Cover crop increases soybean yield cropped after degraded pasture in sandy soil[1]

Plantas de cobertura aumentam a produtividade da soja cultivada após pastagem degradada em solo arenoso

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HIGHLIGHTS:
Black oats improve soil microbial activity, nodulation and soybean yield.
Shoot dry weight, shoot nitrogen, root nodules number and dry weight of root nodules, have a positive correlation with soybean yield.
Nitrogen fertilization is not recommended where cover crops are used.

ABSTRACT: Soybean cropping has been growing in recent years in environments with sandy soils and with climatic risk, but yield is low, especially in the early years. The objective of this study was to evaluate the effect of cover crops and nitrogen management in a sandy soil previously under degraded pastures on soybean yield. The study was conducted in Western São Paulo state, Brazil. The experimental design was randomized blocks, with four replicates, and the treatments were: black oats; black oats + 50 kg ha^{-1} of N in black oats; black oats + 50 kg ha^{-1} of N in soybean; black oats + lupine; black oats + lupine + 50 kg ha^{-1} of N in soybean; lupine; fallow; fallow + 50 kg ha^{-1} of N in soybean. Nitrogen concentration of the microbial biomass was higher with oats + N in soybean applied at the beginning of flowering (R1). The number of nodules in soybean roots increased by 2.3 times with oats and oats + N in soybean as compared to fallow. Soybean yield was higher in treatments with oats + N in oats (2,130 kg ha^{-1}), oats (2,038 kg ha^{-1}) and oats + N in soybean (1,872 kg ha^{-1}). In the absence of cover crops, N fertilization in soybean increased yield by 19% (262 kg ha^{-1}) compared to fallow. Black oats are the best option to increase soybean yield. However, in the absence of cover crops, nitrogen fertilization in soybean is necessary.

Key words: *Glycine max*, climatic risk environment, nodulation, microbial biomass

RESUMO: O cultivo de soja vem crescendo nos últimos anos em ambientes de solos arenosos e com risco climático, mas a produtividade é baixa, principalmente nos primeiros anos. Objetivou-se neste estudo avaliar o efeito do manejo de culturas de cobertura e nitrogênio em solo arenoso previamente ocupado por pastagem degradada na produtividade da soja. O estudo foi conduzido no Oeste de São Paulo. O delineamento experimental foi em blocos casualizados, com quatro repetições e os tratamentos foram: aveia preta; aveia preta + 50 kg ha^{-1} de N na aveia preta; aveia preta + 50 kg ha^{-1} de N na soja; aveia preta + tremoço; aveia preta + tremoço + 50 kg ha^{-1} de N na soja; tremoço; pousio; pousio + 50 kg ha^{-1} de N na soja. O teor de nitrogênio da biomassa microbiana foi maior com aveia preta + N na soja aplicado no início do florescimento (R1). O número de nódulos nas raízes da soja aumentou 2,3 vezes com aveia e aveia + N na soja em relação ao pousio. A produtividade da soja foi maior nos tratamentos com aveia + N na aveia (2.130 kg ha^{-1}), aveia (2.038 kg ha^{-1}) e aveia + N na soja (1.872 kg ha^{-1}). Na ausência de culturas de cobertura, a adubação nitrogenada na soja aumentou a produtividade em 19% (262 kg ha^{-1}) comparado ao pousio. A aveia preta é a melhor opção para aumentar a produtividade da soja. Porém na ausência de planta de cobertura é necessária a adubação nitrogenada na soja.

Palavras-chave: *Glycine max*, ambiente de risco climático, nodulação, biomassa microbiana

INTRODUCTION

Due to restrictions on opening of new areas, soybean cultivation has expanded to areas with lower yield potential (Cordeiro & Echer, 2019; Silva et al., 2020). These areas include pastures that are degraded or in some stage of degradation and sandy soils, which are about 8% of Brazilian soils (Donagemma et al., 2016). No-tillage, previous cover crops and the adequate management of nitrogen can increase soybean yield in these environments, due to improved soil conditions (Pereira et al., 2007; Nivelle et al., 2016; Williams et al., 2018; Silva et al., 2020).

Legume species can fix nitrogen through biological nitrogen fixation (BNF) and, after mineralization of straw and roots, N will be available to plants (Gabriel et al., 2016). Therefore, under conditions where BNF efficiency is limited, legumes can partially provide the N required by soybean. Restovich et al. (2012) reported no increase in soybean yield with preceding crops of grasses and/or legumes under temperate climate with high soil fertility and 2% of soil organic matter (SOM). Garcia et al. (2014) also reported no increase in soybean yield using grasses before soybean and N application in cover crops in a fertile clay soil with 2% of SOM.

