







## SCIENTIFIC ARTICLE

## Regrowth and ornamental traits of bermudagrass fertilized with sewage sludge

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### Abstract

The sewage sludge is a low-cost material and sustainable alternative to substitute chemical fertilizers on ornamental lawns and gardens. Thus, the objective was to evaluate the effects of the application of sewage sludge on the regrowth and ornamental traits of Discovery™ bermudagrass. The experiment was carried out during the fall/winter of 2019. The turf was removed and left the soil exposed for a new grass regrowth. The treatments applied were 0, 357, 714, 1,071 and 1,428 g m<sup>-2</sup> sewage sludge spread evenly on the lawn in a single dose. The evaluations were carried out after 120 days and the soil solution (EC and NO<sub>3</sub><sup>-</sup>), Normalized difference vegetation index, root length, root + rhizome + stolon + leaves volume and digital image analysis were evaluated. The results showed that the increase of sewage sludge positively influenced the turfgrass development, both in the aesthetic aspect and on bermudagrass regrowth. The soil solution can show that the sludge increased the electrical conductivity and NO<sub>3</sub><sup>-</sup> ions; however, it did not hinder the development of the lawn, even having positive correlations between these variables and the biometric evaluations of the plant. It is concluded that the dose of 1,428 g m<sup>-2</sup> presented the best results for the evaluated characteristics, being the recommended one for use in the fertilization of bermudagrass Discovery™.

**Keywords:** biosolid, organic fertilization, ornamental turfgrass, sod production.

### Resumo

#### Características ornamentais e de rebrota da grama bermuda fertilizada com lodo de esgoto

O lodo de esgoto é um material de baixo custo e alternativa sustentável para substituir fertilizantes químicos em gramados e jardins ornamentais. Assim, o objetivo do trabalho foi avaliar os efeitos da aplicação de lodo de esgoto nas características estéticas e na rebrota da grama bermuda Discovery™. O experimento foi realizado durante o outono/inverno de 2019, onde o gramado foi retirado com uma colhedora própria e deixado o solo exposto para uma nova rebrota da grama, e assim foram aplicados 0; 357; 714; 1071 e 1.428 g m<sup>-2</sup> de lodo de esgoto espalhados uniformemente no gramado em dose única. As avaliações foram realizadas após 120 dias, sendo: solução do solo (CE e NO<sub>3</sub><sup>-</sup>), Índice de vegetação por diferença normalizada, comprimento de raiz, volume de raiz+rizoma+estolão+folhas e análise por imagem digital. Os resultados demonstraram que o aumento das doses de lodo influenciou positivamente o desenvolvimento do gramado, tanto no aspecto estético quanto nas avaliações de rebrota do tapete. A solução do solo pôde evidenciar que o lodo aumentou a condutividade elétrica e íons de NO<sub>3</sub><sup>-</sup>, contudo não prejudicou o desenvolvimento do gramado, havendo inclusive correlações positivas entre essas variáveis e as avaliações biométricas da planta. Conclui-se que a dose de 1.428 g m<sup>-2</sup> de lodo de esgoto apresentou os melhores resultados para as características avaliadas, sendo a dose recomendada para uso na grama bermuda Discovery™.

**Palavras-chave:** biossólido, fertilizante orgânico, gramado ornamental, produção de grama.

### Introduction

There has been an accelerated growth of urban centers in Brazil, which has not been accompanied by the basic sanitation; thus, many of these sites do not have an

adequate treatment of the waste generated (Castro et al., 2015). However, the creation of the National Solid Waste Policy (NSWP) Law n° 12,305 of 2010, demands from the Brazilian Government the construction of Sewage Treatment Plants (STP) (Brasil, 2010). At the end of the

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sewage treatment process (separation of the solid from the liquid residue), pollutants, nutrients and contaminants are concentrated in a single mass, called sewage sludge (Pedrosa *et al.*, 2017). Usually, the sewage sludge produced is destined for landfills, generating a huge environmental impact. Consequently, there is a search for alternatives for sewage sludge final disposal, such as its use in agriculture, which is regulated by the National Environment Council (CONAMA, 2006). The decree n° 375, defines the treatment of sewage sludge before its use in agriculture as an organic fertilizer (Martins *et al.*, 2018), due to the presence of pathogenic agents.

