ORIGINAL ARTICLE

Can fertilization and liming affect the amount of litter and roots on *Pinus taeda* forest floor?

A adubação e a calagem podem afetar a quantidade de litter e raízes na serapilheira de Pinus taeda?

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Abstract

The present study evaluated the litter accumulation and the roots contained in the litter as related to different fertilization and liming situations in a commercial stand of *Pinus taeda* (23 years), after five-year application. The study was established in a site of low fertility, located in the forest region of Jaguariaíva - PR. In 2008 an area of 1 ha was selected to establish a subtraction diagnosis, with 7 treatments (Complete; less macro; less micro; less K; less Zn; less lime and control) with 4 replicates. The accumulated litter dry matter, as well as its compartments was quantified: new litter, old litter and humus. The total dry matter of roots contained in litter (≤ 2 mm) and coarse roots (> 2 mm), total root length, root length (fine and coarse) and total volume of the roots contained in the litter were evaluated. No effect of the treatments on the evaluated attributes was verified, except on the dry matter of old litter. A very expressive accumulation of litter in the stands studied (88 to 116 Mg ha⁻¹) was verified mostly in the fine fraction (58.2 to 83.3 Mg ha⁻¹). Dry matter of roots (6 to 8.5 Mg ha⁻¹) and root length found in litter (142 to 176 thousand km ha⁻¹) were also high, being much higher than those found in the literature. The absence of effect of fertilization and liming on the assessed attributes suggests that these practices are not very efficient in stands with more advanced age.

Keywords: Pinus fertilization; Forest site; Decomposition.

Resumo

O presente trabalho avaliou o acúmulo de serapilheira (litter) e as raízes contidas na serapilheira em função de diferentes situações de adubação e calagem, de um plantio comercial de *Pinus taeda* (23 anos), após 5 anos da aplicação. O povoamento estudado foi estabelecido em um sítio de baixo crescimento, situado no polo florestal de Jaguariaíva - PR. Em 2008 uma área de 1 ha foi selecionada para estabelecer estudo de diagnose por subtração, sendo 7 tratamentos (Completo, menos macro, menos micro, menos K, menos Zn, menos calcário e testemunha) com 4 repetições. Foi quantificada a matéria seca de serapilheira acumulada (litter novo, litter velho e litter humificado), além da matéria seca total de raízes contidas na serapilheira, de raízes finas (≤ 2 mm) e raízes grossas (> 2 mm), o comprimento total das raízes, o comprimento das duas classes de raízes (finas e grossas), e o volume total das raízes contidas na serapilheira. Não foi verificado efeito na grande maioria dos tratamentos sobre os atributos avaliados, exceto sobre a massa de litter velho. Foi verificado um acúmulo muito expressivo de serapilheira no povoamento estudado (88 a 116 Mg ha⁻¹), a maior parte de fração fina ($58,21$ a $83,33$ Mg ha⁻¹). A matéria seca de raízes (6 a $8,5$ Mg ha⁻¹) e o comprimento de raízes encontradas na serapilheira (142 a 176 mil km ha⁻¹), podendo ser considerados bastante elevados. A ausência de efeito da adubação e calagem sobre

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os atributos avaliados, sugere que estas práticas são pouco eficientes em povoamentos com idade mais avançada.

Palavras-chave: Adubação de Pinus; Sítio florestal; Decomposição.

INTRODUCTION

Pine plantations are the second forest species planted in Brazil in large areas, occupying 0.72 million hectares. These plantations have been concentrated in the south; the majority in Paraná State with 44% (Indústria Brasileira de Árvores, 2020). In this state, the Jaguariaíva forest region presented more than 160 thousand hectares of pines, which guarantees the supply of wood for several industries related to this sector in the region (Cardoso, 2013).

