

Full Length Research Paper

Growth of *Piper nigrum* L. and nutrients cycling by intercropping with leguminous species

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Received 25 April, 2016; Accepted 19 September, 2016

Nitrogen (N) fixation is performed efficiently by leguminous plants, which can then be used as 'green manure' to reduce the need for synthetic fertilizer (especially N) and thus sustainably provide satisfactory crop yields with low production costs. The aim of this study was to determine the effect of intercropping with leguminous species used as cover crops on branch growth and nutrient cycling of pepper plants. The experiment was carried out in an indoor system with a randomized block design with four replicates. The treatments consisted of seven leguminous plant species which were planted as cover crops for black pepper plants: *Canavalia ensiformis*, *Mucuna deeringiana*, *Mucuna pruriens*, *Cajanus cajan*, *Crotalaria juncea*, *Crotalaria spectabilis* and the uncovered control. Samples of leaves and branches were harvested from flowering leguminous plants to quantify the production of dry matter. In addition, macro- and micro-nutrients content was measured in the legumes. The growth of the main branch of the pepper plants was measured at 30 and 60 days after the leguminous cover crops were cut. The leguminous plants, *M. pruriens*, *C. ensiformis*, *C. cajan* and *C. juncea* provided higher accumulation of nutrients and dry matter production in the pepper plants. At 60 days after cutting, intercropping with *C. ensiformis* supported more growth of pepper plants as determined by main branch measurements.

Key words: Black pepper, cover crops, soil fertility management, soil conservation.

INTRODUCTION

The black pepper (*Piper nigrum* L.), belonging to the family, Piperaceae, is a perennial plant of Indian origin which is cultivated in many tropical regions. Due to its peculiar taste, this plant is commonly used for food flavoring, preparation and food processing, characterizing

it as an important spice which is sold worldwide (Hussain et al., 2011; Nair, 2011).

Among black pepper-producing countries, Brazil stands out as the third largest producer of this crop, with more than 40 thousand tons produced annually. In the Brazilian

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states of Para and Espírito Santo, 79 and 13% of agricultural production is represented by black pepper cultivation, respectively. Espírito Santo Farmers have better access to technologies such as irrigation systems in approximately 80% of the area planted with this crop, which has led to higher yields as compared to Para (Partelli, 2009). In addition to these technologies, these farmers use legumes as a source of nutrients for the cultivated plants to ensure greater agricultural sustainability.

The impact of conventional agriculture on the environment is easily perceived, compromising the sustainability of agricultural activity. Sustainable agriculture seeks to combine aspects of socio-cultural, economic and environmental research in order to implement conservative agricultural practices, such as the use of 'green manure' as opposed to industrial soil additives (nitrogen-N fertilizer), to promote the sustainability of ecosystems. A common form of 'green manure' is provided by the cultivation of legumes alongside food crops, which promotes nutrient cycling, provides biological fixation of atmospheric N (Partelli et al., 2011; Moda et al., 2014), favors the density and diversity of edaphic micro-organisms (Ferreira et al., 2010), improves soil structure (Cunha et al., 2010) and can be an efficient strategy to maintain productivity in the low-fertility soils of the humid tropical (Moura et al., 2010; Aguiar et al., 2010) as compared to fertilization with industrially produced mineral N only.

The effectiveness of green manure provided by legumes is superior to any other group of plants, because of the specific symbiotic associations that legumes form with N fixing bacteria, which result in a high N concentration in the plant (Partelli et al., 2011; Souza et al., 2015). Besides providing N, leguminous plants may enhance the quality of soil organic matter, and consequently the ability to exchange cations (Partelli et al., 2011).

While it is well established that soil fertility can be improved by substituting mineral N for naturally occurring N_2 fixers, resulting in satisfactory yield with relatively low production costs, there are limited studies available in the literature describing the use of leguminous intercrops as green manure to promote the growth of pepper plants. Given this, the present work aimed to establish the usefulness of leguminous intercrops in pepper plants by quantifying the growth of the branches of black pepper plants and nutrient cycling provided by legumes.

MATERIALS AND METHODS

The experiment was conducted in smallholder farmer's field which is located in the municipality of Jaguare, Espírito Santo-Brazil (18° 54' 20" S, 40° 04' 34" W and altitude of 80 m) in Oxisol dystrophic soil (Santos et al., 2013) of sandy-clay texture. The climate is classified as Aw according to the Köppen climate classification and is tropical with a dry winter and a rainy summer (Alvares et al., 2013).

The experiment was conducted in black pepper plants planted in a randomized block design with four replicates, consisting of seven treatments. Each treatment block was planted alongside a different species of legume to be used for green manure: *Canavalia ensiformis* (jack bean), *Mucuna deeringiana* (mucuna dwarf), *Mucuna pruriens* (velvet bean), *Cajanus cajan* (pigeon pea), *Crotalaria juncea*, *Crotalaria spectabilis* and control (without green manure).