Sewage sludge is a material rich in nutrients and organic matter (Zhang *et al.*, 2015; Li *et al.*, 2017) mainly nitrogen (N), the most responsive element in plants (Wang *et al.*, 2019). The presence of organic matter in the residue has a direct influence on physical and biological properties of soils, increasing soil porosity and facilitating water drainage and, avoiding surface runoff (Silva *et al.*, 2018). Several studies have demonstrated the potential of sewage sludge as a fertilizer (Lira *et al.*, 2008; Nobile *et al.*, 2014; Backes *et al.*, 2017; Mota *et al.*, 2019).

The N is the main nutrient involved in the quality, growth and aesthetics of lawns (Lusk *et al.*, 2018). Nitrogen is an important component of molecules involved in chlorophyll synthesis (Santos and Castilho, 2018; Wu *et al.*, 2019), granting an intense green color in lawns, greater density (closed and flawless lawns), reducing the recovery time to mechanical injuries, mainly caused by trampling (Gazola *et al.*, 2016; Santos *et al.*, 2019). Thus, the use of sewage sludge as a source of N, can be a promising alternative to substitute mineral fertilizer (Mota *et al.*, 2019) and to mitigate the impact of sewage sludge production. However, studies on the use of sewage sludge in turfgrass is restricted

to emerald turfgrass (*Zoysia japonica*) (Backes *et al.*, 2017; Mota *et al.*, 2019), and bermudagrass (*Cynodon* spp.) (Nobile *et al.*, 2014).

Discovery™ is the new variety of bermudagrass (*Cynodon dactylon*) (Khan *et al.*, 2017), recently introduced in Brazil, with some specific characteristics, differing from other species of the genus. Its color is composed of a bluish green, being extremely attractive and ornamental, it has a slow vertical growth rate, with pruning requirements 75% lower than others conventional bermudagrass, reducing maintenance fees. It also has good drought tolerance, cold and shade resistance, making it recommended for residential gardens, parks and commercial areas (Emmons and Rossi, 2015; Qually Grama, 2020). Thus, the objective of the work was to evaluate the effects of the application of sewage sludge on the aesthetic characteristics and the regrowth of bermudagrass Discovery™.

## Material and Methods

The study was carried out in an experimental area with a long-lasting planting of Discovery™ bermudagrass. The climate in the region is Cfa (humid subtropical climate with abundant and well-distributed rainfall throughout the year), according to the Koppen climate classification. During the experiment period (autumn/winter 2019), the average temperature was 18.9 °C, 62.59% of relative humidity and 161 mm of accumulated precipitation.

The soil is characterized as a dystrophic red Latosol (EMBRAPA, 2013), with chemical analysis described on Table 1. According to the chemical analysis of the soil, there was no need for liming, because the V% was within the recommended range for sod production (Godoy *et al.*, 2012), since the area had been installed for 1.5 years.

**Table 1.** Chemical analysis of soil in the experimental area.

pH	O.M.	P <sub>resine</sub>	H+Al	K	Ca	Mg	BS	CEC	V%
CaCl <sub>2</sub>	g dm <sup>-3</sup>	mg dm <sup>3</sup>	----- mg dm <sup>-3</sup> -----						
5.0	30	37	35	7.4	44	11	63	98	64

CEC - cation exchange capacity, BS – sum of bases, V% - base saturation percentage, O.M. – organic matter.

The sewage sludge used (Table 2) was obtained from SABESP STS (Sewage Treatment Station), located in Botucatu city, *São Paulo State*, Brazil. The waste went through the disinfection process in the Art. 3 of

CONAMA Resolution n° 375/2006 (CONAMA, 2006), remaining in drying bays for approximately 45 days, producing high temperatures (above 55 °C), which let material dry (21% humidity).

