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Coffee seedling growthafter legume cultivation in soils with contrasting phosphorus contents[1]

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ABSTRACT

Coffee seedling growth depends on soil phosphorus (P) availability and may be influenced by from pre-cultivation with legumes. Efficient and sustainable ways to increase the bioavailability of P through the recovery of P adsorbed by the soil matrix should be sought. This study proposed to evaluate the growth and P-use efficiency of coffee seedlings cultivated in soils with different P availability after cultivation with legumes. The experiment was carried out in a fully randomized design. Treatments were arranged in a factorial scheme [(2 x 4) + 1]: two soil types, pre-cultivation with four legume species, and one control (without pre-cultivation). The studied soils comprised a Typical Acriferic Red Oxisol (LVwf) with low-P availability and a Typical Chernossolic Litholytic Entisol (RLm) with high-P availability. The legume species *Crotalaria juncea*, *Cajanus cajan*, *Canavalia ensiformis,* and *Mucuna aterrima* were previously cultivated for 45 days. Afterwards, coffee seedlings were transplanted to the pots, which were then grown for 120 days until evaluations. We assessed the following parameters: plant height (H), stem diameter (SD), shoot dry matter (SDM), root dry matter (RDM), total dry matter (TDM), and shoot contents of macronutrients. Our results showed that coffee seedlings grew more when cultivated in the high-P availability soil, with increments of 13.05% in H, 4.86% in SD, 46.98% in SDM, 17.61% in RDM, and 41.80% in TDM.We also observed an increase of 28.09% in shoot P contents for coffee seedlings grown in RLm. Moreover, pre-cultivation with *C. juncea* provided the largest increases in coffee seedling growth compared to the control. When grown after *C. ensiformis* cultivation, coffee seedlings had the highest shoot contents of P, Ca, Mg, and S, which, compared to control, increased by 45%, 39%, 18%, and 17%, respectively.

Keywords: *Canavalia ensiformis*; *Crotalaria juncea*; *Coffea arabica*; phosphate fertilization; nutritional efficiency.

1 INTRODUCTION

As coffee has economic and social significance in Brazil, further research intended to increase phosphorus (P) absorption efficiency and its influence on coffee growth is needed since it is related to productivity. Coffee seedlings require macro-and micronutrients for their full development (Vilela Junior et al., 2017). Among the macronutrients, P is one of the most significant since it increases root development and vigor (Santinato et al., 2014), which are indispensable to ensure plant growth, development, and productivity.

Highly weathered tropical soils have low-P availability to plants due to a high interaction between phosphates and soil solid-phase components, adsorbing or precipitating P, thus making it unavailable to plants. The retention of P added to the soil occurs both by its precipitation in solution with ionic forms of iron (Fe), aluminum (Al), and calcium (Ca), and by its adsorption by Fe and Al oxyhydroxides (Novais; Smith, 1999). In soils with low-P availability, applications of high P contents are customary to raise contents close to the critical level (Raij, 2011).

Some studies have evidenced the relationship between P supply via phosphate fertilizers and coffee seedling grow thin plant nursery (Tomaz et al., 2009; Lemos et al., 2015; Silva et al., 2019). There has been a growing concern about how long society will be able to rely on the supply of phosphate fertilizers, which are currently limited to phosphate rock deposits (Elser; Bennett, 2011). In this sense, strategies aimed at replacing phosphate fertilizers, and then prolonging the time of use of such mineral reserves, are needed. Hence, P cycling in native soils and its use by pre-cultivation (legumes) and successor (coffee) plants are relevant factors to be investigated and are thus the focus of our study.

Crop rotation with highly efficient species in extracting P from the soil is one of the methods to cycle this element by converting unavailable into more labile forms (Silva et al., 2011). This practice can increase P efficiency in agricultural production systems since succeeding crops may use mineralized P after previous crop shoot and root decomposition (Soltangheisi et al., 2018). Among the crops commonly used in rotation systems as cover and green manure, legumes stand out.