However, the studies were conducted in favorable environments for BNF activity in soybean (long-term crop rotation associated with cover crops, medium- and high-fertility soils and regular rainfall pattern). Thus, in unfavorable edaphoclimatic environments for BNF, preceding cover crops (Williams et al., 2018) and mineral nitrogen fertilization can improve soybean yield (Mourtzinis et al., 2018; Cordeiro & Echer, 2019). The objective of this study was to evaluate the effect of preceding crops and nitrogen supply on soybean (*Glycine max*) yield.

MATERIAL AND METHODS

A field experiment was carried out during the 2017/2018 season, in Presidente Bernardes, São Paulo, Brazil (22º 11' 53" S, 51º 40' 30" W and 401 m altitude). The climate of this region is classified as Aw or tropical rainy (Köppen) (Alvares et al., 2013). The rainfall and maximum and minimum temperatures recorded during the experiment are shown in Figure 1. Soil of the area is classified as Oxisol, of sandy texture (Soil Survey Staff, 2014). The physical-chemical attributes are shown in Table 1. The analysis were carried out according to the methodologies described by Raij et al. (2001).

The experimental design was randomized blocks with four replicates. Treatments consisted of the combination between cover crops and nitrogen fertilization: 1) black oats; 2) black oats + 50 kg ha^{-1} of N in black oats; 3) black oats + 50 kg ha^{-1} of N in soybean; 4) black oats + lupine; 5) black oats + lupine + 50 kg ha^{-1} of N in soybean; 6) lupine; 7) fallow; 8) fallow + 50 kg ha^{-1} of N in soybean. Each experimental unit had dimensions of 7 x 3.15 m, totaling 22.05 m².

The experiment was set up without soil plowing in an area of degraded pasture (*Urochloa brizantha*). The area has been occupied by pasture in the last 10 years without application of lime and fertilizers and was desiccated in April 2017 (glyphosate 1440 g a.i. ha^{-1}). Liming without incorporation into the soil was performed in the area (1,800 kg ha^{-1} of dolomitic lime), eight months (April, 2017) before soybean sowing.

Table 1. Soil physical-chemical attributes before sowing of cover crops and soybean

Layer (m)	pH	SOM (g dm^{-3})	P$_{resin}$ (mg dm^{-3})	H + Al	K	Ca	Mg	CEC	BS (%)	Sand	Clay	Silt
				(mmol$_c$ dm^{-3})						(g kg^{-1})		
0-0.10	3.9	9.7	5.5	22.9	1.8	2.3	2.2	29.3	21.6	805	135	60
0.10-0.20	3.9	8.9	5.5	26.9	1.4	1.9	1.5	31.7	15.2	790	141	69

SOM - Soil organic matter; CEC - Cation exchange capacity; BS - Base saturation
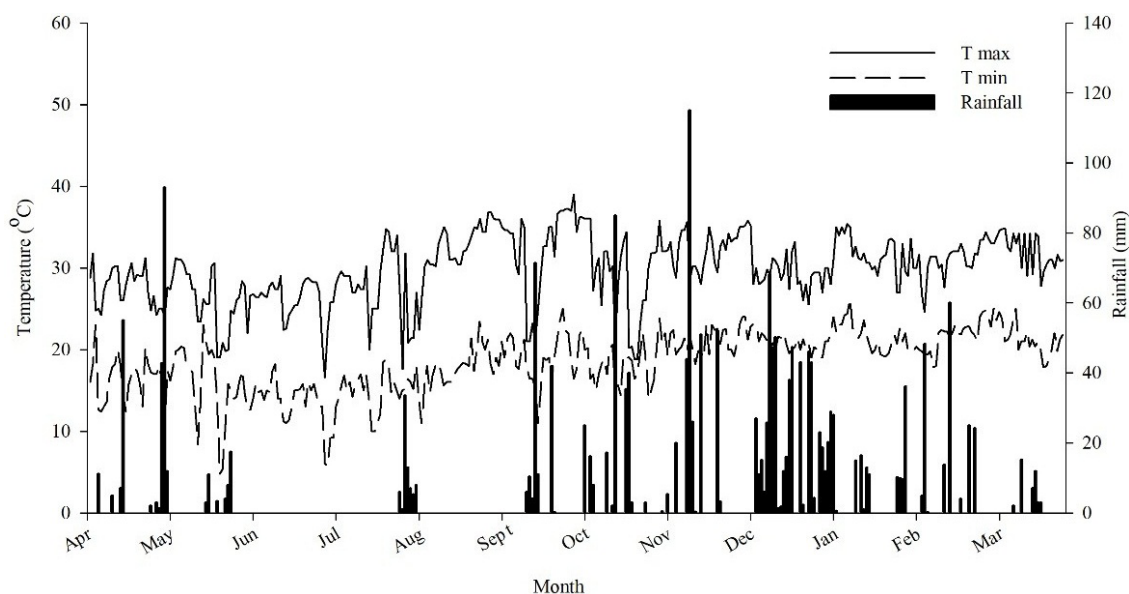
Cover crops (April-November), soybeans (December-March)