Pine plantations in Brazil have been established on poor fertility sites despite of its high national average yield (Moro et al., 2014; Motta et al., 2014; Schumacher et al., 2008). However, some sites in the Jaguariaíva region have been producing low yields and show severe symptoms of nutritional deficiency (Batista, 2011). The occurrence of deficiency symptoms was very significant since pine is considered to be of low nutritional demand (Motta et al., 2014). However, not only the poor fertility of the soil is responsible for the occurrence of lack of nutrients; nutrient exhaustion by successive forest harvesting cycles without nutrient reposition may have large impacts as well (Berthrong et al., 2009).

The forest planted under low fertility sites has been shown to have a larger accumulation of litter on soil when compared to high fertility ones (Motta et al., 2014). Hence, it is possible that the lack of nutrients could be responsible for litter accumulation and consequently fertilization and liming could even diminish litter accumulation. However, nutrients and lime application has been shown to have a different effect on litter accumulation; varying from a large decrease (Jandl et al., 2003) to no effect (Moro et al., 2014); and an increase of the amount (Dames et al., 1998). There is a gap of information about this in Brazilian pine plantations.

Furthermore, it is known that roots play a significant role to explain physiologic processes, especially when related to mineral nutrition and water availability in trees (Lopes et al., 2010). On one hand, litter accumulation on the forest floor provides a new environment for root exploration seeking nutrients and water. This may work as a short cut to nutrient cycling, since the root is able to take up the nutrients right after their release from residues and before they reach the soil. There is an indication that it can be a great contribution to nutrient uptake from litter (Motta et al., 2014). On the other hand, the amount of roots within the litter layer can be influenced by plant nutrition (Sayer et al., 2006); and application of K has resulted in a decrease of the amounts of root under native forests (Sayer et al., 2012; Yavitt et al., 2011). In addition, a planted forest with increased nutrient supply resulted in lower fine root length density for litter and soil surface; but the opposite was observed for specific root density (Bakker et al., 2009).

Bakker (1999) also observed that the effect of moderate amounts of lime (1.4 to 2.8 Mg ha⁻¹ CaCO₃) application on oak root growth lasted for 10 to 25 years; suggesting a long-term effect. Shepard & Mitchell (1990) also reported the effects of K application after 40 years of application on growth and plant biomass. They associated it to the effective K cycling in litterfall. Again, there are few studies on the quantification of roots in litter layers from pine plantations (Lopes et al., 2010; Motta et al., 2014) in Brazil. There are also no studies on the effect of fertilization and liming on pine root growth within litter layers.

Therefore, the aim of this study was to analyze the effect of different fertilization and liming conditions on the dry matter of accumulated litter and on the roots contained in it. The experiment was performed in the Jaguariaíva – PR forest region, on a poor soil site.

MATERIAL AND METHODS

Site description

The experiment was conducted in the municipality of Jaguariaíva, Northeast Region of the State of Paraná, Brazil. According to Köppen, the region climate varies from subtropical (Cfa) and temperate (Cfb), with an average annual rainfall and temperature around 1,480 mm and 19°C, respectively. Frosts may occur in the region and there is a decreased precipitation in the winter months (Batista, 2011).

The soil derives from sandstones, belonging to the geological formation Itararé and Furnas. According to the Brazilian Soil Taxonomy, it was classified as Cambissolo Latossólico, with medium to sandy texture. The soil chemical characterization was done by sampling in each experiment block at three depths (0-20, 20-40 and 40-60 cm) with auger. Twenty sub-samples were collected in each block to form a compost sample. The soil samples were analyzed (Table 1) for pH CaCl_2 0,01 M (1:2,5 soil: solution ratio); exchangeable Al, Ca and Mg extracted by KCl 1M, available K and P extracted by Mehlich I, organic C by chemical oxidation by dichromate and (H + Al) by calcium acetate 0,5 M at pH 7.0.

Table 1. Soil chemical properties of the study area before fertilization and liming – Jaguariaíva – Paraná State.