Legumes have been widely used in crop rotation and green manuring due to their higher nitrogen (N) releases. This occurs due to the ability of legumes to associate with nitrogen-fixing bacteria, in addition to their high potential to mobilize and absorb nutrients, producing large amounts of phytomass (Amabile; Carvalho, 2006). Unlike what is seen for N, there are few studies relating P mineralization rates with biochemical traits of legumes (Casali et al., 2011). In this sense, the use of legumes in crop rotation or green manuring can also act as a complementary source of N and increase P use efficiency by plants.

[1]Part of the first author's PhD thesis.

Unavailable P forms can be solubilized and made available to plants through the cultivation of some legumes, which has been considered an attractive strategy for the management of soils with high phosphate adsorption capacity (Mascarenhas; Wutke, 2014). According to Nziguheba and Bünemann (2005), the use of plants as green manure can increase P availability in two circumstances: (a) during their growth when soil properties change and hence P is mobilized; or (b) during their decomposition when organic P is mineralized.

The inclusion of species with high P absorption efficiency in the crop rotation can contribute to the P nutrition of the subsequent crop, thus reducing the need for phosphate fertilizers. Coffee plants can benefit from pre-cultivation with legumes to provide P. Based on the above considerations, the purpose of our study was to evaluate the growth of coffee seedlings and their P-use efficiency when grown in soils with different P availability and after cultivation with legumes.

2 MATERIALS AND METHODS

2.1 Study location

The experiment was conducted in a green house at the campus of the Federal University of Viçosa (UFV), in the city of Viçosa, Minas Gerais State (Brazil). It is located at the geographical coordinates of X = 518969.59 and Y = 4919485.31, and an altitude of 651 m. According to Köppen's classification (1948), the local climate is warm-temperate, with rainy summers and cold, dry winters (Cwb). From 1968 to 2010, average annual rainfallwas1, 268.2 mm, relative humidity around 81%, and average annual temperature 20 °C, according to data obtained from a local weather station (Lorenzon; Dias; Leite, 2013).

2.2 Soil characterization

Soil samples (horizon A) were collected at the city of Lagoa Formosa (Minas Gerais, Brazil) and corresponded to two soil classes: Typical Acriferic Red Oxisol (in Portuguese: *Latossolo Vermelho Acriférrico típico - LVwf*) and Typical Chernossolic Litholytic Entisol (in Portuguese: *Neossolo Litólico Chernossólico típico – RLm*). The sampled sites had no agricultural use. To collect the soil, mini-trenches were opened along low traffic roads, and the soils was collected at a depth of 0 to 0,3 m. The geographic coordinates of the sampling sites are X: 353390.67; Y: 7926050.50 (soil LVwf); and X: 362621.72; Y: 7919872.49 (soil RLm). The soil samples were characterized physically and chemically, without using repetitions, following the methodologies described by Teixeira et al. (2017), the results of which are shown in Table 1.

Table 1: Physicochemical properties of the soils used in the study.

Property	Soil type	
	LVwf	RLm
Coarse sand (kg kg^{-1})	0.085	0.073
Fine sand (kg kg^{-1})	0.079	0.082
Silt (kg kg^{-1})	0.232	0.360
Clay (kg kg^{-1})	0.603	0.486
Sd (kg m^{-3})	1170.00	1220.00
Pd (kg m^{-3})	3080.00	2890.00
pH H_2O (1:2.5)	5.72	6.01
Ca^{2+} (cmol$_c$ kg^{-1})	0.46	15.08
Mg^{2+} (cmol$_c$ kg^{-1})	0.18	4.32
K^{+} (mg dm^{-3})	29.00	163.00
Available P Mehlich-1 (kg m^{-3})	0.0017	0.768
Total P (mg dm^{-3})	2969.25	7419.85
Remaining P (kg)	0.0062	0.0133
Al^{3+} (cmol$_c$ kg^{-1})	0.00	0.00
H+Al (cmol$_c$ kg^{-1})	5.90	8.30
TB (cmol$_c$ kg^{-1})	0.71	19.82
CEC (cmol$_c$ kg^{-1})	0.71	19.82
$_e$CEC (cmol$_c$ kg^{-1})	6.61	28.12
%BS	10.70	70.50
OM (dag kg^{-1})	2.74	3.91

