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MINERAL AND ORGANIC FERTILIZERS INFLUENCE THE ATTRACTION OF *AVENA STRIGOSA* ROOTS AND THE BEHAVIOR OF *FOLSOMIA CANDIDA*

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Resumo

O objetivo desse trabalho foi avaliar os efeitos de doses crescentes de fertilizantes mineral e orgânico na quimiotaxia de raízes de *Avena strigosa* e fuga de *Folsomia candida*. Cinco amostras de solo foram usadas para a fertilização mineral com NPK e outras cinco amostras de solo foram usadas para a fertilização orgânica com fertilizante orgânico a base de cama de aves. O experimento inclui solo controle (sem aplicação) e solos contendo 4-, 8-, 16- e 32-ton de fertilizante por ha. *A. strigosa* foi plantada e cultivada nos solos por 14, 21, 28 e 35 dias e avaliado o comportamento de fuga de *F. candida*. A quantidade de raízes nos solos contendo fertilizante mineral foram maiores que aquelas nos solos contendo fertilizante orgânico a base de cama de aves. O comportamento de fuga de *F. candida* dos solos contendo fertilizante orgânico a base de cama de aves aumentou de 8- para 32-ton ha^{-1}. Aplicações no solo de altas doses de fertilizante orgânico a base de cama de aves aumenta o efeito tóxico para *A. strigosa* e *F. candida*.

Palavras-chave: bioindicadores, ecotoxicidade, fertilização, cama de aves, quimiotaxia.

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Abstract

The aim of this work was to evaluate the effects of increasing doses of mineral and organic fertilizers on the root chemotaxis of *Avena strigosa* and avoidance of *Folsomia candida*. Five soil samples were used for the mineral fertilization with NPK and other five soil samples were used for the organic fertilization with poultry litter-based organic fertilizer. The experiment included control soils (without applications) and soils containing 4-, 8-, 16- and 32-ton fertilizer per ha. *A. strigosa* was planted and cultivated in the soils for 14, 21, 28 and 35 days and evaluated the avoidance behavior of *F. candida*. The root amounts in the soils containing mineral fertilizer were higher than those of the soils containing poultry litter-based organic fertilizer. The avoidance behavior of *F. candida* from the soils containing poultry litter-based organic fertilizer increased from 8- to 32-ton ha^{-1}. Applications of high poultry litter-based organic fertilizer doses in soil increase the toxic effects for *A. strigosa* and *F. candida*.

Keywords: bioindicators, ecotoxicity, fertilization, poultry litter, chemotaxis.

1. Introduction

Poultry litter-based organic fertilizers have been widely applied in soils with the aim of improving the agricultural production indices (GUIMARÃES et al., 2016). The organic fertilization is an efficient, viable and sustainable strategy for the soil management compared to synthetic mineral fertilization (GEREMIA et al., 2015). The organic fertilization also contributes to increase the productivity of different plants and preserve the environment.

However, inadequate (excessive and/or continuous) application of animal wastes as soil fertilizers may generate environmental pollution resulting in environmental risks and loss of ecosystems services, i.e., nutrient cycling, habitat for soil organisms (URRA et al., 2019). Sometimes, it is necessary to evaluate the effects of doses of organic fertilizers in agricultural soils on the plants and soil organisms (such as Collembola) the aim of obtaining a better understanding of the environment and use appropriate doses to do not prejudice these organisms.

Black oat (*Avena strigosa*.) is an important winter crop in the world, are very efficient in nutrient cycling due to their large dry matter production capacity and the aggressive root system (SORATTO and CRUSCIOL, 2008). Roots are responsible for plant support in the soil and nutrient absorption, growing due to occurrence of chemotaxis (SILVA and DELATORRE, 2009). The development of the root system is related to the availability of nutrients (CECATO et al., 2001), among other factors, such as bulk density, acidity. Nutrient availability, i.e. phosphorus and nitrogen, is a common soil restriction for plant growth and development (SILVA and DELATORRE, 2009), situation that occur for short time to organic fertilizers. A well-developed root system is essential for healthy plant growth and development. The amount and effective sources of nutrients are important management practices to improve plant root systems (Fageria e Moreira, 2011) and consequently, higher yields.