**Table 2.** Chemical analysis of sewage sludge.

N	P <sub>2</sub> O <sub>5</sub>	K <sub>2</sub> O	Ca	Mg	S	Moist.	O.M.	O.C.
----- % (natura) -----								dry
2.8	3.5	0.1	1.5	0.4	0.4	21	37	26
Na	B	Cu	Fe	Mn	Zn	C/N	pH	
--- mg kg <sup>-1</sup> (natura) ----							natura	
1,132	145	185	33,793	259	701	7/1	5.8	

Moist. – moisture, O.M. – organic matter, O.C. – Organic carbon.

To carry out the experiment, 5 cm lawn were removed with a lawn harvester (Classen Hydro-Drive Sod Cutter, SCHV-18/5.5-motor Honda), leaving the soil exposed for new regrowth and lawn growth. After 30 days, the experimental plots of 1.5 m<sup>2</sup> (1 x 1.5 m) with a 0.5 m border were delimited, totaling 15 units, and treatments were applied. The treatments were composed of 0; 357; 714; 1,071 and 1,428 g m<sup>-2</sup> sewage sludge corresponding to 0, 10, 20, 30 and 40 g m<sup>-2</sup> N (Backes et al., 2017), respectively. The doses were spread equally over each experimental plot.

After 120 days of experimentation the following evaluations were performed: a) Ion monitoring of soil solution; b) Normalized difference vegetation index (NDVI); c) Root length and dry biomass volume; and, d) Digital image analysis.

The ion monitoring was performed using 30 cm soil solution extractors, which are composed of tubes with a ceramic end (acts as a filter) in contact with the soil, allowing the access of samples of the aqueous solution of the soil to a pre-established depth, and at the other end, there are two syringes to collect the solution. The extractors were installed 10 cm deep in the soil and a vacuum was applied to remove the air from inside of the extractor, 24 h before the material was collected. The solution was extracted with a syringe and submitted to electrical conductivity (EC) analyzes and NO<sub>3</sub><sup>-</sup> analyses with quick reading kits. According to the adapted methodology of Gomes et al. (2019).

For NDVI, the portable GreenSeeker device was used to diagnostic plant vigor. The equipment emits brief bursts of red and infrared light, which intensity is detected by the equipment and is an indicator of the plant vigor. The sensor displays the values in NDVI that can range from 0.00 to 0.99, measuring the vigor and density of the plants. For root length and dry biomass volume, samples of rhizomes, stolon and leaves from the turfgrass of each experimental plot were collected. The material was washed to remove the adhered soil. The root length was measured, and all samples were dried in an oven at 60 °C, until constant weight for dry biomass.

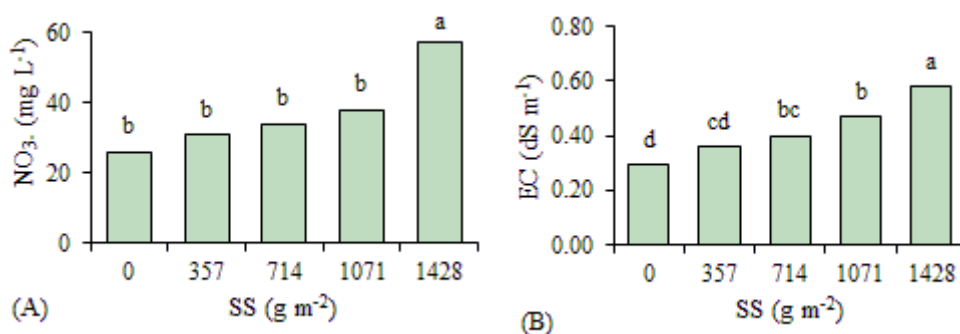
The digital image analysis was performed with a 12 Mp camera, with the support of a structure called “light box”, similar to that produced by Peterson et al. (2011). The

structure is a completely sealed box with lamps connected to a battery, in order to standardize the brightness and the photographed area. The images were transferred to a computer, and the RGB value (Blue, Green and Red) of each image was verified by the Adobe Fireworks® program. As only the green (G) component does not define the green color, depending also on the red (R) and blue (B) components, the RGB results were converted to HSB values (Hue, saturation and brightness), and the Dark Green Color Index (DGCI) was calculated, which varies from 0-1 (Karcher and Richardson, 2003), with the values of G, Hue and DGCI being presented.