LVwf: Typical Acriferic Red Oxisol; RLm: Typical Chernossolic Litholytic Entisol; Sd: soil density; Pd: particle density; TB: total bases; %BS: base saturation; CEC: cation exchange capacity at pH 7; $_e$CEC: effective cation exchange capacity; OM: organic matter.

2.3 Experimental design

The experiment was carried out in a fully randomized design (FRD) with six replicates. Treatments were arranged in a factorial scheme [(2 x 4) + 1] and defined as: two soil types, pre-cultivation with four legume species, and a control (without pre-cultivation) totaling 60 experimental plots.

2.4 Coffee seedling cultivation

The experimental plots consisted of rigid polyethylene pots, with a capacity of 3 dm^3 soil. First, the legume species *Crotalaria juncea*, *Cajanus cajan*, *Canavalia ensiformis*, and *Mucuna aterrima* were cultivated. The soils were fertilized (except for P) according to the recommendations of Novais et al. (1991). For both soils, the doses of nutrients consisted of 0.1, 0.04, 0.00081, 0.00366, 0.00015, 0.004, 0.00133, and 0.00156 kg m^{-3} soil of N, S, B, Mn, Mo, Zn, Cu, and Fe, respectively, and the following sources were used: NH_4NO_3, K_2SO_4, H_3BO_3, $MnCl_2.4H_2O$, $NaMoO_4.2H_2O$, $ZnCl_2$, Cu Cl, and $FeCl_3.6H_2O$, respectively. Besides, in the LVwf soil, 0.3kg

m^{-3} of K was applied, using KCl as the source, as well as 0.00204 kg of calcium ($CaCO_3$ p.a.) and magnesium ($MgCO_3$ p.a.) carbonates in 4:1 proportion, for Ca and Mg supply (Ribeiro; Guimarães; Alvarez, 1999).

After 45 days of cultivation, the legume Crotalaria juncea was starting to flower, thus the shoots and roots of all legumes were removed. The roots were separated from the aerial part at the base of the stem, sieved and washed under running water to remove soil particles. Then, soil samples were taken from each experimental unit for chemical characterization, without repetition (Table 2), following the procedures described by Teixeira et al. (2017).

Coffee seedlings (*Coffea arabica*) from the cultivar MGS Paraíso 2 were grown in plastic bags for 60 days, then the uniform seedlings were transplanted into the experimental pots. Each seedling was transplanted to a separate pot by cutting out the bottom of the bag. Afterwards, the soils were fertilized with nutrient solution, except for P, as indicated by Novais et al. (1991). For both soils, the doses of nutrients consisted of0.1, 0.04, 0.00081, 0.00366, 0.00015, 0.004, 0.00133, and 0.00156 kg m^{-3} soil of N, S, B, Mn, Mo, Zn, Cu, and Fe, respectively, and the following sources were used: NH_4NO_3, K_2SO_4, H_3BO_3, $MnCl_2.4H_2O$, $NaMoO_4.2H_2O$, $ZnCl_2$, CuCl, and $FeCl_3.6H_2O$, respectively. Besides, in the LVwf soil, 0.3kg m^{-3} of K was applied, using KCl as the source, as well as 0.00204 kg of calcium ($CaCO_3$ p.a.) and magnesium ($MgCO_3$ p.a.) carbonates in 4:1 proportion, for Ca and Mg supply (Ribeiro; Guimarães; Alvarez, 1999).