Depth	pH CaCl_2 0.01M	Al	H + Al $\text{cmol}_e \text{ dm}^{-3}$	Ca	Mg	K mg dm^{-3}	CEC* pH 7,0	P g dm^{-3}	C	m%	V %
0-20	4,0	1,5	7,2	0,1	0,1	0,03	7,5	0,7	20,3	86	3,3
20-40	4,1	1,1	6,0	0,1	0,1	0,02	6,2	0,6	15,3	84	3,4
40-60	4,1	0,9	5,9	0,1	0,1	0,02	6,1	0,2	15,5	81	3,5

*CEC = cation exchangeable capacity; m = aluminum saturation of effective CEC; V = base saturation of the CEC a pH 7.

Experimental design

An omission type experiment or diagnosis for subtraction was used with seven treatments in order to evaluate the limiting factor to yield at 18 years of age from a commercial forest plantation. The experimental design was randomized blocks, with four replicates (blocks). The seedlings were planted with three meters spacing between rows and two-meter spacing between plants (3 m x 2 m). The plot had 64 trees, eight lines each with eight plants, in an area of 384 m². The evaluated area was composed by 16 trees centrally located (4 x 4) with two border rows separating plots.

The experimental was established in 2008; an area of 1.5 hectare was selected with the application of the following treatments:

- Treatment 1: Complete treatment: application of all nutrients (N, P, K, Zn, Cu, B, Mo, Ca + Mg in the dolomitic limestone form);
- Treatment 2: Complete treatment minus macronutrients: application of all nutrients with macronutrients (N, P, K) omission;
- Treatment 3: Complete treatment minus micronutrients: application of all nutrients with micronutrients (Zn, Cu, B, Mo) omission;
- Treatment 4: Complete treatment minus K: application of all nutrients with potassium (K) omission;
- Treatment 5: Complete treatment minus Zn: application of all nutrients with zinc (Zn) omission;
- Treatment 6: Complete treatment minus liming: application of all nutrients with limestone (Ca + Mg) omission;
- Treatment 7: Control treatment: control, absence of fertilization and liming.

The amount of fertilizer applied in 2008 in the different treatments, referring to 1 hectare, were: 40 kg of nitrogen (89 kg of urea, 45% of N), 60 kg of P_2O_5 (146 kg of triple

superphosphate, 41% of P_2O_5), 80 kg of K_2O (133 kg of KCl, 60% of K_2O), 3 kg of zinc (20 kg of $ZnSO_4$, 15% of Zn), 2 kg of boron (20 kg of ulexite, 10% of B), 2 kg of copper (8.3 kg $CuSO_4$, 24% Cu), 20 g of molybdenum (61 g of Na_2MoO_4 , 39% of Mo) and 1.3 tons of dolomitic limestone.

The different sources of fertilizers were weighed in the laboratory, brought to the field where they were mixed and applied manually by broadcasting over the litter layer. The lime was applied just after fertilizer application in the same manner.

Sampling and processing of samples

The litter sampling was carried out in 2013, by collecting four samples in each plot. A frame with 10 x 10 cm was placed and pressed against the litter surface. Then, the litter layers were cut with knife and the outside litter part was removed. The litter monolith was gently removed and brought to the lab. This litter was divided into new litter (NL), old litter (OL) and fragmented humus litter (FHL); separated in the field and kept in a 4°C refrigerator until analysis.

The NL and OL were separated into needles, residues (other tissues such as reproductive structures and branches) and roots, when present. The roots from FHL were manually separated using tweezers. Mycorrhizae were taken with the roots. These roots were then separated in coarse and fine roots, using a standard root as a parameter, with a threshold of 1 mm. Using a scanner (Epson Expression 11000XL), they were analyzed in WinRhizo Pro 2013 A software with which it was possible to determine the diameter, length and volume of roots.

After root separation, the FHL was sieved through a 2 mm sieve, dividing it into two fractions: fine (< 2 mm) and coarse (>2 mm). All other separated materials from NL, OL e FHL were dried in the greenhouse at 65°C until constant mass, and then weighed.