The experiment was carried out in randomized 3 blocks with 5 sewage sludge doses. The results were subjected to analysis of variance (ANOVA) and Tukey's test at the level of 5% probability. Results showed an upward straight tendency and in the absence of the maximum point, for that reason regressions were not considered for analysis. Data analysis was performed by the Statistix 10 program.

## Results and Discussion

The N (NO<sub>3</sub><sup>-</sup>) analysis of the soil solution (Figure 1) showed that as the sewage sludge doses increased, the nitrate content in the soil augmented. The higher nitrate contents in soil (40 g m<sup>-2</sup> N,  $p \leq 0.05$ ) was observed on treatment with the highest sewage sludge dose (1.428 g m<sup>-2</sup>). Due to the importance of N fertilization for the crop, the result demonstrates the potential of sewage sludge to supply N on turfgrass maintenance, with positive responses for plant dry biomass (Figure 2C). The availability of N in soil solution is due to the nutritional composition of the residue, in our study sewage sludge had 2.8% N, percentage higher than many organic residues used in agriculture such as charcoal (obtained from pyrolysis of eucalyptus), pine-bark, cattle manure, coconut fiber, eucalyptus sawdust or vermiculite (Higashikawa et al., 2010). The utilization sewage sludge as a fertilizer also enhances organic matter to soils, contributing to the greater availability of N. However, when organic materials are used as fertilizer, the availability of N for plants depends on of organic matter mineralization (Sempiterno et al., 2017).

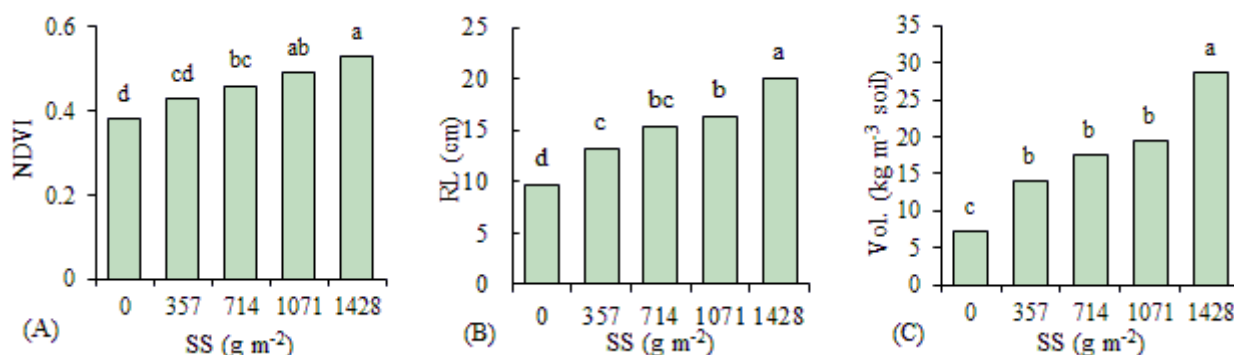


**Figure 1.** Nitrogen in soil solution (A) and electric conductivity (EC) (B) of Discovery™ bermudagrass on sewage sludge (SS) treatments.