2.5 Coffee agronomic and nutritional evaluations

After 120-day coffee cultivation, seedling growth and nutritional status were estimated. The following growth parameters were determined: plant height (PH), stem diameter (SD) at 5 cm height from ground level, shoot dry matter (SDM), root dry matter (RDM), total dry matter (TDM), and Dickson's quality index (DQI). The PH was measured using a ruler graduated in centimeters (cm), while SD was measured with a digital caliper with a precision of 0.01 mm. Both SDM and TDM were quantified after collecting plants and separating roots from shoots by cutting them out at the base of stems. After cutting the shoots, the roots were collected, washed in running water over a 0.0005 m sieve. Both shoot and root were packed separately in paper bags, dried in an oven at 65 °C for 72 h, and weighed on an analytical scale.

The DQI was calculated using the formula proposed by Dickson, Leaf and Hosner (1960), as follows:

$$DQI = \frac{TDM}{(PH / SD) + (SDM / RDM)} \quad (1)$$

Macronutrient contents (N, P, K, Ca, Mg, and S) were determined by processing shoots and roots in a Willey type mill. Besides, nitro-perchloric digestion (Malavolta; Vitti; Oliveira, 1997) was performed to quantify levels of P, K, Ca, Mg, and S, which were determined by ICP-OES (inductively coupled plasma optical emission spectrometry). The content of N was quantified by sulfuric digestion and Kjeldahl distillation (Tedesco; Gianello, 1979).

Nutrient-use efficiency indexes were calculated using dry matter weight and P content of each plant part (Amaral et al., 2011), as follows:

$$NUE = \frac{(TDM)^2}{\text{P content in the entire plant}}, \text{in } kg^2 kg^{-1} \quad (2)$$

Nutrient-use efficiency for shoot production (NUESP):

Table 2: Chemical characterization of the soils after 45-day cultivation with legumes.

Legume species	pH	Ca^{2+}	Mg^{2+}	K-available *	P-available *	Al^{3+}	H+Al
	H_2O	$cmol_c\ kg^{-1}$		--------- kg m^{-3} ---------		$cmol_c\ kg^{-1}$	
LVwf							
Crotalaria juncea	5.63	0.83	0.53	0.0186	0.0003	0.00	3.54
Cajanus cajan	5.16	0.70	0.40	0.0216	0.0003	0.00	3.69
Canavalia ensiformis	5.75	0.76	0.53	0.0097	0.0015	0.00	3.98
Mucuna aterrima	5.88	0.95	0.63	0.0255	0.0002	0.00	3.98
RLm							
Crotalaria juncea	6.32	13.04	4.42	0.0898	0.8327	0.00	6.49
Cajanus cajan	6.11	14.14	5.30	0.1043	0.8521	0.00	6.63
Canavalia ensiformis	6.17	14.02	4.54	0.0898	0.7615	0.00	6.78
Mucuna aterrima	6.32	14.76	4.51	0.0898	0.8424	0.00	6.63

* Determined by the $Mehlich^{-1}$ extractor.

LVwf: Typical Acriferic Red Oxisol (in Portuguese: *Latossolo Vermelho Acriférrico típico*); RLm: Typical Chernossolic Litholytic Entisol (in Portuguese: *Neossolo Litólico Chernossólico típico*).

$$NUESP = \frac{(SDM)^2}{\text{P content in the entire plant}}, \text{in } kg^2kg^{-1} \quad (3)$$

Nutrient-use efficiency for root production (NUERP):

$$NUERP = \frac{(RDM)^2}{\text{P content in the entire plant}}, \text{in } kg^2kg^{-1} \quad (4)$$

Using the P contents, P translocation index (PTI) was calculated (Abichequer; Bohnen, 1998), as follows:

$$PTI = \frac{\text{P content in the plant shoot}}{\text{P content in the entire plant}} \times 100 \quad (5)$$

2.6 Statistical analysis

The data were submitted to analysis of variance (ANOVA) by the F-test ($p<0.05$) and, if significant, the averages were compared by the Tukey's test ($p<0.05$). All statistical procedures were conducted with the aid of the SISVAR software (Ferreira, 2014).