Collembola is one of the most abundant organisms in agricultural soils and are responsible for the decomposition of organic matter and nutrient cycling, being the best representative for evaluating soil quality (BUCH et al., 2016). The springtail species *Folsomia candida* was selected due to its widespread use in ecotoxicological studies and is known to be sensitive to a widespread group of chemical substances (BUCH et al., 2016; MACCARI et al., 2016; SEGAT et al., 2015; ZORTÉA et al., 2015). Therefore, the study of the response of Collembola is of great interest, principally for organic fertilizers, that have a complex matrix with many potential contaminants (RENAUD et al., 2017).

Better understand how that mineral and organic fertilizers influence the development of roots and in the presence of springtails, is very important to protect the structure and functioning of soil communities, being an essential factor for the maintenance of ecosystem services. Thus, the hypothesis of the study is the poultry litter will favor the growth of roots of *A. strigosa* in its direction and the presence of *F. candida*. The aim of this work was to evaluate the effects of the application of doses of mineral and organic fertilizers in agricultural soils on the root chemotaxis of *A. strigosa* and avoidance rates of *F. candida* from the poultry litter-based organic fertilizer enriched soils.

2. Material and Methods

2.1 Soil samples

Oxisol Rhodic Hapludox, further designated by Oxisol (SOIL SURVEY STAFF, 2014) samples with clayey texture were collected at depths ranging from 0 to 20 cm in the West of the State of Santa Catarina, Brazil [27º05'274" S and 052º 38'085" W]. Afterwards, the soil samples were homogenized, dried in an oven at 65.0 ± 1.0 °C, milled, sieved to 2.0 mm using a granulometric sieve. The pH values of the Oxisol samples were adjusted to 6.0 ± 0.5 by adding calcium carbonate, and humidity adjusted to 65% of the maximum water absorption capacity prior to mineral and organic fertilization. A tropical artificial soil containing 75, 20 and 5% of thin sand, kaolin and coconut fiber, respectively, was used as control in the avoidance assays of *F. candida*. The physical-chemical parameters of the natural and artificial soils were, respectively: clay 73 and 9%; cation exchange capacity at $pH_{7.0}$ 8.89 and 1.2 $cmol_c\ dm^{-3}$; pH (H_2O) 5.4 and 6.2; organic matter 2.0 and 2.2%; P 4.0 and 11.1 mg dm^{-3}; K 116 and 262 mg dm^{-3}; Ca 4.2 and 2.0 $cmol_c\ dm^{-3}$; Mg 1.3 and 0.9 $cmol_c\ dm^{-3}$; Al 0.0 and 0.0 $cmol_c\ dm^{-3}$; H+Al 3.09 and 1.8 $cmol_c\ dm^{-3}$.

2.2 Assays of root chemotaxis

The treatments consisted in the use of organic fertilizer doses in the following amounts 0- (control soils), 4-, 8-, 16-, and 32-ton ha^{-1}. The physical-chemical properties of the poultry litter-based organic fertilizer was: humidity 5.83%; P_2O_5 4.59%; K_2O 1.65%; nitrogen 2.11%; organic carbon 27.06%; calcium 11.80%; magnesium 1.23%; sulfur 1.07%; pH 6.5. Based on the levels of nutrient inclusion via organic fertilizer (Table 1), the levels of N, P and K were calculated, which served as a basis to formulate the mixtures for the mineral treatment, in other words, the treatments were isonutritive for N, P and K, and differed only as to the source of the nutrients. The mineral source was composed of urea (45% of N), triple superphosphate (42% of P_2O_5) and potassium chloride (60% of K_2O), mixed, homogenized and added to the soil.