Figure 1. Rainfall and maximum (T max) and minimum (T min) temperatures observed during the experiment in Presidente Bernardes, state of São Paulo, Brazil - 2017/2018 season

Liming was performed aiming at 70% base saturation. Cover crops - black oats (*Avena strigosa*) and white lupine (*Lupinus albus*) - were sown (April, 24) at a spacing of 0.22 m between rows, and fertilization was applied in the seeding row at rates of 6, 45 and 15 kg ha^{-1} of N, P$_2$O$_5$ and K$_2$O, respectively. The same amount of seeds (65 kg ha^{-1} for black oats and 60 kg ha^{-1} for lupine) was used in the single cropping and intercropping treatments. Nitrogen fertilization (ammonium nitrate) in oats (treatment 2) was performed at 60 days after emergence (DAE).

In November 2017 (45 days before soybean sowing), cover crops were desiccated (glyphosate at 840 g a.i. ha^{-1} and clethodim - 168 g a.i. ha^{-1}). Shoots of cover crops and weeds in the fallow treatment were sampled immediately after the desiccation of the cover crops using a rectangle (0.4 × 0.5 m), by collecting the straw on the soil surface. Soil samples of 2000 cm^3 (0.1 × 0.1 × 0.2 m) were collected to evaluate root biomass. Samples were washed, dried in an oven at 65 ºC for 48 hours (straw and roots), and then weighed and ground for carbon and nitrogen analysis (Malavolta et al., 1997).

Soybean (TMG 7062 IPRO) was sown on 12/28/2017 with 14 seeds m^{-1}, in rows spaced by 0.45 m. Inoculation was performed in the seed beds, with a liquid inoculant based on *Bradyrhizobium japonicum*, SEMIA 5079 and SEMIA 5080 (6 x 109 CFU mL^{-1} - one dose is equivalent to 100 mL), using eight doses (33% above the recommended dose for areas of first-year soybean cropping), and the solution was applied with a flow rate of 50 L ha^{-1}. Fertilization at sowing consisted of the application of 16.8, 126 and 42 kg ha^{-1} of N, P$_2$O$_5$ and K$_2$O, respectively. At V3 stage the micronutrients cobalt and molybdenum were sprayed at rates of 8 and 40 g ha^{-1}, respectively. Nitrogen fertilization in soybean (ammonium nitrate) was performed in treatments 3, 5 and 8. Potassium fertilization was performed at 60 DAE with 85 kg ha^{-1} K$_2$O (potassium chloride). Weeds, pests and diseases were monitored and controlled as needed following the crop recommendation practices.

Root nodulation (number and weight of nodules) was evaluated at R4 stage in six plants per plot. The root nodules were manually counted, dried in an oven at 65 °C for 48 hours, and weighed on a precision scale (0.01 g) to determine their dry weight. Also, in R4, 10 trifoliate leaves were sampled for N leaf diagnosis and six plants per plot were sampled for shoot dry matter and nitrogen accumulation evaluation (Malavolta et al., 1997). Leaf area index (LAI) was evaluated at the R4 stage (55 DAE) using a ceptometer (AccuPAR LP-80˚ - Decagon Devices). Plant height was evaluated at the R4 stage by measuring three plants per plot.

Soil microbiological indicators such as soil respiration (Rodella & Saboya, 1999), dehydrogenase enzyme (Casida et al., 1964), nitrogen (N-mic) (Joergensen & Brookes, 1990) and carbon (C-mic) (Ferreira et al., 1999) in the microbial biomass, following the extraction methods (Vance et al., 1987), were evaluated in samples from 0-0.10 m depth in three sub-samples per plot at soybean R4 stage.