3 RESULTS

3.1 Coffee plant growth

After 120-day cultivation, there was no significant interaction ($p>0.05$) between soil types and legume species for coffee seedling growth parameters (Table 3). When cultivated in RLm, coffee seedlings showed higher values of PH, SD, SDM, TDM, and DQI, with increments of 13.05% in PH, 4.86% in SD, 46.98% in SDM, 17, and 41.80% in TDM compared to seedlings grown in LVwf.

3.2 Coffee shoot and root contents of macronutrients

There was an interaction ($p<0.05$) between soil types and legume species only for coffee shoot K contents (Table 4). However, no interaction was observed for the other macronutrients (N, P, Ca, Mg, and S). By analyzing the effect of each factor individually, we observed that coffee seedlings grown in RLm had superior shoot contents of N, P, Ca, Mg, and S compared to those grown in LVwf.

The interaction between legume species and soil types was broken downinto coffee seedling shoot K contents and root P, Mg, and K contents separately (Table 5). In terms of pre-cultivation with legumes, except for root K content, there was no difference among treatments when coffee seedlings were cultivated in LVwf. But when grown in RLm, the seedlings in treatments without pre-cultivated plants and after *Crotalaria juncea* cultivation showed the lowest values of shoot K content and root P, Mg, and K contents, and after *Mucuna aterrima* cultivation for root P, Mg, and K contents.

3.3 Nutritional efficiency of phosphorus in coffee seedlings

There was no significant interaction between legume species and soil types for nutrient use efficiency (NUE), nutrient use efficiency for shoot production (NUESP), and nutrient use

Table 3: Coffee seedling growth parameters during 120-day cultivation, after cultivation with legumes in soils with contrasting P contents.

Treatment	PH	SD	SDM	RDM	TDM	DQI
	------- m -------		---------------- kg ----------------			
Legume species (L)						
Crotalaria juncea	0.3811a	0.00607a	0.0112a	0.0023a	0.0135a	1.21a
Cajanus cajan	0.3850a	0.00554ab	0.0098ab	0.0019a	0.0116ab	0.94ab
Canavalia ensiformis	0.3753a	0.00598a	0.0095ab	0.0019a	0.0114ab	1.01ab
Mucuna aterrima	0.3862a	0.00580a	0.0101ab	0.0019a	0.0119ab	0.93ab
Without pre-cultivated plant	0.3863a	0.00508b	0.0085b	0.0016a	0.0101b	0.79b
Soil type(S)						
LVwf	0.3593b	0.0056b	0.0079b	0.0018a	0.0097b	0.88b
RLm	0.4062a	0.0058a	0.0117a	0.0021a	0.0137a	1.07a
L*S	ns	Ns	ns	ns	ns	ns
C.V. (%)	5.91	8.77	17.36	39.68	19.10	32.83
Overall average	0.3828	0.0057	0.0098	0.0019	0.0117	0.0009

PH: plant height, SD: stem diameter, SDM: shoot dry matter, RDM: root dry matter, TDM: total dry matter, DQI: Dickson's quality index. LVwf: Typical Acriferic Red Oxisol; RLm: Typical Chernossolic Litholytic Entisol; L*S: legume species X soil type interaction; C.V.: coefficient of variation; ns: non-significant by the ANOVA F-test; Different letters in the column differ significantly by the Tukey's test at 5% probability.

efficiency for root production (NUERP) for coffee seedlings (Table 6);yet, there was an interaction for P translocation index (PTI). When considering the studied factors, the two soil types (RLm and LVwf) showed no significant difference for NUE, EUPA, and NUERP. Moreover, pre-cultivation with *Canavalia ensiformis* provided the lowest NUESP and shoot production per P unit, with no significant difference to NUERP.