The black pepper plants were planted with 3.0 m between rows and 1.8 m between plants, under a sprinkler irrigation system. Each experimental unit consisted of two rows of seven plants, totaling 75.6 m². Within each experiment unit, the leguminous plants were planted in pits by hand, in January 2012. Two rows of leguminous plants were planted, with 0.4 m between plants and 0.5 m between rows, one on each side of the rows of pepper plants. On average, four legume seeds per hole were planted.

In order to measure the dry biomass production of the various leguminous plants, about 300 g of fresh biomass consisting of branches and leaves were collected from each experimental unit (one or two plants, depending on the species) at 70 days after sowing, when the plants were flowering. These were obtained from the four central meters of each row in each experimental unit, comprising an area of 24 m². Subsequently, these plants were dried in an oven with forced air ventilation at 65°C, until they reached a constant mass. After drying, the plant material was weighed on an analytical balance. Immediately after weighing, the plant material was ground in a Wiley mill and a subsample of ground plant material was digested in concentrated sulfuric acid. These samples were analyzed for macronutrient content according to the methodology described by Silva (2009). The nutrient content of the aerial parts of these leguminous plants was obtained by multiplying the weight of dry matter by the nutrient content measured in that dry matter.

In order to assess the effectiveness of each leguminous plant species as green manure, the vegetative growth of the apical branches of the pepper plants was measured at 30 and 60 days after cutting the leguminous plants (DAC). The distribution of the data was assessed using Lilliefors test, and transformed using the function $y = \text{Log}(x+10)$. Subsequently, a one way analysis of variance (ANOVA) was performed on the transformed data. Duncan test ($P \leq 0.05$) was used to compare the treatment means of all available variables. All statistical analyses were performed with the statistical software ASSISTAT 7.7 beta (Silva and Azevedo, 2006).

RESULTS AND DISCUSSION

In this study, the effect of using green manure generated by leguminous intercropping for six species of legumes was determined for black pepper plants by measuring the nutrient content of the legumes as well as the growth of the pepper plants. The legumes, *Mucuna* and *C. ensiformis* generated higher average dry biomass, which did not differ significantly from the average of legumes *C. cajan* and *C. juncea* (Table 1). This is similar to the findings of Partelli et al. (2011), who studied the cycling of nutrients in coffee plants. According to Partelli et al. (2011), this increase in biomass production by these legume species as compared to other leguminous plants studied, can be attributed to their generally increased seasonal growth.

It is known that dry matter yield in these species is lower in soil without supplementation by mineral fertilizer and without liming, due to a greater need for soil fertility

Table 1. Dry matter production of leguminous plant grown alongside with pepper plants.

Treatments	Dry matter (kg ha ⁻¹)
<i>Mucuna pruriens</i>	2949.67 ^a
<i>Canavalia ensiformis</i>	2301.96 ^a
<i>Cajanus cajan</i>	2231.67 ^{ab}
<i>Crotalaria juncea</i>	1387.64 ^{ab}
<i>Crotalaria spectabilis</i>	567.73 ^{bc}
<i>Mucuna deeringiana</i>	522.90 ^c
CV ¹ (%)	8.35

Means followed by the same letters in the same column do not differ by Duncan test ($p > 0.05$).

¹Coefficient of variation.

for these species (Nyatsanga and Pierre, 1973; Howard and Rees, 1996). For this study, the low dry biomass production demonstrated by these legumes may indicate that the soil fertility under study conditions may have been insufficient. This shows that the choice of cover crops must be adapted to the local climate and soil conditions in cases where high biomass production capacity is required. Higher amount of macro/micro-nutrients was measured in *M. pruriens* and *C. ensiformis* followed by *C. cajan* and *C. juncea* (Table 2), probably because these species present the greatest bio-matter production index (Table 1) and ability to adapt to the study climate.

Similar results were reported by Silva et al. (2014) when studying the effect of straw, nutrient levels and soil cover for plant coverage seeded in summer for directly sowed beans. These authors observed that the greatest accumulation of N, P, K, Ca, and S was obtained in *M. pruriens*, and attributed the results to higher biomass of the shoots produced by this crop. The accumulation of nutrients depends on the species used, the phenological stage, the dry matter production of shoots and the cultivation period (Partelli et al., 2011).

Studies revealed that the capacity to accumulate nutrients in the tissues of legumes contributes to the ability of these species to recycle nutrients from the soil and atmosphere, and later (after cutting and decomposition) make them available to plants in 'green manure' (Oliveira et al., 2002). These desirable characteristics are augmented by the beneficial role of cover plants to the soil, where they reduce the flow of water loss at the surface and reduce soil erosion and the loss of nutrients, increase the amount of available organic matter, and provide improved nutrient bio-availability for plants (Partelli et al., 2011; Souza et al., 2015).

