólido apresentou potencial elevado para a produção de mudas de arborização urbana das três espécies utilizadas, proporcionando crescimento estatisticamente igual ou superior ao observado com esterco bovino. para os parâmetros analisados. Os resultados demostraram que o biossólido pode ser usado para produção de mudas das três espécies florestais e o seu uso como substrato é uma alternativa viável de destinação final.

Palavras-chave: Lodo de esgoto e esterco bovino.

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1. INTRODUCTION

The world's urban population has grown rapidly on the last decades, having increased from 30% in 1950 to currently 55% and has a projection of 68% by 2050, according to the last World Urbanization Prospects published by the United Nations (UN) in 2018. Most urbanized regions are Northern America followed by Latin America and the Caribbean where 81% of its population lives in urban areas (UN, 2018). In Brazil, the urban population increased from 81,2% in 2000 to 84,4% in 2010 demonstrating a record urbanization rate for the country so far (IBGE, 2018).

One of the main environmental challenges for Brazilian cities lies on the final disposal of sewage sludge generated in Sewage Treatment Stations (STS). Despite presenting an enormous potential for agricultural use in Brazil, the main final disposal for sewage sludge are still the landfills (ABREU et al., 2017b). Therefore, it is important to find sustainable alternatives for disposal in order to reduce costs and ensure environmental and population protection.

After treatment and stabilization sewage sludge is known as biosolid and constitutes an important source of organic matter and nutrients for plants. Sewage sludge composition is influenced by its origin. Therefore, prior to application in agriculture the material must be verified and assessed in order to stablish if heavy metals and pathogenic agent rates comply with legislation. In Brazil, the 375/2006 CONAMA Resolution stipulates procedures and standard quality limits for the agricultural use of sewage sludge in order to prevent human and environmental contamination.

Urbanization changes several aspects of a region including social, economic and environmental setups. Environmental negative impacts including adverse changes in the microclimate and in the landscape can directly affect population's life quality and health. In this scenario, urban afforestation is a feasible approach to mitigate such impacts (FARIA et al., 2007). The presence of trees can improve urban environments acting both on the dwellers physical and mental perceptions. According to Martini and Biondi (2015) urban afforestation provides several benefits including microclimate control, acoustic mitigation, air pollutants filter, soil protection, urban landscape improvement, as well as, many other ecological services.

For urban afforestation projects to succeed trees are expected to withstand stressful environments and conditions. Seedlings production is an essential part of the process since it ought to provide resistant seedlings thus reducing future management operations. Seedling's quality is strictly related, among other aspects, to container (ABREU et al., 2015) and substrate (CARNEIRO, 1995) choices. CEMIG (2011) stablishes standards for urban afforestation seedlings including the minimal height of 2,5m and the minimum collar diameter of 5cm. Seedlings are usually produced in large containers of 15L, or more, in order to sustain long production periods in forestry nurseries and to provide space and nutrients for a balanced development between root system and aerial parts. Therefore, due to containers volume a great amount of substrate is necessary for urban afforestation seedlings production.

According to Carneiro (1995), substrate's quality is important since its chemical, biological and physical properties will impact seedlings development. Substrates solid phase is composted by mineral and organic particles. Organic matter provides nutrients to the system, retain water, nutrients and improve substrate physical conditions (TRIGUEIRO and GUERRINI, 2003).

The main organic matter source in substrates composition for seedlings production in the state of Rio de Janeiro, Brazil is cattle manure (SEA, 2013). Nonetheless, this material is considered a major limiting aspect for seedlings production in the metropolitan region of Rio de Janeiro where the majority of forestry nurseries are located (ALONSO, 2013). The state does not have a significant livestock production and it is mainly performed in extensive production systems hence cattle manure is not available in large quantities neither in good quality conditions.

Biosolid used as substrate component in forestry seedlings production has provided positive results for native and exotic species (TRIGUEIRO and GUERRINI, 2003; SCHEER et al., 2012; CABREIRA et al., 2017b). In addition, the final disposal for biosolid holds environmental and economic advantages in comparison to orthodox and current means since it promotes principles of residues reuse or recycling. However, most studies in the forestry subject has focused on seedlings production towards environmental restoration and still few information (RIBEIRO, 2017) is available for urban afforestation seedlings.

It is important to ensure that substrate components are accessible and of low cost in order to attend rural producers that in general terms have low income. Caldeira et al. (2014) states that researches aiming to inventory available materials in distinct regions are extremely necessary. These researches allow the identification of local and inexpensive inputs for substrates composition consequently reducing production costs and improving nurseries economical independency.

Therefore, biosolid stands as a potential alternative to cattle manure in Rio de Janeiro. Besides providing nutrients and organic matter, biosolid is generated in urban areas where the demand for substrates components in urban afforestation also lies. Therefore, it would possibly improve production, reduce costs and provide a sustainable final disposal alternative.

2. OBJECTIVES

This study aimed to evaluate the feasibility of biosolid as a substrate component for *Schinus molle* L, *Licania tomentosa* (Benth) Fritsch. and *Peltophorum dubium* (Springer.) urban afforestation seedlings production.

Moreover, this study aimed to verify the biosolid proportion that promoted higher seedlings growth and compare seedlings produced with biosolid and cattle manure in the same proportion.

3. LITERATURE REVIEW

3.1. Urban Afforestation

According to Magalhães (2006), the concept of urban afforestation can be confused with urban forests and, especially in Brazil, the consolidation of such term is still on a dynamic process of debates. Despite sharing some equal benefits, urban afforestation does not stand for the same meaning as urban forests. Nowak (2016) defines urban forests as the composition of all trees within urban lands, while Konijnendijk and Randrup (2004) states the concept of urban forestry as the art, science and technology of managing trees and forest resources for psychological, sociological, aesthetic, economic and environmental benefits.

Therefore, for the purpose of this present work, urban afforestation is defined as isolated trees or small groups present in urban centers. Individual trees are well established in the entire urban area including parking lots, streets and private properties. Urban afforestation trees are planned, cultivated, maintained and interact with the environment as individuals and not as a forest fragment (MAGALHÃES, 2016).