Yield and yield components (number of pods per plant, number of grains per pod, and 100-grain weight) were determined at R8 stage. Plants in one meter of row were harvested for yield components determination, and mechanical harvest of the three central rows (7 m each) was performed

to evaluate grain yield. Grain moisture was corrected to 13%. Grain N content was determined according to Malavolta et al. (1997).

After testing for homogeneity of variance and normality, data were submitted to ANOVA and means were compared by the Scott-Knott test at p ≤ 0.05 . Pearson's correlation was evaluated between soybean yield and shoot dry matter accumulation, shoot N accumulation, number and weight of root nodules.

RESULTS AND DISCUSSION

Treatments with black oats produced a larger amount of root and total (shoot + root) dry matter than other treatments
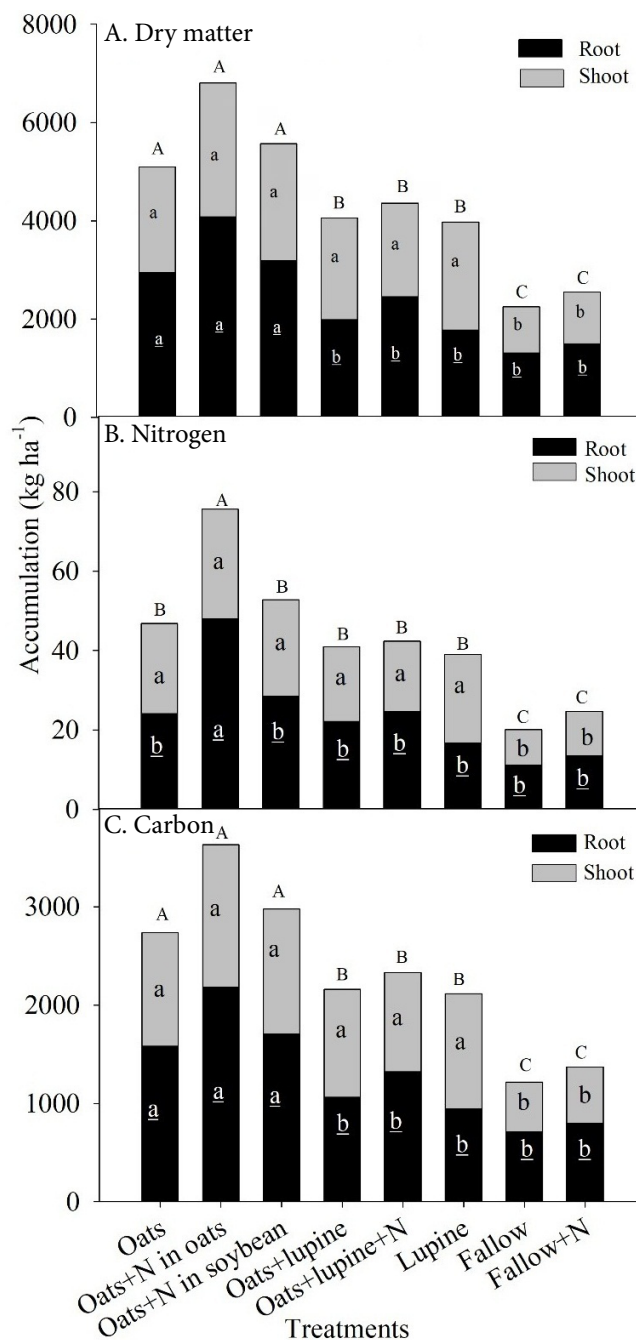
Means followed by the same letter do not differ by the Scott-Knott test at p ≤ 0.05; Different uppercase letters indicate significant differences between treatments adding shoot and root. Different lowercase letters indicate significant difference between separate shoot and root treatments

Figure 2. Dry matter (DM) and accumulation of nitrogen and carbon in shoot and roots (0-0.20 m of depth) of cover crops

(Figure 2). On the other hand, shoot dry matter was similar between treatments with black oats, lupine or intercropping (oats and lupine) (Figure 2). Nitrogen accumulation (shoot and roots) was higher in the black oats + N in black oats treatment with 75.5 kg ha^{-1}, due to a higher concentration of N in the roots (11.8 g kg^{-1}) compared to black oats without N (8.3 g kg^{-1}), since the N in the shoot was similar in treatments with black oats and lupine or in the intercropping. Total carbon accumulation (shoot and root) was higher in black oats treatments, mainly due to root production increase in these treatments (Figure 2). In these cases, N mineral fertilization was needed to increase cover crops biomass production (shoot and roots), as a consequence of the low biological nitrogen fixation (BNF) of lupine.