The interaction between the effects of legume species and soil types was broken down for coffee seedling PTI. For seedlings grown in LVwf, all legume species used in pre-cultivation showed no differences. But when grown in RLm, pre-cultivation with *Mucuna aterrima* provided the highest PTI values, while pre-cultivation with *Crotalaria juncea* had the lowest ones. Among the legume species used in pre-cultivation, only *Mucuna aterrima* exhibited different PTI values for coffee seedlings grown in both soils, with seedlings cultivated in RLm showing the highest ones (Table 7).

4 DISCUSSION

4.1 Coffee plant growth

Our findings revealed that pre-cultivation with *Crotalaria juncea* provided increases in SD, SDM, TDM, and DQI for coffee seedlings, with increments of 20%, 33%, 34%, and 52%, respectively, compared to the control (without pre-cultivated plant). DQI is taken as a seedling quality indicator since it considers the biomass balance distribution of seedlings (Fonseca et al., 2002). The higher the DQI, the better the plant biomass

Table 4: Macronutrients concentration in the shoots and roots of coffee seedlings grown for 120 days, after cultivation with legumes, in soils with contrasting P contents.

Treatment	N	P	Ca	Mg	K	S
Shoot content (kg)						
Legume species (L)						
Crotalaria juncea	0.0283a	0.0009ab	0.0065ab	0.0026ab	-	0.0016ab
Cajanus cajan	0.0291a	0.0011ab	0.0078ab	0.0030a	-	0.0019ab
Canavalia ensiformis	0.0296a	0.0012a	0.0081a	0.0032a	-	0.0019a
Mucuna aterrima	0.0283a	0.0011ab	0.0073ab	0.0029ab	-	0.0018ab
Without pre-cultivated plant	0.0303a	0.0008b	0.0059b	0.0022b	-	0.0014b
Soil type (S)						
LVwf	0.0278b	0.0009b	0.0062b	0.0025b	-	0.0015b
RLm	0.0305a	0.0011a	0.0080a	0.0030a	-	0.0018a
L*S	ns	ns	ns	ns	**	ns
C.V. (%)	7.94	27.34	24.60	25.30	44.47	23.20
Overall average	0.0291	0.0010	0.0071	0.0028	0.0076	0.0017
Treatments	N	P	Ca	Mg	K	S
Root content(kg)						
Legume species(L)						
Crotalaria juncea	nd	-	0.0078a	-	-	0.0034a
Cajanus cajan	nd	-	0.0078a	-	-	0.0034a
Canavalia ensiformis	nd	-	0.0076a	-	-	0.0033a
Mucuna aterrima	nd	-	0.0076a	-	-	0.0036a
Without pre-cultivated plant	nd	-	0.0077a	-	-	0.0034a
Soil type(S)						
LVwf	nd	-	0.0058b	-	-	0.0034a
RLm	nd	-	0.0096a	-	-	0.0035a
L*S	nd	**	ns	*	**	ns
C.V. (%)	nd	13.61	11.66	10.88	16.79	8.57
Overall average	nd	0.0012	0.0077	0.0092	0.0112	0.0034

LVwf: Typical Acriferic Red Oxisol; RLm: Typical Chernossolic Litholytic Entisol; L*S: legume species X soil type interaction; C.V.: coefficient of variation; ns: non-significant, nd: non-determined, ** and * significant at 1% and 5% probability, respectively, by the ANOVA F-test; Different letters in the column differ significantly by the Tukey's test at 5% probability.

distribution. The other legumes used in pre-cultivation showed similar results for the studied parameters, except for *Canavalia ensiformis* and *Mucuna aterrima*, which led to increases in SD. Our results also showed no differences for PH and RDM.