Early records of trees and green areas as contributors to more attractive cities date back to ancient civilizations for example the Egyptian, Greek and Roman Empires. During the Middle Age, cities would preserve and maintain green areas inside its walls, especially for resources, agriculture and wood. Interest in the aesthetic and recreational benefits of urban green spaces emerged during the Renaissance period and stimulated the creation of parks and green areas for leisure and social prestige. The awareness for urban afforestation continued to strengthen in Europe as space planning and management became more established parts of municipal activity during modern times (KONIJNENDIJK and RANDRUP, 2004).

In Brazil the first urban afforestation experience took place in Recife with seedlings planting on the streets. Afterwards in Rio de Janeiro, Belém, Olinda, Vila Rica and São Paulo mainly expressed in Public Parks and Botanic Gardens as an European influence. In Rio de Janeiro urban afforestation started with the creation of the Passeio Público in 1783 and later in 1808 the creation of the Rio de Janeiro Botanic Garden. An important mark for the city was the arrival of Auguste Marie Glaziou in 1860, a French architect responsible for projects such as Quinta da Boa Vista and Campo de Santana. Glaziou established the use of native tree species and laid the ground for urban afforestation planning and regulation that has been developed until present days (MILANO and DALCIN, 2000).

Unlike natural environments, urban centers exhibit several artificial features for instance soil impermeabilization, residues generation, atmospheric pollution and loss of green cover. Consequently, urban afforestation plays a crucial role concerning cities basic planning and administration in order to mitigate urban problems and enhance environmental services (MILANO and DALCIN, 2000). According to Zhang and Zheng (2011) urban trees are able to enhance the liveability of cities as they act as a municipal infrastructure and provide valuable services. Grise et al. (2016) affirms that all vegetation, native or exotic, is a central component of urban landscapes acting over climate, ecology and sociological features.

From the several benefits provided by urban afforestation it is possible to highlight pavement and urban soil protection (MCPHERSON and MUCHNICK, 2005), erosion control (DA SILVA et al., 2011), social benefits (VECCHIATO and TEMPESTA, 2013), energy savings (ZHANG and ZHENG, 2011), microclimate regulation (ROCHA et al., 2004), noises absorption, health benefits, habitats for wildlife (ZHENG et al., 2011), absorption of heavy metal pollutants (MU et al.,2004) and recreation opportunities. Furthermore, according to Dantas and Souza (2004) urban afforestation reduces the feeling of oppression, improves cities aesthetics, provides thermal comfort, reduces erosion and water runoff as well as many other ecological services.

Moreover, the perceptions and opinions of cities residents concerning urban afforestation have been well documented on many countries. Schroeder et al. (2006) surveyed communities both on the U.S. and UK and discovered that residents in all the communities held similarly high levels of satisfaction with the trees outside their homes. Other researches held in U.S. corroborate the high level of acceptance of street trees and demonstrate a positive correlation between education level and urban afforestation support (GORMAN, 2004; ZHANG and ZHENG, 2011).

Furthermore, studies in Brazil corroborate with previous results showing that most people relate positively to urban trees and that the aesthetical and climate regulation are some of the most perceived effects (MARTINI et al., 2014; BODROWSKI and BIONDI, 2016). However, urban afforestation in Brazilian cities many times are marked by lack of management and maintenance causing problems such as limited growing spaces, vandalism, inadequate pruning, transmission lines conflicts and equivocated species selection (FARIA et al., 2007; VOLPE-FILIK, 2007; FARIA et al., 2013).

3.2 Urban Afforestation Seedlings Production

For an accurate and efficient urban afforestation planning several aspects are ought to be taken into consideration, such as: urban environment, physical available space and species characteristics. Selected tree species must tolerate adverse soil conditions, air pollution, limited space, extreme climate variations, rain regime, luminosity alterations and vandalism (MILANO and DALCIN, 2000). Urban afforestation planners frequently choose native tree species since they are more adapted to local conditions and use available resources in most efficient way thus ensuring superior success rate for afforestation projects (FARIA et al., 2007; OLDFIELD et al., 2013).

Urban afforestation success also relies on trees performing the intended function planned even under stressful environments and conditions. Therefore, the selection of adequate tree species and genotypes as well as good quality seedlings are important factors in promoting environmental benefits and decreasing costs in the establishment and management of urban green areas (SAEBO et al., 2003). Using unsuitable seedlings in sidewalks may incur on future problems to both urban equipment and other trees. Most common problems found in this situation are low survival rate and bad canopy formation due to lack of silvicultural traits (BIONDI et al., 2007). Urban afforestation seedlings are expensive mainly due to long production periods on forestry nurseries and the large amount of demanded input.

Forest seedlings quality is strongly related to container choice, substrate and silvicultural traits during production (CARNEIRO, 1995). Substrates are the appropriate growing media characterized by adequate water retention, oxygen and nutrients provision, compactible pH, absence of chemical elements in toxic rates and proper electrical conductivity (TRIGUEIRO and GUERRINI, 2003). Several studies have focused on different substrates composition and its influence on seedlings development, sanity and strength (CUNHA et al., 2005).

According to CEMIG (2011), urban afforestation seedlings should meet quality standards such as minimal height for first crotch of 2,5 m, minimal collar diameter of 5,0 cm, adequate container volume, absence of diseases and pests and well distributed canopy structure. Shorter seedlings are more susceptible to vandalism. In São Paulo state 50 to 80% of planted urban afforestation seedlings are destroyed due to vandalism (CPFL ENERGIA, 2008).

Urban afforestation seedlings are sowed in smaller containers and only months later transplanted into larger containers of, in general, 15 to 50 L. Containers of greater volume are necessary in order to provide nutrients, organic matter and space for root growth considering that the production can take from 1 to 2 years in the nursery. Important silvicultural traits, for instance staking-out and pruning, must be undertook during this production stage to guarantee seedlings development in shorter time and stem structure that will not cause problems for streets establishment (BIONDI et al., 2007).