Table 1. Contents of available nitrogen, phosphorus and potassium in the soils after organic fertilization with doses of 4-, 8-, 16- and 32-ton poultry litter-based fertilizer per ha soil.

Available nutrient dose in soil (kg ha^{-1})	4	8	16	32
Nitrogen	84	169	338	675
Phosphorus	184	367	734	1469
Potassium	66	132	264	528

The root chemotaxis studies were performed according to the following procedure. Firstly, 10 cm diameter cylindrical vessels were divided by metallic plates. A side of each vessel received 550 g of mineral fertilizer-enriched soil and the other side received 500 g of poultry litter-based fertilizer-enriched soil. In total, each vessel received 1050 g of soil previously fertilized. Seeds of *A. strigosa* supplied by a local market were planted and cultivated in each side of the vessels for 14, 21, 28 and 35 days, with the aim of evaluating the root growth, development and distribution in different land fertilization systems. Each vessel was considered one experimental unit. It was used a total of 100 units with 20 replicates for each fertilizer dose.

Experimental casual factorial design was studied with 5 levels (fertilizer doses of 0-, 4-, 8-, 16-, and 32-ton ha^{-1}) and two treatments (mineral and organic fertilizer), with five replicates. The experimental units were left in laboratory with controlled conditions of temperature (20±2 °C) and photoperiod (12:12 h). All vessels were weekly irrigated with clean water. The amounts of green and dried masses and root percentages after 14, 21, 28 and 35 of growth were determined according to Silva and Queiroz (2002). Analysis of variance (ANOVA One-way) and t-tests were employed for the data treatment. The results were expressed as mean ± standard deviation.

2.2 Avoidance behavior assays of *F. candida*

The avoidance assays of *F. candida* were performed according to the protocol ISO 17512-2 (ISO, 2008). Spherical plastic flasks (volume: 150 mL; height: 6 cm; diameter: 6.5) were divided in two parts by using metallic plates. 30 g of natural soil (without poultry litter-based organic fertilizer) were added in one part and 30 g of soils containing 4-, 8-, 16-, and 32-ton poultry litter-based organic fertilizer per ha soil were added in the other part. Next, twenty springtails with ages from 10 to 12 days were placed in each part. Finally, the metallic plates were removed and experimental procedure conducted at temperature (20±2 °C) and photoperiod (12:12 h) for 48 h, considering the standards for validation. Afterwards, the soils containing springtails were separately transferred to clean plastic flasks for counting.

A similar procedure was used for the dual-control test (with natural and artificial soils) where the control soil was added to each part of the spherical plastic flask. This dual-control test was used to validate the main avoidance test. The avoidance percentages (A, %) were calculated using the formula: $A = ((C - T)/N) \times 100$, where: A = percentage of avoidance, C = springtail number in the control soil, T = springtail number in the poultry litter-enriched soil, N = total number of springtails in the system. The total number of springtails found in the control and poultry litter-enriched soils were evaluated using the Fisher exact test, which is applied considering a 95% significance level ($p \leq 0{,}05$).

3. Results and Discussion

3.1 Root chemotaxis

Contrary to expected (first part of the hypothesis), in the initial growth stage, the roots preferred the soil fertilized with mineral fertilizer than organic fertilizer. A higher root percentage was determined in the soil containing mineral fertilizer equivalent to 4-ton organic fertilizer per ha soil compared to the soil containing mineral fertilizer equivalent to 16-ton organic fertilizer per ha soil, after 14 days (Table 2). This result can be related to the sources of chemical fertilizers used that can cause effects on pH and salinity (FERNANDES, 2006). The lower root percentage in the soils containing 4-ton poultry litter-based fertilizer per ha soil, is due to the fact that organic fertilization needs much longer time to release nutrients (CORRÊA and MIELE, 2011), than mineral fertilizer. Higher doses (16-ton) can supply the initial amount of nutrients, as well as maintain an environment with higher humidity (CORRÊA and MIELE, 2011).