The highest values of coffee growth parameters in RLm can be justified by its higher available P contents. The Law of the Minimum says that plant growth is limited by the low availability of an essential element in the soil, failing to meet plant demands, even if the other nutrients are available in balanced amounts (Lepsch, 2016). When growing coffee seedlings in soils with high and low levels of P, Neto et al. (2016) also observed a reduction in the production of plant biomass from plants when cultivated in soil with low P content.

Despite the lack of P availability to the growth of coffee seedlings in LVwf, pre-cultivation with *Crotalaria juncea* showed to be a promising alternative to supply P contents to the following crop. Therefore, this legume species stood out for pre-cultivation, providing increases in coffee growth parameters.

Table 5: Breakdown analysis of the interaction between legume species (L) and soil types (S) for coffee seedling shoot K content and root P, Mg, and K contents after 120-day cultivation.

L x S	*Crotalaria juncea*	*Cajanus cajan*	*Canavalia ensiformis*	*Mucuna aterrima*	Without pre-cultivated plant
			Shoot K content (kg)		
LVwf	0.0068aA	5.44bA	0.0064bA	0.0052bA	0.0049aA
RLm	0.0049aB	12.10aA	0.0107aA	0.0126aA	0.0072aAB
			Root P content (kg)		
LVwf	0.0011aA	0.0009bA	0.0009bA	0.0010bA	0.0010aA
RLm	0.0012aB	0.0017aA	0.0016aA	0.0013aB	0.0011aB
			Root Mg content (kg)		
LVwf	0.0094aA	0.0087bA	0.0085bA	0.0087aA	0.0089aA
RLm	0.0092aAB	0.0105aA	0.0103aA	0.0087aB	0.0095aAB
			Root K content (kg)		
LVwf	0.0092aB	0.0110bAB	0.0085bB	0.0097aB	0.0129aA
RLm	0.0103aB	0.0152aA	0.0129aAB	0.0112aB	0.0113aB

LVwf: Typical Acriferic Red Oxisol; RLm: Typical Chernossolic Litholytic Entisol; Different lower-case letters in the column and different upper-case letters in the row differ significantly by the Tukey's test at 5% probability.

Table 6: Nutrient use efficiency (NUE), nutrient use efficiency for shoot production (NUESP), nutrient use efficiency for root production (NUERP), and P translocation index (PTI) in coffee seedlings after 120-day cultivation.

Treatments	NUE	NUESP	NUERP	PTI
	--------- kg^2kg^{-1}---------			%
		Legume species(L)		
Crotalaria juncea	15.45a	10.64a	0.47a	-
Cajanus cajan	10.72ab	7.61ab	0.29a	-
Canavalia ensiformis	9.77b	6.70b	0.31a	-
Mucuna aterrima	12.17ab	8.54ab	0.38a	-
Without pre-cultivated plant	12.46ab	8.82ab	0.34a	-
		Soil type (S)		
LVwf	11.81a	7.96a	0.40a	-
RLm	12.42a	8.96a	0.32a	-
L*S	ns	ns	ns	*
C.V. (%)	35.00	34.53	64.52	8.82
Overall average	12.11	8.46	0.36	80.40

LVwf: Typical Acriferic Red Oxisol; RLm: Typical Chernossolic Litholytic Entisol; L*S: legume species X soil type interaction; C.V.: coefficient of variation; ns: non-significant, and * and * significant at 1% and 5% probability; Different letters in the column differ significantly by the Tukey's test at 5% probability.

Table 7: Breakdown analysis of the interaction between legume species (L) and soil types (S) for PTI of coffee seedlings after 120-day cultivation.

L x S	*Crotalaria juncea*	*Cajanus cajan*	*Canavalia ensiformis*	*Mucuna aterrima*	Without pre-cultivated plant
LVwf	78.82aA	82.06aA	80.31aA	73.86bA	78.02aA
RLm	77.70aB	80.82aAB	83.14aAB	89.48aA	79.76aAB

LVwf: Typical Acriferic Red Oxisol; RLm: Typical Chernossolic Litholytic Entisol; Different lower-case letters in the column and different upper-case letters in the row differ significantly by the Tukey's test at 5% probability.