3.3 Biosolid

Sewage treatment stations, or plants, are a set of facilities and equipment designed to treat both industrial and domestic sewage. This service is crucial for the municipality and environmental conservancy since it avoids the discharge of a large amount of organic load and pollutants directly to water bodies. The final product generated during this process is named sewage sludge and when this material is stabilized by compost process, it is called composted sewage sludge or biosolid (SILVA et al., 2004).

The choice for the biosolid nomenclature has been used in substitution for sewage sludge in order to emphasize the potential benefits for use in agriculture and forestry after treatment and stabilization (ASSENHEIMER, 2009). After treatment, the composted sewage sludge final disposal represents a major environmental problem for municipalities around the world (GUERRERO et al., 2002), for example, in Connecticut where storage sites are reaching their maximum capacity (BUGBEE, 2002). Brazil faces similar challenge as most sewage treatment stations dispose biosolid mainly in landfills posing both an economical and environmental issue (ABREU et al., 2017b).

Due to high organic matter content and nutrients levels, biosolids hold a high agronomic potential usage as soil amendments and nutrients source (LOUZADA et al., 2015; GUERRINI et al., 2017). Composted sewage sludge has high contents of nutrients and organic matter thus enabling this material for reuse aiming to improve plants productivity and decrease chemical fertilizers dependency (LOBO et al., 2015). Despite sharing common features, chemical composition of biosolids is strictly related to sewage origin and treatment process (BACKES et al., 2009).

Organic matter, macro and micronutrientes present in biosolids play an important role in maintaining soil fertility and physical attributes. For instance, organic matter may improve soil water storage and infiltration thus decreasing erosion (BETTIOL and CAMARGO, 2006). Peñarete et al. (2013) working on agronomical sites in Colombia and Sampaio et al. (2012) on degraded areas restoration projects, observed both a tendency of bulk density decrease, soil aggregates induction, total porosity and structural stability increase in soils where biosolid was applied. Biosolids composts are also a feasible alternative for nurseries productions impacting on appropriate drainage, moisture retention and good fertility to container substrates at lower costs in contrast to commercial components (BUGBEE, 2002).

According to Backes et al. (2009), in general cases, biosolid is rich in N which is present in both organic and non-organic forms thus this material has a strong potential for usage as nitrogen fertilizer. Carbon and nitrogen are predominantly found in biosolid as organic composts while phosphorus is mainly in inorganic form (CARVALHO et al., 2015). Moreover, due to an average relation between N and P of, approximately, 1:4 in composted sewage sludge, biosolids are a suitable source of phosphorous for Brazilian soils (CHIBA et al., 2009). Application of biosolid for agricultural purposes has demonstrated an improvement in production and, in some cases, promoted similar or superior results when compared to mineral fertilization (LOBO et al., 2013).

Silva et al. (2001) observed that sewage sludge positively affected soil fertility through acidity reduction, nutrients supply (Ca, P, S and Zn) and effective cation exchange capacity (CEC) increase. Further benefits from biosolid usage are related to micronutrients since this material can provide elements such as zinc, copper, iron, manganese and molybdenum. Composted sewage sludge is poor in potassium and may demand a combined use with mineral fertilization in order to attend plants demands (BETTIOL and CAMARGO, 2008).

Despite biosolid valuable potential for agronomic application it is essential to assess whether the material generated in sewage treatment stations comply with legislation and regional needs (ABREU et al., 2017b). Since physical and chemical properties of sewage sludge changes in time and according to the source (HERNANDEZ-APAOLAZA et al., 2005), biosolid should always be tested before agronomical use. Main restrictions for use are possible presence of contaminants (heavy metals, pathogenic agents, trace elements, organic chemicals, glass), potential phytotoxicity (immaturity and/or salt level, pH) and differences in species responses (OSTOS et al., 2008).

The 375/06 CONAMA resolution is the current legislation that establish standards and procedures for sewage sludge use for agronomical purposes. The resolution affirms that the agronomic use of such residue guarantees environmental benefits and represents an adequate reuse alternative. Moreover, according to the legislation, sewage sludge characterization must comprehend: agronomical potential; organic and inorganic potential substances; bacteriological indicators; pathogenic agents and stability (CONAMA, 2006).

4. MATERIAL AND METHODS

The study was carried out in the Botanic Garden at the Universidade Federal Rural do Rio de Janeiro, located in the city of Seropédica, Rio de Janeiro state. The region climate is AW, according to Koppën's classification, characterized by high temperatures throughout the year, distinct wet and dry seasons and most of precipitation occurring on summer.

The experiment was conducted from seedlings transplantation into plastic pots in July 2016 until January 2018 when the last measurement took place, completing a total of 18 months study.

Biosolid used in this study was provided by the Water and Sewage State Company of Rio de Janeiro (CEDAE) and came from the Ilha do Governador plant, located in the North region of the city, which receives only sewage of domestic and commercial origin. This sewage treatment station plant performs a secondary treatment with activated sludge system, biological anaerobic stabilization, centrifugal dewatering and drying in semipermeable open-air beds.

For chemical and biological characterization purposes and also agronomical potential evaluation a representative sample of biosolid was collected. The analysis was conducted in accordance with standards from Attachment IV of the 375/06 CONAMA (2006) resolution, made available by CEDAE. Biosolid samples were sent for chemical total content analysis in laboratory, results are shown in Table 1. The elected method for this analysis was the EPA 3050 with acid digestion (HNO_3 + H_2O_2) in digestor blocks at 95 °C. Results for inorganic substances were in accordance with resolution standards.

Table 1: Chemical analysis result for total content for biosolid and cattle manure (CM) used in the production of urban afforestation species before nursery pot's filling

Subst.	P	K	Ca	Mg	Al	N	C	OM	OC	C/N
	------------ g kg^{-1} ------------							------- % --------		
Biosolid	5,7	2,1	14,7	3,6	35,4	12,5	110,5	15,39	8,93	8,84
CM	3,2	4,5	14,6	4,2	7,0	14,3	185,8	27,08	15,71	12,99

K - Flame Photometry; P, Ca, Mg and Al - ICP-OES; C and N - Dry Combustion method; OM - Calculate by Total Organic Carbon Content using Van Blender factor (1,724), OM= 1,724 X C.