The same patterns were observed for electrical conductivity of the soil solution (EC), which increased as the sewage sludge doses augmented. The highest sewage sludge dose (1,428 g m<sup>-2</sup>) resulted in a higher EC (0.58 dS m<sup>-1</sup>). The high nutrients and organic matter contents inserted in the soil by the sewage sludge contributed to the increase of EC in soil solution, which is related to the higher occurrence of ions (Menezes and Matos, 2018). The increase of EC in soil solution indicates the higher greater presence of ions in the soil (Schlotter *et al.*, 2012), and this technique is very accurate to estimate ions contents in soil (Gomes *et al.*, 2019). Organic waste affects soil fertility properties; consequently, other parameters as soil acidity buffering capacity and the waste liming and agronomic

value must be considered than only nutrient composition for sewage sludge use in agriculture (Carmo *et al.*, 2016).

The highest dose of sewage sludge (1,428 g m<sup>-2</sup>) positively influenced the Normalized Difference Vegetation Index (NDVI), with an average of 0.53 (Figure 2A), 39.41% higher compared to control treatment (0 g m<sup>-2</sup>). This index can indicate when the lawn is exposed to stresses such as trampling/compaction, utilization, and water deficit (Kerr *et al.*, 2019; Straw *et al.*, 2020). NDVI can be used as an alternative to evaluate the DCGI in the turfgrass (Caturegli *et al.*, 2020). Higher NDVI indexes can be obtained according to the increase in turfgrass cover, density, and above-ground biomass, which may vary between grass species (Bremer *et al.*, 2011).



**Figure 2.** Normalized difference vegetation index (NDVI) (A), Root length (B) and Root + Rhizome + Stolon + Leaves volume (C) of Discovery™ bermudagrass on sewage sludge (SS) treatments

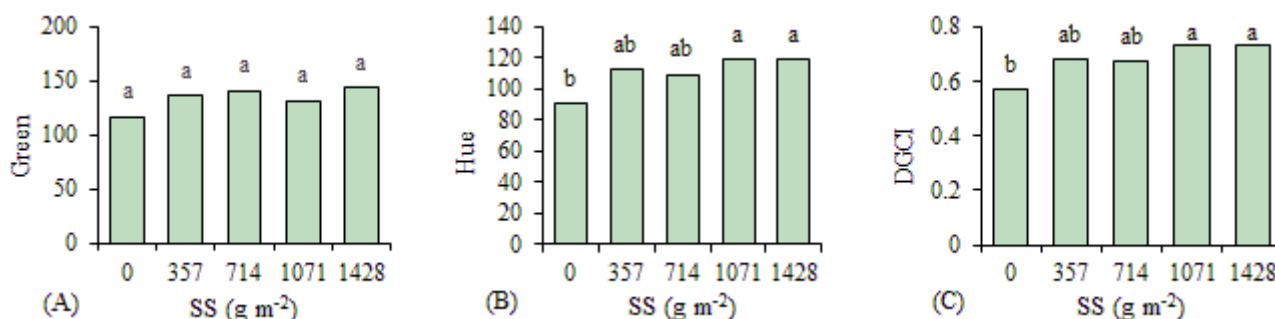
The root length (RL) was superior in treatment 1,428 g m<sup>-2</sup> sewage sludge (Figure 2B), with 20 cm. This variable is extremely important for the establishment and regrowth of the lawns, once higher RL values result in great uptake of nutrients and water, and consequently the formation of lawns in a lesser amount of time. The Root + Rhizome + Stolon + Leaves volume (Figure 2C) reached the highest value (28.67 kg m<sup>-3</sup>) with the application of 1,428 g m<sup>-2</sup> sewage sludge, value 291.67% higher than the control

treatment. These data show the importance of adequate nutrition when soil still exposed, due to the lower regrowth observed in the area of control treatment (0 g m<sup>-2</sup>). Higher production of Root + Rhizome + Stolon + Leaves dry mass of emerald grass was also obtained with high doses of sewage sludge (30 Mg sewage sludge ha<sup>-1</sup>, equivalent to 300 kg ha<sup>-1</sup> of N), and similar to the area that received mineral fertilization (Backes *et al.*, 2017). Backes *et al.* (2010) observed that high doses of sewage sludge,

produced an excessive growth of the shoots in relation to root system. However, Lima et al. (2018) highlights the importance of the superior growth of roots, rhizomes and stolons compared to the shoots, once the formation and resistance of the lawn depends exclusively on these organs. The lower growth of the root system related to high doses

of sewage sludge has already been observed in other plant species, as reported by Zabotto et al. (2020) and Gonçalves et al. (2015).