The cycling of P and other nutrients is important for coffee nutrition (Cardoso et al., 2018) due to reductions in costs and dependence on industrial inputs for coffee growing (Vilela et al., 2011). Green manuring is a technique that has shown promising results for coffee plants (Paulo et al., 2006; Araújo et al., 2013; Vilela et al., 2011; Cardoso et al., 2018; Jaeggi et al., 2019; Franco Junior et al., 2019), either in pre-cultivation or intercropped in-between rows of already-established crops.

4.2 Macronutrients in coffee shoots and roots

Our results show that coffee seedlings grown after *Canavalia ensiformis* had higher shoot levels of P, Ca, Mg, and S compared to the control. Unlike, no differences were observed for Namong legume species. Thus, pre-cultivation with *Canavalia ensiformis* promote benefits to posterior coffee cultivation. Legumes can mobilize nutrients and make them available to successor crops. Therefore, green manuring in coffee plantations can reduce the requirements for fertilizer application over time.

Our findings also revealed a significant interaction between soil types and legume species for P, Mg, and K levels. Oppositely, the interaction between Ca and S levels in coffee roots (Table 4) had no significant results. The content of Ca was higher in roots of seedlings grown in RLm, while those of S showed no differences between soil types. Regarding the effect of pre-cultivation with legume species, the contents of Ca and S in coffee roots exhibited no significant differences.

4.3 Nutritional efficiency of phosphorus in coffee seedlings

According to Amaral et al. (2011), the term "nutritional efficiency" has been used to characterize plant ability to absorb and use nutrients, which is related to its efficiency of absorption, translocation, and use of nutrients. In most studies on nutritional efficiency, P-use efficiency has been increasingly evidenced (Amaral et al., 2011). However, in our study, an increase in P availability did not allow us to identify differences between coffee seedlings grown in soils with different P availability. This might have occurred because only a single species and cultivar was assessed. Different plant species differ in their responses to soil nutrient availability, as they show differences in their absorption capacity and use efficiency (Tomaz et al., 2008).

Pre-cultivation with legumes, in turn, enabled us to detect significant differences between coffee seedlings, especially for those grown after *Crotalaria juncea*. Phosphorus mobilization by this legume species during its cultivation may have increased P-use efficiency and coffee shoot and root productions per P unit. Franco Junior et al. (2019) observed that the consortium with *Crotalaria juncea* showed positive responses to the coffee initial development in the field and better conditions of soil humidity and temperature. *Crotalaria juncea* can still bring other benefits to coffee, as observed by Supriyad et al. (2019), in which *Crotalaria juncea* stimulated the increase in abundance of predators and parasitoid insects in the coffee ecosystem.

Translocation refers to the movement or transfer of an ion from its absorption site in roots to any other point in the plant, whether inside or outside the root system (Malavolta; Vitti; Oliveira, 1997). Phosphorus translocation index (PTI) is expressed as the percentage of P transferred to plant shoot in relation to the total amount absorbed (Abichequer; Bohnen, 1998). In this sense, the higher the PTI, the greater the translocated amount from plant roots to shoot (Silva; Vitti; Trevizam, 2007). In our study, there was a high P translocation from coffee seedling roots to shoot, even when grown in the low-P availability soil. Contrary to what occurs in soil, P has high mobility within plant tissues, easily translocating from roots to shoot (Novais; Smyth, 1999).

5 CONCLUSIONS

Pre-cultivation with legume species increases coffee seedling growth, mainly with *Crotalaria juncea* plants;

Coffee seedlings grown after *Canavalia ensiformis* cultivation have higher shoot levels of P, Ca, Mg, and S;

Pre-cultivation with *Crotalaria juncea* can be a promising alternative to supplement phosphorus to successor crops in low-P availability soils.

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