4.1 Seedlings Production

For the production of urban afforestation seedlings three tree species were chosen: *Schinus molle* L, *Licania tomentosa* (Benth). Fritsch. e *Peltophorum dubium* (Springer.) Taub. Species selection was due to the ornamental attributes and vast use in streets and car parks urban afforestation in the city of Rio de Janeiro. Moreover, all three species are listed in several Urban Forest Master Plans and Urban Afforestation Manuals across the country (CEMIG, 2001; BARBED0 et al., 2005; CPFL ENERGIA, 2008; FPJ, 2015). Each species represented an individual experiment.

Firstly, seedlings were produced in 280 cm³ tubes using a substrate composed of 80% biosolid and 20% vermiculite. Seedlings production proceedings and management followed usual standards. For each species 24 seedlings were produced on a total of 72. Approximately 120 days after sowing they were transplanted to 18L plastic nursery pots. Biosolid was used as a substrate component in three distinct treatments in order to assess an alternative organic matter source in contrast to cattle manure.

Seedlings were selected for uniform shoot height and diameter before transplantation into 18L plastic nursery pots. Substrates were homogenized for each treatment and subsequently nursery plastic pots where filled and identified according to the corresponding treatment and replication. After transplantation pots were placed at the Botanic Garden respecting the experimental design and an average spacing of approximately 1,5 m between replications.

Subsoil used in substrates composition (treatments) was extracted from a hill located in the city of Queimados, Rio de Janeiro, and is classified as a Yellow Ferralsol by Fonseca (2010). Chemical analysis for this soil is presented in Table 2. Due to elevated aluminum content liming was undertook in order to increase pH using 780 g of calcitic limestone (Total Neutralizing Power = 85%) for 600 kg of soil. This calculation was based on recommendation described by Carneiro (1995) for soil pH adjustment in forest seedlings production. A second chemical analysis was undertaken 30 days after liming, results are also presented in Table 2.

Table 2: Chemical analysis result for subsoil used in the production of urban afforestation species in two distinct times, before nursery pot's filling

Characteristic	Unit	Before Liming	After Liming
pH	-	4,3	5,0
P	mg L^{-1}	11	19
K^+	mg L^{-1}	8	10
Ca^{2+}	$cmol_c$ dm^{-3}	0,8	1,7
Mg^{2+}	$cmol_c$ dm^{-3}	0,6	0,8
Al^{3+}	$cmol_c$ dm^{-3}	2,6	0,3
$H+Al^{3+}$	$cmol_c$ dm^{-3}	6,4	3,7
T	$cmol_c$ dm^{-3}	7,8	6,3
V	%	18	41
M	%	64,4	8,9
Corg	(g kg-1)	1,1	1,1

pH in water; P and K: Mehlich-1 Extraction; Ca, Mg and Al: KCl extraction 1,0 mol L; H+Al: calcium acetate extraction Calcium acetate; T= cation Exchange capacity in pH = 7,0; V= base saturation index; m= aluminium saturation index.

The cattle manure used in this experiment was provided by the Agricultural Research Company of the State of Rio de Janeiro (Pesagro-Rio) located in the city of Seropédica. Before using as substrate, the material was aged and sifted. Results obtained from total content chemical analysis for cattle manure are shown in Table 1.

The experiment was composed by four treatments on which T1 represented a control treatment. This treatment represents a commonly used substrate in forest nurseries for urban afforestation tree species seedlings production. Experimental treatments were: T1 - cattle manure (50%), subsoil (40%) and sand (10%); T2– biosolid (25%), subsoil (65%) and sand (10%); T3 – biosolid (50%), subsoil (40%) and sand (10%); T4 – biosolid (100%).

The experiment was conducted using a completely randomized design for each species with four treatments (cattle manure and increasing biosolid doses) and six replications. Every individual seedling represented a replication.

Immediately after experiment assemblage seedlings shoot height and collar diameter were measured using a graduated ruler (cm) and a digital caliper (mm), respectively. Measurement followed at 6, 12 and 18 months after transplantation.

Seedlings were irrigated whenever necessary, approximately three times a week, in the beginning of the morning or late afternoon. During the entire experiment plastic pots were kept clean by hand weeding, where undesired plants were removed from the substrate.

Collected data were processed and analyzed for each species and treatment. The data residuals normality and homogeneity were tested for each variable evaluated using the Lilliefors and Bartlett tests, respectively, with 5% probability. Results were submitted to analysis of variance and when significance was detected, the Tukey test was used ($P \geq 0,95$). For all analysis, the software used was the 'Sistema de Análise Estatística e Genética' (SAEG).

4.2 Soil Analysis

Throughout the experiment substrate chemical fertility and physical properties were also assessed. Substrate samples for chemical fertility analysis were obtained at 0, 4 and 8 months after transplantation. Four samples were collected for each treatment (16 total), without distinction between species using a screw auger. Samples were air dried and sent for analysis in an agronomic laboratory.

In order to evaluate physical characteristics samples were obtained at one year after transplantation and porosity, particle and bulk density were determined. Firstly, for bulk density, using a Kopecky ring of known volume (50 cm^3). In total five samples were obtained per treatment (20 total) without distinction between species. Samples were dried in oven at 105 °C for 24 hours and weighted after cooling. Collected data allowed the following equation to be applied according to the Manual for Methods of Soil Analysis (CLAESSEN, 1997).

$$Bulk\ Density\ (g\ cm^{-3}) = \frac{weight\ of\ sample\ dried\ at\ 105°C\ (g)}{ring\ volume\ (cm^{-3})}$$

Particle density was assessed through the volumetric flask protocol described in Claessen (1997). Samples used in this method were the sieved, weight and dried samples obtained with the Kopecky ring for previous bulk density determination. Mathematical equation applied for particle density is shown below.

$$Particle\ Density\ (g\ cm^{-3}) = \frac{weight\ of\ sample\ dried\ at\ 105°C\ (g)}{50 - alcohol\ volume\ spent}$$

Furthermore, substrate porosity was assessed for each treatment based on bulk and particle density results. Mathematical equation was used according to Claessen (1997).

$$Total\ Porosity = 100 * \frac{(particle\ density - bulk\ density)}{particle\ density}$$

Finally, an empirical test was undertaken aiming to evaluate substrate aggregation for each treatment. For this purpose, one seedling per treatment (total 4) of *Schinus molle* was removed from 18L and aggregation was assessed by visual and mechanical examination. We submitted seedlings clod to three light impacts and designated grades ranging from 5 (excellent aggregation) to 1 (poor aggregation). Seedlings were posteriorly placed in 25L forest nursery pots and identified by treatment and replication. Aggregation was classified by a scale of 1 until 5, from worst to best substrate aggregation.