Preceding black oats + N in soybean, black oats + lupine and black oats + lupine + N in soybean showed the highest soil respiration rates (Table 2). The activity of the enzyme dehydrogenase in the soil was higher after black oat + lupine compared to fallow or fallow + N in soybean (Table 2). The N-mic content was higher in black oats + N in soybean (Table 2). C-mic was higher after oat, oats + N in black oats and oats + N in soybeans (Table 2).

Nitrogen fertilization and cover crops can improve soil microbiological properties (Tahir et al., 2015; Nivelle et al., 2016). This is due to root growth, release of exudates and increased soil organic carbon. In this study, the application of N in soybeans (Fallow + N) only increased the N-mic, and associated with cover crops improved soil respiration (black oats + N in soybean), enzyme dehydrogenase and C-mic (Table 2). However, lupine in single crop or intercropped with oats had little effect on soil microbiology, contrasting to the findings from Nivelle et al. (2016), who reported an increase in soil respiration rates and N-mic by leguminous cover crops. In this case, the lupine did little to improve the soil, due to the low vegetative growth.

Under temperate climate, monoculture and high doses of mineral N (up to 150 kg ha^{-1}) can reduce soil microbiota, due to soil acidification (Zhaou et al., 2017). Ramirez et al. (2010) claim that the soil biological activity decreases as N doses are increased; however, the addition of carbon in the soil (straw) can reverse this effect. Thus, the balance in the inputs of N and C in the system is important, which must be adjusted using cover crops with medium C:N ratio (< 30) or increase the system´s nitrogen mineral fertilization.

The number of soybean root nodules was lower when cultivated after fallow (Figure 3B). On the other hand, the pre-crop of black oats or black oats + N in soybean caused soybean nodulation. Nitrogen-fertilized black oats

Table 2. Soil respiration, dehydrogenase activity, nitrogen and carbon of the microbial (mic) biomass of a soil cultivated with cover crops

Treatments	Soil respiration (mg CO_2 kg^{-1} soil day^{-1})	Dehydrogenase (mg of TTF g^{-1} soil)	N-mic	C-mic
			(mg kg^{-1} of soil)	
Oats	2.7 b	5.6 a	9.6 c	110.8 a
Oats + N	1.3 c	6.2 a	11.7 b	101.6 a
Oats + N in soybean	4.4 a	5.0 b	14.0 a	108.1 a
Oats + lupine	3.9 a	6.7 a	8.6 c	48.2 c
Oats + lupine + N	4.4 a	5.8 a	11.1 b	90.6 b
Lupine	2.4 b	6.1 a	8.7 c	46.1 c
Fallow	1.7 c	3.7 b	6.6 d	46.7 c
Fallow + N in soybean	1.6 c	4.3 b	11.6 b	53.6 c
CV (%)	14.13	17.92	9.39	9.75

Means followed by the same letter do not differ significantly by the Scott-Knott test at p > 0.05
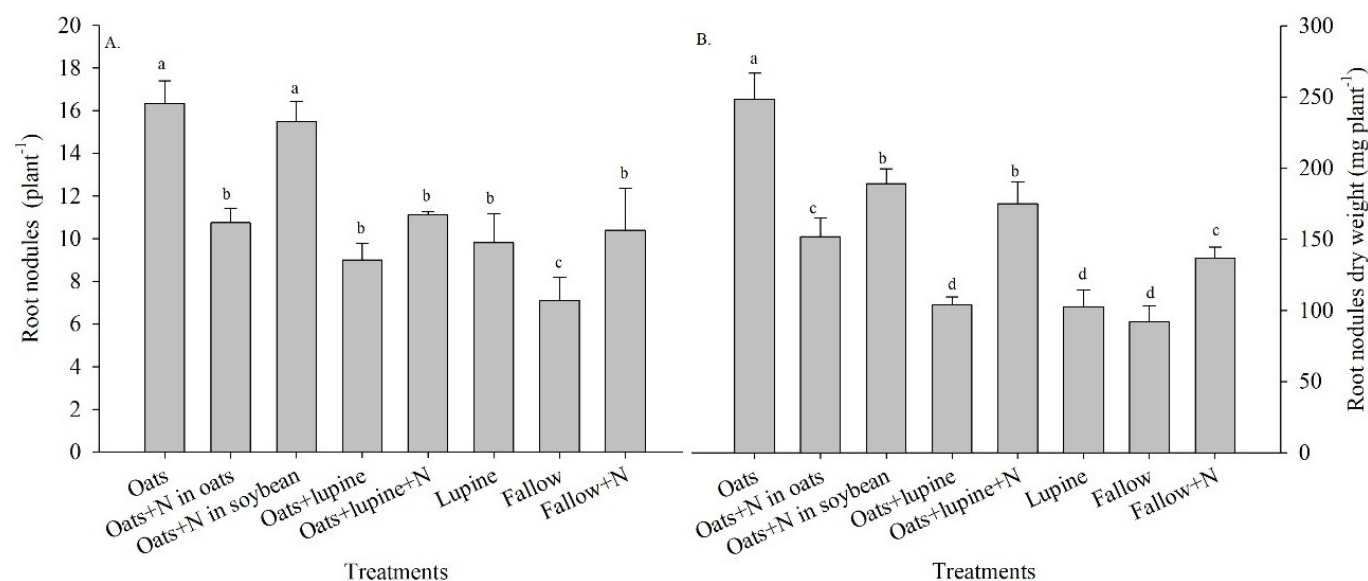
Means followed by the same letter do not differ significantly by the Scott-Knott test at p > 0.05