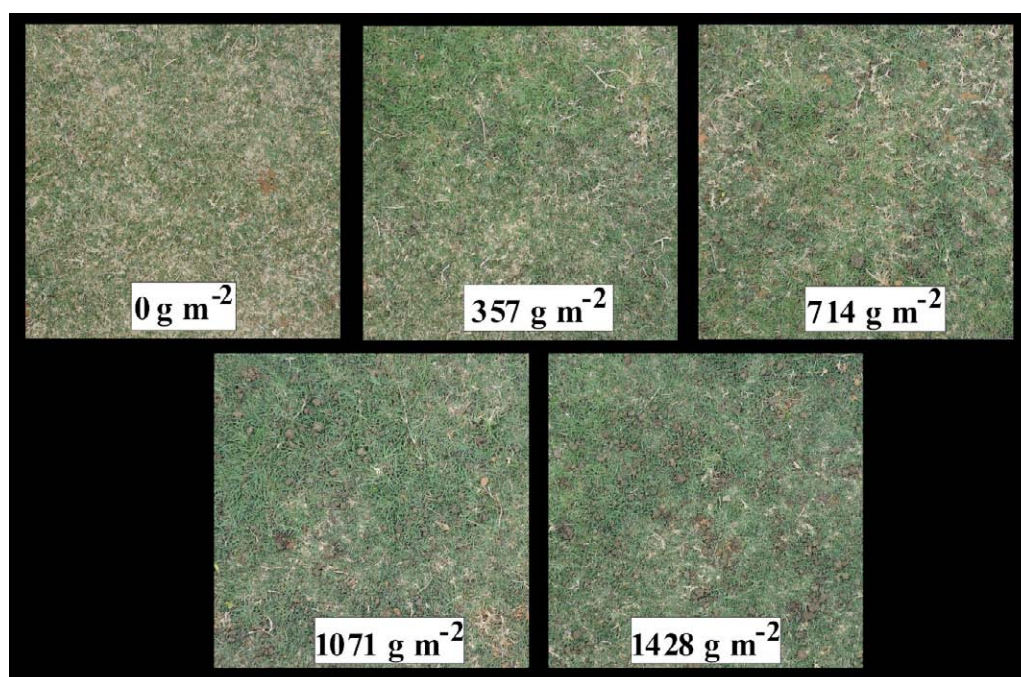
The parameter green component (G) of the RGB color system do not show statistical difference with the application of sewage sludge (Figure 3A).



**Figure 3.** Green component (G) of the RGB color system (A), Hue (B) and Dark Green Color Index (DGCI) (C) of Discovery™ bermudagrass on sewage sludge (SS) treatments.

The G do not showed to be an accurate technique to estimate the color of the lawn, dependent of the blue and red values, as observed by Backes et al. (2010) and Gazola et al. (2016) in color evaluation of emerald grass. However, for the parameters hue and Dark Green Color Index (DGCI) (Figures 3B and C), the doses 1,071 and 1,428 g m<sup>-2</sup> sewage sludge were statistically higher than the other treatments. High doses of N in lawns promote a more intense green color (Godoy et al., 2012; Gazola et al., 2016) due to the increase of leaf chlorophyll (Santos et al., 2019). The hue of the lawn describes the pigment of a color at angles ranging from 0 to 360°, and for lawns, the values are generally between 60 to 120°, where the closer to 120° the more intense the green

color is (Godoy et al., 2012), with the highest doses of sewage sludge (1,071 and 1,428 g m<sup>-2</sup>), with values of 119°. The present study showed that all treatments with sewage sludge results in superior hue (103°) and DGCI (0.63) values to those recommended for bermudagrass (Godoy et al., 2012). Higher DGCI values reflect darker green on the turfgrass, making it a desirable attribute for ornamental lawns (Gazola et al., 2016) as seen in Figure 4. The control treatment had the lowest value, showing that the application sludge influences this parameter. Lower index was also observed in Zeon grass (*Zoysia matrella*) due to low fertilization (Kamimura et al., 2020), demonstrating that the nutritional management may result in distinct DGCI.