Substrate physical results were processed and analyzed for each treatment. The data residuals normality and homogeneity were tested for each variable evaluated using the Lilliefors and Bartlett tests, respectively, with 5% probability. Results were submitted to analysis of variance and when significance was detected, the Tukey test was used ($P \geq 0,95$). For all analysis, the software used was the 'Sistema de Análise Estatística e Genética' (SAEG).

5. RESULTS

5.1 Seedlings Production

Seedlings of *Schinus molle* produced in substrates with biosolid proportions of 50 and 100% did not statistically differ in average shoot height, at the age of 18 months after transplantation, from the one produced with substrate composed by 50% of cattle manure (Figure 1). For this species the treatment that promoted lower average growth in shoot height was 25% biosolid when compared to the other three treatments.

Considering *Pelthoporum dubium* and *Licania tomentosa*, seedlings produced with substrate composed of 100% biosolid showed superior average growth in shoot height when compared to other treatments (Figure 1). However, no statistic difference was detected between treatments T1, T3 and T4 in average shoot height growth in both cases. Substrate composed of 25% biosolid showed lower average growth in shoot height and statistically different result from T4 for the two species.

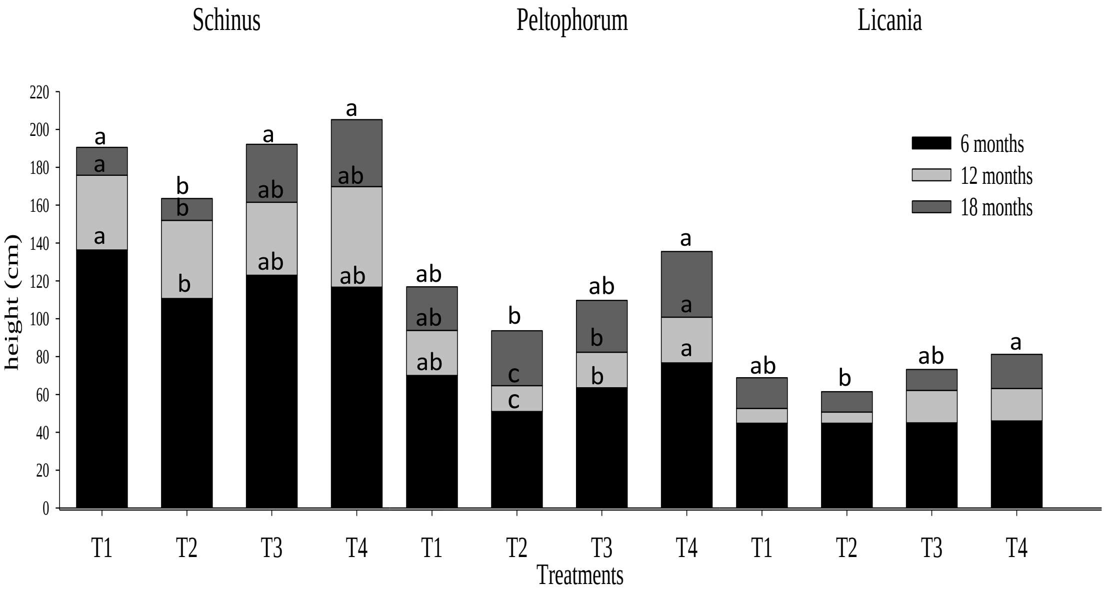

Figure 1: Average shoot height growth of *Schinus molle*, *Peltophorum dubium* and *Licania tomentosa* seedlings in different times after transplantation, produced in a substrate with different proportions of cattle manure, subsoil and sand 50*:40:10 (T1) and three substrates with different proportions of biosolid, subsoil and sand 25:65:10; 50:40:10 and 100, respectively T2, T3 and T4. For each age and species average values followed by the same letter do not statistically differ by the Tukey test ($P \geq 0,95$). No statistically difference was detected for *Licania tomentosa* at age 6 and 12.

Results for mean collar diameter at 18 months for seedlings of the three species are presented on Figure 2. Highest mean value of collar diameter for *Schinus molle* was found in the 50% cattle manure treatment and did not statistically differ from 100% biosolid substrate. Also, for this species, seedlings produced with substrate containing 50% biosolid were statistically equal to 25 and 100% biosolid proportion by the Tukey test.

For *Peltophorum dubium* higher mean collar diameter values were observed in seedlings produced in 100% biosolid and 50% cattle manure, both treatments did not statistically differ for this characteristic at 18 months after transplantation. Experimental treatment containing 50% biosolid did not statistically differ from 50% cattle manure and 25% biosolid for collar diameter mean values.

Seedlings of *Licania tomentosa* showed superior mean growth in collar diameter when produced with 100 and 50% of biosolid proportion. Both treatment results did not statistically differ for this morphological feature. At 6 and 12 months after transplantation no significant difference was detected by the F-Test between treatments. Collar diameter mean value for T3 at 18 months did not statistically differ from T1 and T2.

In the seedlings production of all three species lowest mean values for collar diameter were observed for substrate composition of 25% proportion of biosolid.