Figure 3. Number and dry weight of root nodules of soybean grown after cover crops and nitrogen management

decreased nodule number and dry weight (Figures 3A and B) compared to unfertilized black oats. This is because at the beginning of nodulation (early stages of soybean) the availability of N in the soil was higher, limiting biological fixation. Pre-crop of lupine, in single crop or intercropped with oats, did not significantly affect the dry weight of root nodules (Figure 3B).

Even though soybean nodulation increased after black oats and black oats + N in soybean, the nodulation was still considered low. Mendes et al. (2008) reported a dry weight of root nodules above 320 mg plant^{-1} to achieve soybean yields above 2.8 Mg ha^{-1}. Hungria et al. (2017) reported yields above 4.0 Mg ha^{-1} with 25 root nodules plant^{-1} in a first-year soybean area in medium- and high-fertility soils, different from what we found in a low-fertility sandy soil, whose maximum nodulation was 16 root nodules plant^{-1} and root nodules dry weight was 248 mg plant^{-1}.

Nitrogen fertilization can reduce the efficiency of BNF, because it increases the availability of inorganic N available for crops and requires less energy for plant uptake in comparison to biological fixation (Kaschuk et al., 2016; Saturno et al., 2017). In this study, N-fertilized black oats reduced soybean nodulation (Figures 3A and B). In addition, N-fertilized soybean after fallow increased nodule number by 46%, compared to fallow, probably because a small supply of N was beneficial and did not harm BNF, as previously reported (Cordeiro & Echer, 2019). Xia et al. (2017) report higher nodulation of soybeans with the addition of low levels (< 50 mg L^{-1}) of nitrogen due to increased nitrogenase activity in soybean roots; however, high levels of mineral N (> 50 mg L^{-1}) reduce nitrogenase activity and nodulation of soybeans. Additionally, in degraded post-pasture areas with low N stock in the soil, the application of up to 50 kg ha^{-1} of N does not reduce soybean nodulation (Cordeiro & Echer, 2019), as reported in this study (Figure 3A).

Higher BNF efficiency in black oats and black oats + N in soybeans resulted in higher LAIs (leaf area index), but still below the recommended for high yields in soybeans (3.5 to 4.5) (Specht et al., 1999; Tagliapietra et al., 2018) (Table 3). Single black oats also showed the highest plant height in comparison to the other treatments, being similar to black oats + N in black oats and black oats + N in soybean for shoot dry weight, but was inferior to black oats + N in soybean for shoot N accumulation (Table 3). Leaf nitrogen content was within the sufficiency range (45 to 55 g kg^{-1}) in all treatments

(Malavolta et al., 1997). This is because the vegetative growth of soybeans was small, due to late sowing and low soil fertility. Thus, even the fallow levels were within the sufficiency range, but did not result in high soybean yields.

Shoot dry weight, shoot nitrogen, root nodules number and root nodules dry weight showed positive correlation with soybean grain yield (Figures 4A and D). In addition, root nodules number and dry weight had the highest coefficients of determination for soybean yield (Figures 4C and D).