**Figure 4.** Digital images of Discovery™ bermudagrass on sewage sludge treatments.

Pearson's correlation analysis showed a significant interaction between the N of the soil solution, volume of Root + Rhizome + Stolon + Leaves ( $p \leq 0.01$ ) and DGCI ( $p \leq 0.05$ ) (Table 3). The correlation shows the linearity between two quantitative variables, that is, the association between them (Figueredo Filho and Silva Junior, 2009). Thus, the data demonstrate the importance of N for the initial development of the crop, contributing to greater growth and production of lawn biomass. Also, the correlation between  $\text{NO}_3^-$  and the DGCI is associated

with an increase in chlorophyll content, reflecting the intense green in lawns, and higher  $\text{NO}_3^-$  concentration more intense is grass color (Oliveira *et al.*, 2018; Santos *et al.*, 2019). Chlorophylls are magnesium porphyrins composed of a central Mg atom, linked to another four of N (Santos and Castilho, 2018; Taiz and Zeiger, 2017). The correlation between nitrogen and green color index was also evidenced in emerald grass in work by Santos and Castilho (2015) and in bermudagrass 'Tifway 419' (Santos *et al.*, 2019).

**Table 3.** Pearson's correlation coefficient.

Treatments	$\text{NO}_3^-$	Volume	DGCI	NDVI	RL	G	Hue
Volume	0.9264**						
DGCI	0.5445*	0.6434*					
NDVI	0.8583**	0.9313**	0.7561**				
CRN	0.8883**	0.9857**	0.7126**	0.959**			
G	0.4292 <sup>ns</sup>	0.5475*	0.236 <sup>ns</sup>	0.448 <sup>ns</sup>	0.5148*		
Hue	0.5679*	0.6654**	0.9761**	0.7778**	0.7275**	0.4048 <sup>ns</sup>	
CE	0.9534**	0.9663**	0.6856**	0.9567**	0.9607**	0.4252 <sup>ns</sup>	0.6965**

\* - significant 5%; \*\* - significant 1%; ns – No significant. DGCI – Dark Green Color Index, NDVI - Normalized Difference Vegetation Index, RL – Root Length, G – Green.

One of the positive factors for the use of sewage sludge in the nutritional management of lawns is the fact that the release of N occurs slowly, due to the mineralization of organic matter. In this way, the possibility of cost reduction in lawn maintenance, once N fertilization can be reduced or even eliminated, depending on the management adopted. Still, other macro and micronutrients are added to the soil, which are essential for plant development. Another important factor with the transformation of sewage sludge into fertilizer is the environmental contribution, significantly reducing the risks of pollution of the environment caused by incorrect waste disposal.

## Conclusions

Our results demonstrate that sewage sludge has the potential to increase fertilizer in Discovery™ bermudagrass. The application of sewage sludge positively influenced the availability of nitrate in the soil solution, the aesthetic traits, and the regrowth of the bermudagrass. It is recommended to use 1,428 g m<sup>-2</sup> sewage sludge to fertilize Discovery™ bermudagrass.

### Author Contribution

**P.S.T.S.:** Field analysis, preparation and writing of the article, critical review. **A.R.Z.:** Field analysis, interpretation, preparation

and writing of the article, critical review and translation. **P.L.F.S.:** Idea of the experiment, field analysis, data collection and analysis, interpretation, preparation and writing of the article and critical review. **M.V.L.N.:** Idea of the experiment, field analysis and data collection. **A.R.T.:** Critical review, translation and approval of the final version of the article. **R.L.V.B.:** Critical review, approval of the final version, work advisor.

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