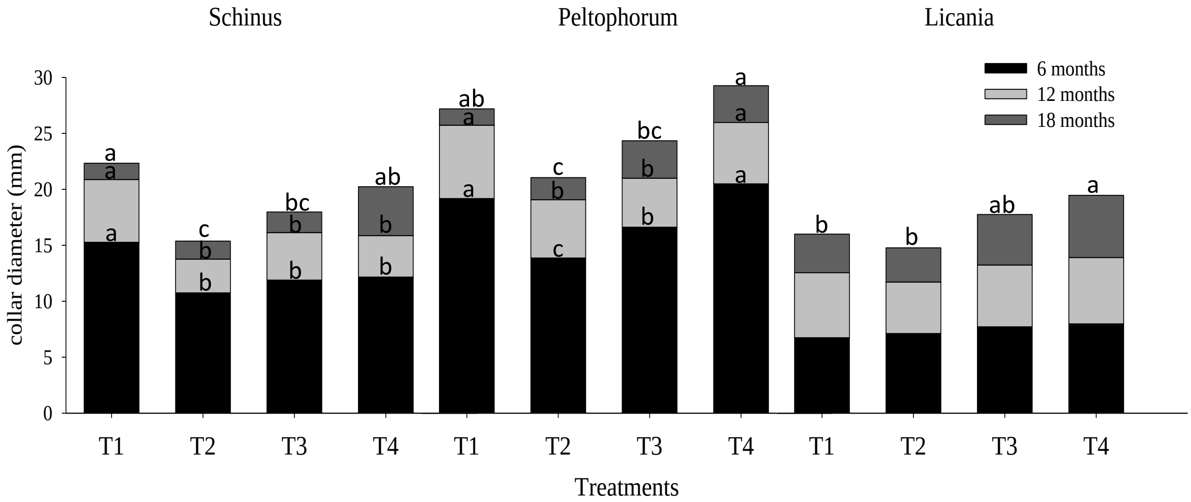

Figure 2: Average collar diameter growth of *Schinus molle*, *Peltophorum dubium* and *Licania tomentosa* seedlings in different times after transplantation, produced in a substrate with different proportions of cattle manure, subsoil and sand 50*:40:10 (T1) and three substrates with different proportions of biosolid, subsoil and sand 25:65:10; 50:40:10 and 100, respectively T2, T3 and T4. For each and species average values followed by the same letter do not statistically differ by the Tukey test ($P \geq 0,95$). No statistically difference was detected for *Licania tomentosa* at age 6 and 12.

5.2. Soil Analysis

Table 3 shows chemical properties of the different growing substrates used in the experiment in three different ages. Lower values of pH were observed in 100% biosolid substrate as well as higher contents of N, P and C when compared to other treatments in all ages. Substrate with 50% of cattle manure in the composition presented superior K^+ content in all ages and Al^{3+} values equals to zero.

Table 3: Chemical analysis result for soil fertility of four different substrate compositions used in the production of urban afforestation species, in three different times after transplanting

Time	Treat.	pH	P	K^+	Ca^{2+}	Mg^{2+}	Al^{3+}	H+Al	N	C
			----- mg L^{-1} -----		------------ $cmol_c$ dm^{-3} -----------				---g kg^{-1}--	
0	T1	7,0	143,4	566,6	4,15	2,27	0,00	0,66	1,9	-
	T2	5,7	234,0	44,7	4,00	0,58	0,00	3,70	1,6	-
	T3	5,3	462,4	79,7	8,07	0,72	0,02	5,59	3,6	-
	T4	4,7	743,7	197,5	1,75	4,11	0,15	12,59	11,5	50
4	T1	7,6	187,9	551,9	6,30	2,74	0,00	0,38	2,3	21
	T2	5,1	244,2	53,5	6,71	0,90	0,03	3,66	1,7	14
	T3	4,9	418,1	82,3	12,87	0,95	0,06	5,48	3,6	27
	T4	5,0	607,7	141,1	8,85	0,22	0,07	12,52	10,6	69
8	T1	7,8	155,6	311,2	6,45	2,74	0,00	1,85	2,1	18

continue.......

Continuacion ....

T2	5,2	214,7	34,0	5,26	0,76	0,04	5,22	1,9	13
T3	4,9	358,8	57,4	11,81	0,93	0,10	13,37	4,2	30
T4	4,8	655,7	90,5	15,09	1,20	0,15	12,72	12,8	33

Al, H + Al -Titration method; Ca and Mg- Atomic Absorption; K - Flame Photometry; N - Kjedahl Method; P - Colorimetric Method; pH - Potentiometric.

Physical characteristics assessed for substrate distinct compositions at 12 months after transplantation are presented in Table 4. Bulk density mean results were statistically equal for 25% biosolid substrate compared to 50% cattle manure and 50% cattle manure to 50% biosolid treatment. Substrate composed of 100% biosolid showed the lowest mean bulk density value. For particle density mean values T1, T2 and T3 treatments presented very similar results and statistically different in comparison to T4.

It is observed that the substrate composed of 100% biosolid presented the highest mean total porosity value and a result statistically different to other substrate compositions. Whereas substrate with 25% proportion of biosolid showed lowest mean total porosity value. It was not possible to identify significant difference between experimental treatments by the F-Test for substrate aggregation. Pictures in Figure 3 show substrate aggregation on each treatment.

Table 4: Physical analysis average results for different substrate compositions used in the production of three urban afforestation species, by the time of 12 months after transplantation

Physical aspect	T1	T2	T3	T4
Bulk density (g cm^{-3})	1,13 ab	1,24 a	1,0 b	0,56 c
Particle density (g cm^{-3})	2,55 a	2,57 a	2,47 a	1,93 b
Total porosity (%)	55,5 bc	51,7 c	59,3 b	70,9 a
Aggregation (grade)	5,0	4,2	3,0	4,0

Treatments are: cattle manure, subsoil and sand 50*:40:10 and three substrates with different proportions of biosolid, subsoil and sand 25:65:10; 50:40:10 and 100. Mean values followed by the same letter, in columns, do not statistically differ by the Tukey test (P ≥ 0,95). For substrate aggregation no significant difference was detected by the F-Test.

Figure 3: Aggregation for different substrate compositions used in the production of three urban afforestation species, by the time of 12 months after transplantation.