Table 2. Root percentages in the soils containing doses of poultry litter-based organic fertilizer (PL-BOF) and NPK-based mineral fertilizer (NPK-BMF) after 14, 21 and 35 days.

Fertilizer Concentrations (ton ha $^{-1}$)						
Fertilizer	0	4	8	16	32	p-value
14 days						
NPK-BMF	33.98^{Bab}	70.73^{Aa}	62.74^{Aa}	19.6^{Bb}	46.29^{ab}	0.0008
PL-BOF	66.02^{Aab}	29.26^{Bb}	37.26^{Bb}	80.39^{Aa}	53.71^{ab}	0.0008
p-value	0.0100	0.0012	0.0062	0.0001	0.5300	
21 days						
NPK-BMF	70.31^{A}	70.43^{A}	38.24	50.46	47.2	0.0967
PL-BOF	29.69^{B}	29.57^{B}	61.76	49.54	52.8	0.0967
p-value	0.0058	0.0056	0.0989	0.9477	0.7207	
35 days						
NPK-BMF	64.21^{A}	76.99^{A}	41.23	74.64^{A}	69.12^{A}	0.0993
PL-BOF	35.79^{B}	23.01^{B}	58.77	25.36^{B}	30.88^{B}	0.0993
p-value	0.0487	0.0004	0.2166	0.0011	0.0092	

^{AB} – Means followed by capital letters in the columns are statistically different by the F-test at a 95% significance level ($p \leq 0.05$). ^{ab} – Means followed by small letters in the lines are statistically different by the Tukey test at a 95 % significance level ($p < 0.05$).

A higher root percentage was determined in the soil containing mineral fertilizer equivalent to 4-ton organic fertilizer per ha soil compared with the other doses, after 21 days (Table 2). Moreover, the growth and development of the roots were affected by the fertilizer type due to nutrient bioavailability in the soils. It can be result of the higher nutrient bioavailability in the soil containing mineral fertilizer than soil containing organic fertilizer. Generally, it has been observed slow nutrient release in agricultural soils containing poultry litter-based organic fertilizers during its degradation (GOMES et al., 2018). Thus, beneficial effects of poultry litter can be observed in longer periods of time (since it takes much longer time for microorganisms to act to perform mineralization), mainly due to the addition of organic matter and its influence on the chemical, physical and biological properties of the soil (CORRÊA and MIELE, 2011; VALADÃO et al., 2011).

No significant interaction was found between factors at 28 days ($p > 0.05$). However, significant effects were found by varying individually the fertilizer types ($p < 0.05$). The most of the roots were found in the soil containing mineral fertilizer 71.24%, whereas in the soil containing organic fertilizer was 28.76%. This is related with the root migration from the nutrient-poor soil areas to soil areas with high bioavailable nutrient concentration. Mineral fertilization promotes an initial start of plant development, due to the immediate supply of nutrients in the early stages of plants (MARQUES et al., 2014).

Finally, a significant interaction between the fertilizer types and doses was also observed after 35 days of experiment, although the fertilizer type influenced more significantly the root distributions (Table 2). Higher root percentages were observed with doses of 4-, 16-, and 32-ton mineral fertilizer equivalent to 4-ton organic fertilizer per ha soil. However, similar root growth and distribution were found with doses of 8-ton ha^{-1} for both organic and mineral fertilizers. These results can be associated to bioavailability of nitrogen and potassium in soil and their interactions with the plant roots

(SILVA and DELATORRE, 2009). In general, mineral fertilizer is more appropriate for the initial growth and development of plants due to fast nutrient release in soil, whereas poultry litter-based organic fertilizer is more appropriate for the final stages of growth and development of plants due to slow nutrient release. In this sense, a mixture of these two fertilizers could be efficient for all stages of growth and development of plants.