The information was extrapolated to the area, obtaining the dry matter of fine roots (≤ 2 mm diameter), dry matter of coarse roots (> 2mm diameter), the total length of roots, length of fine roots, length of coarse roots and total root volume of each sample by hectare.

Statistical analysis

The data was analyzed using the analysis of variance with 5% significance. The assumptions of normality and homogeneity of variance was verified. In case of no normality, variables were transformed. Tukey's test was applied in case of significant difference among data. The statistical analysis was performed on software R (version 3.4.4).

RESULTS AND DISCUSSION

Total, new, old and humus litter

The total litter dry matter (DM) varied from 88 to 116 $Mg\ ha^{-1}$ (Figure 1), considering the three fractions NL, OL and FHL. The obtained values of total litter dry matter values can be considered as high, compared to 9 to 26 $Mg\ ha^{-1}$ found for *Pinus* spp stands in different ages Rio Grande do Sul (Kleinpaul et al., 2003; König et al., 2003) and Paraná state (Watzlawick & Caldeira, 2004). However, Schneider (2011) and Bizon (2005) working in the region of our experiment site found from 55 to 78 and 20 to 91 $Mg\ ha^{-1}$ of total litter, respectively. These results confirmed the high potential of litter accumulation in the region. Additionally, the low soil fertility in the region, also confirmed the possible inverse relationship between soil fertility and amount of litter on the forest floor (Motta et al., 2014). The age of the plantation can also contribute since there is a direct relationship between age and the amount of litter on the forest floor (Haag, 1985; Moro et al., 2014; Viera et al., 2014).

These results reinforce the idea that litter plays an important role for the productivity maintenance in *Pinus* stands on low fertility soils; especially so in stands of later age in which the underlying soil layers have already been depleted (Vogel et al., 2004; Piovesan et al., 2012).

Figura 1. Litter total dry matter from a stand of *Pinus taeda* with 23 years old after 5 years of experiment with fertilization and liming in different treatments – Jaguariaíva – Paraná State – Brazil.

The humus litter corresponds to the major layer of the litter, with average values between 74 and 104 Mg ha⁻¹ (Figure 2). This fraction represents 86% of the total litter dry matter, which comprises most of the root dry matter found in this experiment. The fine and coarse fractions of humus litter varied from 58.2 and 83.3 Mg ha⁻¹ for the fine fraction and 12.7 and 20.7 Mg ha⁻¹ for the coarse fraction (Figure 3). It may be explained by the advanced decomposition stage this part is in, favoring the occurrence of roots. From the results, an average proportion of 4:1 (fine/coarse) could be estimated, indicating that most of the humus litter is composed by fine materials. The variation coefficient (VC) estimated for this proportion was 17,8%, since the distribution of the values obtained from this proportion varied between 6:1 and 2.5:1, considering that there was a high variability in the coarse fraction values.

Figura 2. Humus litter dry matter from a stand of *Pinus taeda* with 23 years old after 5 years of experiment with fertilization and liming in different treatments – Jaguariaíva – Paraná State – Brazil.

Figura 3. Fine fraction dry mass (a) and coarse fraction dry mass (b) of humus litter from a stand of *Pinus taeda* with 23 years old after 5 years of experiment with fertilization and liming in different treatments – Jaguaruaíba – Paraná State.

The new litter and the old litter fractions were 3.9 to 4.8 $Mg\ ha^{-1}$ and 6.7 to 11.4 $Mg\ ha^{-1}$ on average, respectively (Figure 4). Mainly needles composed the new litter (no shown data). In the old litter it was possible to identify other components, such as productivity structures, fine branches and needles (no shown data). However, needles still made up the main compartment, corresponding to 82.7% of the total dry matter in old litter. The variation coefficient for this attribute was low, since the percentage of needles in old litter varied in average from 79.8 to 86.9%. Thus, the data indicate that there is a pattern of the needle proportion and residues in the old litter.