6. DISCUSSION

Observing growth results for all three species it is possible to infer that biosolid positively influenced shoot height and collar diameter seedlings development. Fonseca (2015) observed equivalent results for *Dalbergia nigra*, *Cariniana legalis* and *Caesalpinia echinata* producing seedlings in plastic bags of 9,7 x 20 cm and studying morphological features including shoot height and collar diameter. The author observed better results with 80% biosolid content in substrates and affirmed that induced growth was due to increasing organic matter and available nutrients. The improved growth mainly in shoot height for *Schinus molle* and *Peltophorum dubium* in contrast to *Licania tomentosa* is expected since both species present pioneer characteristic and accelerated initial development (LEAL, 2015; CABREIRA et al., 2017b).

Despite liming undertook in subsoil during substrates composition it is observed that increasing biosolid contents induced lower pH values compared to cattle manure (Table 3). Equivalent results were observed by Bezerra et al. (2006) that applied sewage sludge in degraded area revegetation and observed lower pH values for higher doses. Organic acids are produced during microbiological decomposition of organic matter and are an important complexation agent for heavy metals and tend to increase substrate acidity (NÓBREGA et al., 2007). However, pH modifications rely on different sewage sludge treatments. Several experiments showing acidity decrease were conducted with sewage sludge that received lime (CaO) during treatment for sanitation and stabilization (BACKES et al, 2009). Biosolid with alkaline characteristic may be used as soil amendments for pH regulation (CORRÊA et al, 2007). Furthermore, for biosolid and cattle manure substrates pH values were outside the range of 5,5 to 6,5 indicated by Malavolta (1989) for improved macro and micronutrients absorption.

Chemical fertility analysis showed that increasing doses of biosolid induced higher doses of N. Moreover, biosolid treatments T3 and T4 presented superior N content than T1 (50% cattle manure). Ribeiro (2017) producing urban afforestation seedlings of *Handroanthus impetiginosus*, *Libidibia ferrea* and *Poincianella pluviosa* observed at six months after transplantation higher contents of N in substrates containing distinct biosolid doses when compared to cattle manure substrate. Working with *Peltophorobium dubium*, *Lafoensia pacari* and *Ceiba speciosa* seedlings Caldeira et al. (2017b) tested one cattle manure substrate and three substrates with increasing biosolid contents. Authors observed that N and biosolid content were positively related and that for all species 80% biosolid substrate promoted higher growth in height and collar diameter.

Sewage sludges are considered a good source of N and an alternative for nitrogen fertilization in agricultural activities (LOBO et al., 2015). According to Caldeira et al. (2017a) N and P are highly demanded during initial development stages and are important nutrients to impulse seedlings growth. Therefore, superior results observed for biosolid treatments in this present work are likely to be caused by the high concentration of such nutrients. Although substrates containing biosolid presented higher concentrations of N it is possible to observe in Table 1 that for total N content, cattle manure held higher concentrations of N than biosolid. The majority of N portion (about 80%) found in sewage sludge is in the organic compartment and therefore is readily available for plants uptake after organic matter degradation (CARVALHO et al., 2015; ABREU et al., 2017b). Biosolid substrates are able to gradually provide high levels of nitrogen readily available to the system attending plants nutritional demands more efficiently than common artificial chemical fertilizers (ASSENHEIMER, 2009).

It is possible to observe that concentrations of P in substrates followed similar pattern to N meaning that higher biosolid doses demonstrated higher concentrations (Table 3). Treatment with 100% biosolid provided the highest P concentration in substrate whereas 50% cattle manure treatment had the lowest concentration for this nutrient. This result was consistent for both substrate chemical fertility and total content analysis. Phosphorus is an important nutrient for plants initial growth inducing root system development and enabling important process such as energy transference and photosynthesis (CABREIRA et al., 2017b). Bugbee (2002) tested treatments with increasing biosolid doses (0, 25, 50 and 100%) for the production of flowering annuals, herbaceous perennials and woody ornamental shrubs and detected the increase of N, P, K, Ca and Mg to sewage sludge addition. The author also stated that the best biosolid concentrations for the production of studied plants were 50 and 100% thus affirming that biosolid is suitable for high concentrations application.

Sewage sludge is a potential source of phosphorus for forest species and agronomical crops and a more sustainable alternative for traditional P fertilizers (SILVA et al, 2001; MARTINEZ et al, 2014). Lima Filho (2015) produced *Ceiba speciosa* seedlings in 18L pots testing four substrates with different biosolid contents, a control treatment and one substrate treatment containing super simple phosphate. Comparing equivalent treatments in P_2O_5 between biosolid doses and phosphorus fertilizer application the author observed that seedlings presented significantly higher growth when produced in biosolid substrate. According to Paiva et al. (2009) native forest species are highly adapted to using phosphorus concentrations available in organic matter for instance from litter cycling. Therefore, phosphorus contained in biosolid organic matter is gradually released into the system and may be more effectively absorbed by plants in contrast to more soluble sources, including super simple phosphate, which can easily be adsorbed in soil colloids (LIMA FILHO, 2015).

Cattle manure presented a K total content of 4,5 g kg^{-1} against 2,1 g kg^{-1} for biosolid at the same age. Chemical fertility results for treatments T2, T3 and T4 also showed smaller K^+ concentrations in contrast to 50% cattle manure treatment. This result is corroborated by Nascimento (2016) and Abreu et al. (2017b) that characterized sewage sludge from several stations both in São Paulo and Rio de Janeiro, respectively, and observed low K average rates. This aspect is directly related to sewage sludge treatment process due to potassium’s ionic form and high solubility in water. Consequently, potassium is diluted and solubilized in the wastewater thus being lost during dehydration (ABREU et al., 2017b).