Nitrogen is fundamental for the growth and development of aerial parts and roots of plants (GUO et al., 2019). Its release in soil is faster in presence of mineral fertilizers compared to organic fertilizers (COSTA et al., 2011). It explains the higher root percentages of *A. strigosa* in the soils containing mineral fertilizer, mainly in the first stages of growth. It can also be inferred that the root volume is an indicative of environmental stress and pollution during the growth and development of plants (SILVA et al., 2016). The root length and mass can increase in soils containing a mixture of organic and mineral fertilizers compared to the use of chemical fertilizer only (HATI et al., 2006). Then, a mixture of poultry litter-based organic fertilizer and NPK-based mineral fertilizer could be an excellent alternative for the fertilization of either Oxisol.

3.2 Avoidance behavior of *F. candida*

The ecotoxicological tests based on avoidance behavior of springtails were in agreement with the validation standards. When exposed to soil containing 4-ton poultry litter-based organic fertilizer, *F. candida* did not show an avoidance behavior (Figure 1), where 72.9% of the springtails preferred the soil containing poultry litter-based organic fertilizer. It was associated to high nutrient amounts in organic fertilizer-enriched soils. The use of poultry litter-based organic fertilizer increases the nutrient concentration in soil for the growth and development of plants (HAN et al., 2016), in addition to be a food source for edaphic organisms (GEREMIA et al., 2015).

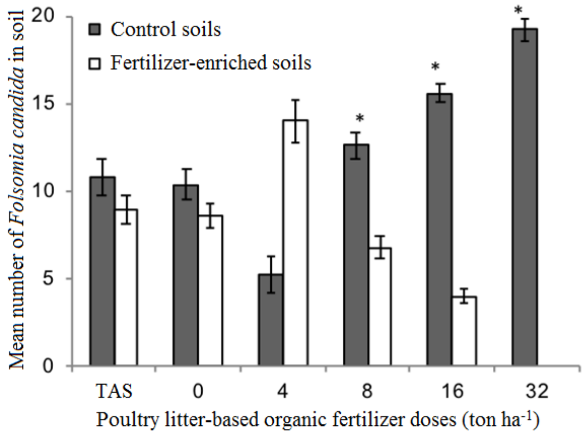

Figure 1. Avoidance assays of *F. candida* in soils containing poultry litter-based organic fertilizer.

The avoidance behavior increased with the organic fertilizer doses from 8- to 32-ton ha^{-1} (Figure 1), confirming our second part of the hypothesis. It confirms that the tolerable limits of poultry litter-based organic fertilizer in land use systems of the State of Santa Catarina ranges from 1- to 4-ton ha^{-1} (CQFS-RS/SC, 2016). Poultry litter-based organic fertilizer doses higher than 4-ton ha^{-1} increase the nitrogen concentrations in soils (84.4 kg ha^{-1} 4- to 675.1 kg ha^{-1} in 32-ton ha^{-1}), increasing the avoidance behavior percentages of springtails from enriched soils to control soils (SEGAT et al., 2015). Moreover, the nutrient excess in soils increases the toxic effects for plants and soil fauna (MACCARI et al., 2016). It is important to note that poultry litter may contain other contaminants, such as Cu and Zn (CORRÊA and MIELE, 2011), insecticide residues (ZORTÉA et al., 2015) and veterinary medicines (HANH et al., 2012), which do not were measured, but that may have contributed to the toxicity of poultry litter in the springtails.

4. Conclusions

The mineral fertilization improves the initial growth and development indices of *A. strigosa* due to the immediate supply of nutrients. For other hand, roots were negatively affected with doses of poultry litter-based organic fertilizer, after 14, 21, 28 and 35 days. For Collembola, the avoidance behavior percentages of *F. candida* from 8-ton ha^{-1}. Thus, applications of high poultry litter-based organic fertilizer doses in soil can increase the toxic effects for *A. strigosa* and *F. candida*, impacting the soil biodiversity and environment.

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