Black oats and black oats + N in black oats (50 kg ha^{-1}) increased yields by 50 and 57%, respectively, compared to fallow (Table 4). Without cover crops (fallow), N fertilization (fallow + N in soybean) increased yield by 19% (262 kg ha^{-1}) (Table 4). The increase in yield in black oat + N in black oats is mainly due to the higher number of pods per plant (Table 4). Besides, the highest number of grains per pod and 100-grain weight (Table 4) in oat sustained the yield in this treatment. Black oat + N in soybean, lupine and fallow led to the lower N contents in soybean grains (Table 4). The export of N in the grains was higher in black oats and black oats + N in soybean treatments, reflecting the higher yield in these treatments (Table 4).

Therefore, preceding black oats, black oats fertilized with N and oats with nitrogen in soybean were the treatments that caused the highest soybean yields in a first-year cultivation area (Table 4), as a result of the improvements in the microbial activity of the soil, mainly higher carbon content of the biomass (Table 2), soybean nodulation (Figure 3) and higher plant growth, principally shoot dry matter accumulation. Besides that, cover crops before soybean increases water use efficiency, nitrogen and crop yield (Restovich et al., 2012); improves soil physical, chemical (Calonego & Rosolem, 2013; Calonego et al., 2017) and biological attributes (Nivelle et al., 2016). Additionally, cover crops improve soil environment for agricultural production, even in areas of sandy soils and climatic risk.

In this study we report the importance of adequate soil management before planting of soybeans over post-degraded pasture, being evident that in these environments, in the absence of cover crops, soybeans depend on mineral N, due to low nodulation of soybeans (Figure 3). Black oats as cover crops before soybean cultivation were the best option to improve soybean yield in areas of transition from degraded pastures to agricultural production, since the absence of cover crops demands N inputs (Table 4) (Cordeiro & Echer, 2019), which increases the production cost.

Table 3. Leaf area index (LAI), plant height, shoot dry weight, shoot N accumulation and leaf nitrogen concentration in soybean

Treatments	Leaf area index	Plant height (m)	Shoot dry weight (kg ha^{-1})	Shoot N (kg ha^{-1})	Leaf N concentration (R4) (g kg^{-1})
Oats	2.37 a	0.44 a	2060 a	63.7 b	48.7 b
Oats + N	2.22 a	0.40 b	1989 a	55.4 b	46.0 b
Oats + N in soybean	2.03 b	0.41 b	2198 a	84.9 a	53.0 a
Oats + lupine	2.18 a	0.42 b	1740 b	58.3 b	46.4 b
Oats + lupine + N	1.90 b	0.40 b	1944 a	57.3 b	46.6 b
Lupine	1.86 b	0.40 b	1659 b	49.6 c	48.9 b
Fallow	1.90 b	0.41 b	1593 b	57.3 c	48.6 b
Fallow + N in soybean	1.76 b	0.42 b	1724 b	52.6 c	47.4 b
CV (%)	6.5	2.1	7.6	10.72	4.6