Soil fertility chemical analysis demonstrated that at ages 0 and 4 treatment containing 50% of biosolid showed the highest Ca^{2+} concentration and at 8 months after transplantation 100% biosolid presented the highest concentration. Considering total content analysis biosolid and cattle manure showed very similar Ca concentrations at age 0. According to Silva et al. (2001) sewage sludge increases soil fertility through nutrients contribution mainly Ca, P, S and Zn. For the production of *Schinus terebinthifolius* seedlings, Nóbrega et al. (2007) observed that increasing doses of biosolid in substrate composition elevated concentrations of P, K^+, Ca^+, Mg^{2+} and CEC. Sewage Treatment Stations that employ liming for sludge sanitation tend to produce biosolids with higher Ca^{2+} concentrations (CARVALHO et al., 2015; NASCIMENTO, 2016; ABREU et al., 2017b).

Cattle manure showed superior Mg concentrations in comparison to biosolid in both chemical analysis. Observing the chemical fertility analysis T1 presented a steady Mg^{+2} concentration throughout the experimental period and a superior value at ages 4 and 8 after transplantation. Total Mg content showed that for cattle manure was 4,2 g kg^{-1} in contrast to 3,6 g kg^{-1} for biosolid at age 0. According to Abreu et al. (2017b) Mg is found mainly in mineral form in biosolids.

The Al^{+3}concentrations were considered low for all analysed treatments. Treatment containing 100% biosolid presented higher Al^{+3} concentrations. T4 at age 0 presented 0,15 $cmol_c\ dm^{-3}$ for chemical fertility analysis and biosolid presented 35,4 $g\ kg^{-1}$ for total content analysis. Treatments with 50 and 100% showed higher C concentration than cattle manure treatment for chemical fertility analysis at 4 and 8 months after transplantation. Considering the total content analysis at age 0 cattle manure showed higher C concentration than biosolid. Cattle manure also presented higher concentration of organic carbon and C/N ratio than biosolid.

Sewage sludges that go through long drying processes, as the one produced at the Ilha do Governador STS, tends to present a lower C concentration and consequently lower C/N ratio due to the exposure to microorganism in the presence of moisture and oxygen (CARVALHO et al., 2015; ABREU et al., 2017b). Nascimento (2016) aiming to evaluate the agricultural potential of sewage sludge produced in São Paulo Plant assessed sludges produced in 19 different stations and found a mean C/N ratio of 8 thus consistent to the present work. However, results are contrasting with those found by Guerrini et al. (2017) whom assessed the impact of biosolid and cattle manure application on urban soil fertility and tree growth. The authors found statistically higher contents of OC in soils treated with biosolid associated to slag and justified such behaviour to the potential complexation of highly reactive organic matter by slag thus leading to soil organic matter stabilisation. On the other hand, they assessed a rapid decrease of OC in the presence of cattle manure probably induced by a higher C/N ratio found in this material. The elevated N concentration as the low C/N ratio found for biosolid in this work favours mineralization processes and validates the potential use for nutrients source (FERREIRA et al., 2013; NASCIMENTO, 2016).

Moreover, for chemical total content analysis at 0 months after transplantation cattle manure presented 27,08 % organic matter content and biosolid 15,39%. Results for biosolid are below the ones found by Abreu et al. (2017) which found values ranging from 51,6% to 57,9% studying sewage sludge from different STS in Rio de Janeiro. However, Carvalho et al. (2015) verified that most of C and N concentrations in swage sludge are contained in the organic compartment. Ferreira et al. (2013) affirms that organic matter holds the majority of P, S and also is a suitable source of micronutrients. Nóbrega et al. (2007) evaluating sewage sludge as substrate component in the production of *Schinus terebinthifolius* seedlings verified the organic matter increased CEC improvement due to biosolid application. Furthermore, organic matter affects physical attributes including soil structure, substrate aeration, particles aggregation and moisture retention (LIMA FILHO, 2015).

For physical characteristics it is possible to observe on Table 4 that T2 presented the highest average bulk density whereas T4 the lowest. This result was expected since T2 had the highest subsoil content and T4 was exclusively formed by biosolid. Comparing 50% cattle manure to 50% biosolid substrates, although both treatments did not statistically differ T1 presented a higher bulk density than T3. Regarding particle density T1, T2 and T3 showed similar average values and did not statistically differed while T4 showed the lowest mean value and statistically different from other treatments. Corroborating previous results T4 presented the highest total porosity mean value and statistically different from the others and T2 the lowest total porosity. Results corroborate with Sampaio et al. (2012) that evaluate the effects of sewage sludge application in increasing doses on physical soil properties. Authors verified aggregate formation, soil porosity and soil humidity increase mainly due to the formation of bigger aggregates and organic matter addition. De Maria et al. (2007) and Campos and Alves (2008) both verified positive influence of biosolid application on Red Latosols including macro porosity, aggregates diameter and organic matter increase.

Lower density substrates incur on lighter seedlings containers and provide an advantage for urban afforestation planting operations. Lighter seedlings may decrease transport and mobilization costs within and from nursery to planting site. Since urban afforestation seedlings are produced in larger containers choosing less dense substrates for weight reduction can support forest nurseries in reducing costs and increasing profits.

For substrate aggregation T1 showed best result (Figure 3) nonetheless no significant statistically difference was detected between treatments by the F-Test. According to Sampaio et al. (2012) sewage sludge characteristic improves particles aggregation thus positively influencing substrates aggregation. Biosolids are suitable for soil and substrates amendments due to high organic matter contents that induce aggregates formation and stabilization. Enhanced soil structure leads to improved bulk density, porosity, water capacity, CEC, aeration, drainage, soil fauna and soil erosion and leaching mitigation (LU et al., 2012).

7. CONCLUSION

Biosolid presented potential for the production of urban afforestation seedlings. Regarding the production time and experimental conditions, substrate containing 100% biosolid favoured seedlings growth in collar diameter and shoot height and is considered suitable for the production of urban afforestation seedlings of *Schinus molle* L, *Licania tomentosa* (Benth) Fritsch. and *Peltophorum dubium* (Springer.). In general terms, seedlings produced with biosolid presented similar or superior average growth when compared to cattle manure in the same proportion.

In order to perform the best choice for substrate material it is important to consider input transport from source to forest nursery and material availability in the region. Taking into account the need for a proper use of sewage sludge and the demand for organic matter input in the production of urban afforestation seedlings this work demonstrates that biosolid use as substrate is a feasible alternative for final disposal.

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