Figura 4. Old litter dry mass (a) and new litter dry mass (b) per hectare from a stand of *Pinus taeda* with 23 years old after 5 years of experiment with fertilization and liming in different treatments – Jaguaruaíba – Paraná State – Brazil.

There was no difference between treatments, except for treatment 7 (control), which clearly indicates that the site is indeed poor, and the fertilization and liming had an effect to increase the FOL content. Also, there is a trend for higher amount of litter on K omission. Early senescence of needles was pointed out as an adaptation to lack of potassium in *Pinus pinaster* stands, with an increase of material deposition (Eimil et al., 2015). Viera & Schumacher (2010) also found that the content of N and K relate negatively with the litter amount in a *Pinus* forest. Residue input, its decomposition rate and rate of humus formation have been responsible for the three major factors that control the amount of litter on the forest floor. Thus, it is possible to infer that on highly productive sites litter decomposition is faster, and in less productive sites the litter layer is thicker (Viera et al., 2014), which could not be found in this experiment.

On the other hand, it can be also inferred that with nutrient input from fertilizers and lime, there would be an increase of dry matter production of litter, but this has not occurred.

Therefore, it seems that fertilization and liming application did not change this balance. In addition, there was indication of a lower input of residue in the control than other treatments, since it yielded the smallest value for the new and old litter fraction. Therefore, it is possible that the increase on decomposition rate with fertilization and liming compensates for the input.

Root matter

The total amount of roots present in the litter varied in average from 6.4 to 8.5 Mg ha⁻¹, as shown in Figure 5. The fertilization and liming did not affect the dry matter amount of roots within the litter layer, except for treatment 5. Somehow, the omission of Zn caused a significant reduction in the roots found in the litter. It can indicate that this nutrient was already a limiting nutrient in the poor fertility soil, since its omission affected the root growth negatively. No explanation could be drawn to elucidate this effect, since no information in the literature was found describing this root biomass reduction. Thus, morpho-physiological studies would be needed in order to understand this effect properly.

Figura 5. Total dry mass of roots contained in the litter per hectare from a stand of *Pinus taeda* with 23 years old after 5 years of experiment with fertilization and liming in different treatments – Jaguariaíva – Paraná State – Brazil. Different letters indicate significant difference between treatments (Tukey test, $P < 0.05$)

The amount of fine roots contained in the litter was high (Figure 6) and varied from 6.4 to 8.6 t ha⁻¹ in average; since other research done in *Pinus* stands by Vogel et al. (2004), Lopes et al. (2010) and Selle et al. (2010) have shown fine root values with 3.5 Mg ha⁻¹ at a maximum, considering whole soil profile. The coarse root matter presented a high variation in values: from 3 to 17 kg ha⁻¹ on average. So, the amount of coarse roots was insignificant compared to fine roots in the litter layer.

The high occurrence of fine roots in litter of *Pinus* can be related to the role of this layer in the recycling processes, provision of nutrients and water, especially under limiting conditions (Motta et al., 2014; Lopes et al., 2010). A strict relation between the litter and fine roots was observed, since the stand is on a poor nutrient soil; acid and with medium texture, which indicates a low water retention capacity. Thus, in order to overcome the low nutrient supply, the roots concentrate in the litter, which may function as an expressive source of nutrients, creating better conditions for root development (Motta et al., 2014).

On the other hand, Sayer et al. (2006) studying the relationship between roots and litter in a tropical forest, found high biomass content of fine roots in litter freshly added to the soil,

202 kg ha⁻¹. The author states that despite seasonal influence, the amount of fine roots also varied with the content of fresh litter added to the soil, indicating a strong relationship between nutrient release during litter decay and fine root growth. Thus, this relationship suggests that root proliferation occurs in response to the more easily obtainable nutrients in the litter, rather than an adaptation to soil fertility.

Figure 6. Fine fraction dry mass (a) and coarse fraction dry mass (b) of litter per hectare from a stand of *Pinus taeda* with 23 years old after 5 years of experiment with fertilization and liming in different treatments – Jaguariáiva – Paraná State.