Means followed by the same letter do not differ significantly by the Scott-Knott test at p > 0.05
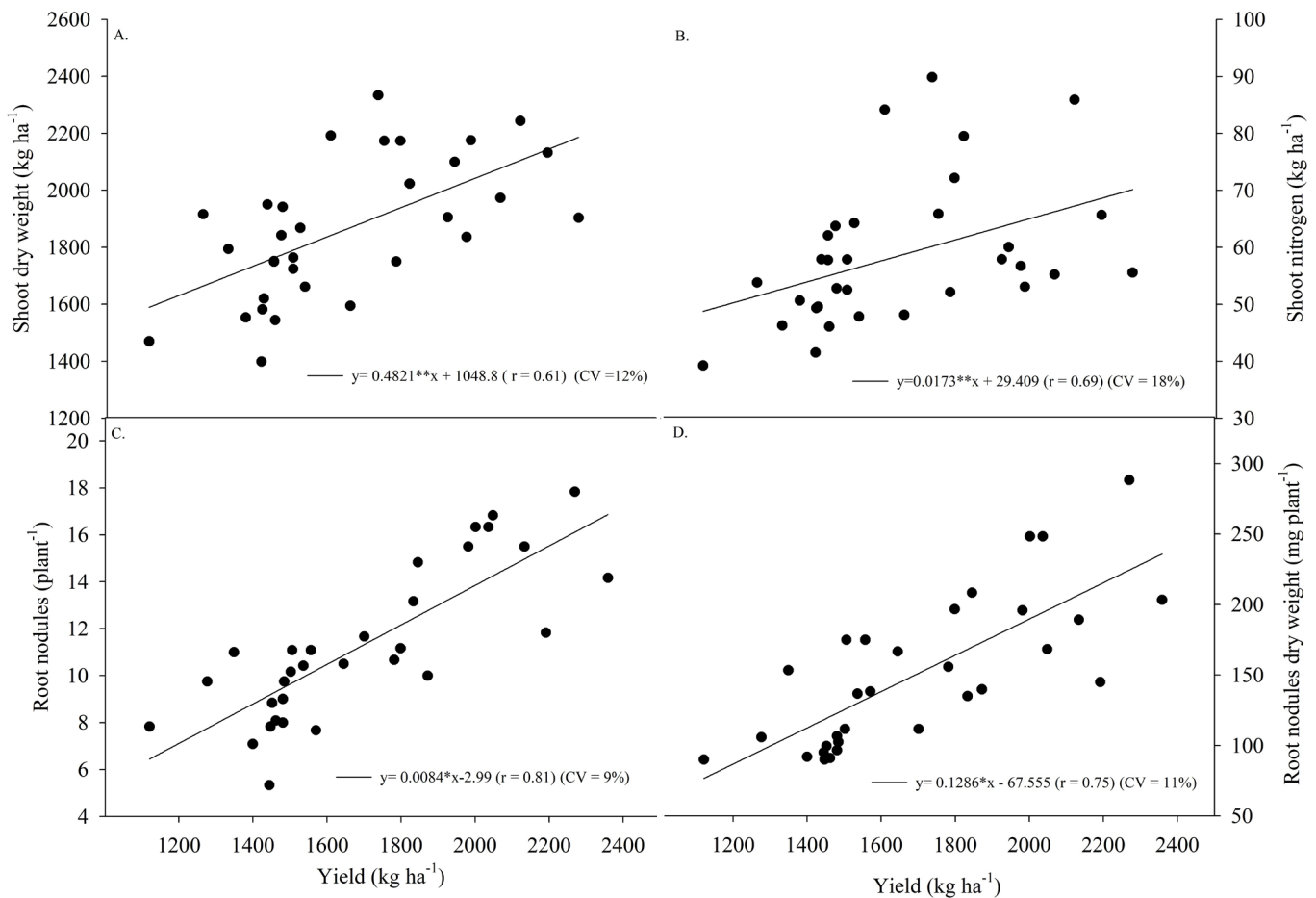
*·** - Significant at p ≤ 0.05 and at p ≤ 0.01 by t test, respectively; r - Cofficient of correlation

Figure 4. Relationship between grain yield and shoot dry weight, shoot nitrogen, root nodules and root nodules dry weight

Table 4. Soybean yield, yield components and export of N in different treatments

Treatments	Pods (plant⁻¹)	Grains (pod⁻¹)	100-grain weight (g)	Grain N concentration (g kg⁻¹)	Yield (kg ha⁻¹)	Export of N (kg ha⁻¹)
Oats	19.6 b	2.9 a	15.1 a	61.7 a	2038 a	125.5 a
Oats + N	30.0 a	1.8 c	15.9 a	61.9 a	2130 a	131.9 a
Oats + N in soybean	22.7 b	2.8 a	12.1 c	58.5 b	1872 a	109.4 b
Oats + lupine	21.9 b	1.9 c	14.1 b	64.6 a	1478 b	95.5 c
Oats + lupine + N	22.5 b	2.1 c	14.0 b	62.7 a	1552 b	97.3 c
Lupine	17.5 d	2.4 b	14.8 a	60.2 b	1477 b	88.7 c
Fallow	19.6 c	1.9 c	14.4 b	56.9 b	1356 b	80.1 c
Fallow + N in soybean	16.2 d	2.7 a	15.7 a	63.1 a	1618 b	97.4 c
CV (%)	8.5	5.5	4.1	4.1	9.8	11.0

Means followed by the same letter do not differ significantly by the Scott-Knott test at p > 0.05

Conclusions

1. In areas of first year of soybean cultivation after degraded pasture in sandy soil there is no need for nitrogen fertilizer application, as long as cover crops are cropped before soybean.

2. Black oats in pre-cultivation of soybeans are an option to reduce the cost of nitrogen fertilizer, by improving soil microbiology, efficiency of biological nitrogen fixation and soybean yield, while using only nitrogen fertilizer brings few benefits to the system.

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