It is also possible that potassium is highly related to root proliferation. Figure 6 may support this hypothesis, where in the treatment with K omission, there was a higher presence of roots. It suggests that K is potentially a limiting factor on these sites. Following the same logic Sayer et al. (2012), studying the nutrient cycling in tropical forest found that fine roots were lower in added K sites, which indicates that with a higher supply of nutrients, the fine root biomass is decreased, so the plant allocates its energy to another compartment for growth. Using K, Yavitt et al. (2011) also found a lower root biomass in the soil of a tropical forest in Panama, in 4 years evaluations.

Under temperate conditions, Bakker et al. (2009) also found that the addition of nutrients (N, P, K, Ca, and Mg) decreased the *P. pinaster* root density in both litter and soil. On the other hand, Albaugh et al. (1998) found less biomass production under fertilization treatments, which they associated as being a result of greater allocation of metabolics to the foliage (photosynthesizing tissue) over to fine roots. Although it describes the opposite effect of this study, the morpho-physiological concept described by the author may explain the effect found in this study. This may suggest that the plants had allocated more biomass to their fine roots over the foliage tissue under lack of potassium conditions in order to enhance the area exploited by the roots in search of more resources.

Root length

Total length of roots present in the litter varied in average from 142 to 176 thousand km ha⁻¹, as shown in Figure 7.

The coarse root length in the litter had a high variation in the estimated values between 25 and 123 km ha⁻¹, shown in Figure 8. The coefficient of variation estimated was also very high (118.9%).

Figura 7. Total length of roots contained in the litter per hectare from a stand of *Pinus taeda* with 23 years old after 5 years of experiment with fertilization and liming in different treatments – Jaguariaíva – Paraná State.

Figura 8. Length of fine roots (a) and coarse roots (b) contained in the litter per hectare from a stand of *Pinus taeda* with 23 years old after 5 years of experiment with fertilization and liming in different treatments – Jaguariaíva – Paraná State.

Possibly there was no statistic difference among treatment, due to the same reason as for dry matter of fine roots, since there is a similar behavior of dry matter and length of roots. The high values for the length of roots in the litter can be explained by Gonçalves & Mello (2004), in which the trees tend to produce longer roots in order to explore higher volume of soil, especially under conditions with low availability of water and nutrients. On the other hand, it would also be possible to expect that root length was reduced because roots need less length to locate nutrients in a fertile soil (Yavitt et al., 2011). This was not the case in this study, as explained before; since the fertilization was applied in later ages and the roots had already developed in the low nutrient environment, which did not reflect any differences.

Root volume

It was estimated that the values for root volume in the litter, varied in average from 25.1 to 31.8 m³ ha⁻¹ (Figure 9). There was not significant difference among treatments, since the volume of roots is related to the diameter and the total length in the litter.

Figura 9. Total length of roots contained in the litter per hectare from a stand of *Pinus taeda* with 23 years old after 5 years of experiment with fertilization and liming in different treatments – Jaguariaíva – Paraná State.

If we consider that practically all root dry matter was contained in the humus litter, and it had in average 8 cm of depth in the stand; we would have an equivalent volume of humus litter of 800 m³ ha⁻¹. Based on this, it is possible to calculate that the roots present in the occupied area, accounted in average for 3 to 4% of the volume occupied by the FHL.

The data of this experiment confirms the idea that litter may play an important role for nutrient cycling, water and nutrient absorption in stands of *Pinus taeda* L. established in poor sites.

CONCLUSION

The expressive quantity of litter in the studied area confirms the accumulation of litter in sites with low fertility soils. A high content of dry matter and length of roots in the litter was also observed. This may indicate that the litter under low soil fertility stimulates the growth of roots in the litter in order to absorb the nutrients present in the residues.

For the period evaluated no effect from fertilization and liming in the stand was verified, indicating that this management procedure is not efficient in later age stands of *Pinus*.

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