The ability of the cover crops used in this study to absorb nutrients and immobilize them followed a similar trend regarding the production of dry biomass. The most efficient legume for green manure would ideally be efficient both at accumulating and absorbing nutrients and producing the most biomass. Another aspect to

consider is the environmental contribution of intercropping afforded by soil protection, which reduces losses by processes such as leaching, due to better soil structure, and also reduces the entrainment of particles by erosion and others that promote loss in quality soils (Chaves et al., 1997).

Intercropping with the leguminous plants *C. ensiformis*, *C. juncea* and *C. cajan* provided the highest average growth of pepper branches at 30 days after cutting (Table 3). At 60 days after cutting, the pepper plants grown alongside the species *C. ensiformis* had the highest average growth of apical branches. These results indicate that the growth of pepper plants is improved with complementary fertilization arising from the decomposition of plant material from intercropped leguminous plants. This improvement may be attributed to the greater dry matter production (Table 1) and accumulation of macronutrients measured for *C. ensiformis* (Table 2) because of their proven ability to fix nitrogen in the soil and their contribution to integral agricultural production systems, which is responsible for growth and development of pepper plant. According to Tabaldi et al. (2012), leguminous plants, *C. ensiformis* are adapted to conditions of low fertility and high temperatures, have the fast growing ability, are capable of producing large amounts of biomass in a short period of time, and are capable of uptake nutrients from deeper layers, ensuring a large residual effect for the culture.

At 30 and 60 days after cutting, the species, *M. deeringiana* and *M. pruriens* did not differ from the control treatment. These results indicate that these two species may provide inadequate green manure for pepper plants; however, these species could still be used as a cover crop to maintain soil moisture. The choice of legume species for cover crop is related to the quantity and quality of material produced by the legume, with the amounts of nutrients released from the waste during the decomposition process required to meet the needs of subsequent cultures (Mendonça and Stott, 2003). The other species may be referred to as high quality, or important for nutrient cycling, in this regard. Still, these results indicate the phytotechnic benefit of implementing a consortium of plants grown alongside legume species.

The species, *M. pruriens* produced the highest average dry biomass and the highest accumulation of macro and micronutrients measured. On the other hand, this species apparently contributed the least to the growth of nearby pepper plants. This may be because this species is a natural climber which somehow entangled the branches of the pepper plants, resulting in reduction in vegetative growth. For this reason, *M. pruriens* is not indicated for intercropping with pepper plants, unless it is plowed under before it begins to climb.

Conclusions

Leguminous *M. pruriens*, *C. ensiformis*, *C. cajan* and *C.*

Table 2. Accumulation of macronutrients and micronutrients in shoots of leguminous grown alongside with pepper plants.

Treatments	Macronutrients (kg ha ⁻¹)					
	N	P	K	Ca	Mg	S
<i>Canavalia ensiformis</i>	92.65 ^a	5.66 ^{ab}	43.16 ^{ab}	48.87 ^a	6.90 ^a	5.59 ^a
<i>Mucuna pruriens</i>	85.27 ^a	6.87 ^a	64.54 ^a	38.67 ^{ab}	6.46 ^a	5.81 ^a
<i>Cajanus cajan</i>	61.70 ^{ab}	4.77 ^{ab}	33.47 ^b	23.41 ^b	4.32 ^{ab}	3.72 ^{ab}
<i>Crotalaria juncea</i>	38.46 ^{ab}	2.31 ^{ab}	19.94 ^b	14.21 ^c	3.30 ^{ab}	1.91 ^b
<i>Mucuna deeringiana</i>	21.33 ^b	1.39 ^{ab}	9.15 ^c	7.80 ^c	1.34 ^b	1.11 ^b
<i>Crotalaria spectabilis</i>	18.95 ^b	0.82 ^b	9.93 ^c	8.37 ^c	1.24 ^b	1.12 ^b
CV ¹ (%)	12.11	8.35	11.71	11.28	6.17	5.56

Treatments	Micronutrients (g ha ⁻¹)				
	Fe	Zn	Cu	Mn	B
<i>Canavalia ensiformis</i>	250.9 ^b	85.2 ^b	11.5 ^{ab}	200.3 ^a	59.9 ^{ab}
<i>Mucuna pruriens</i>	436.6 ^a	150.4 ^a	23.6 ^a	209.4 ^a	88.5 ^a
<i>Cajanus cajan</i>	183.0 ^b	75.9 ^b	11.2 ^{ab}	93.7 ^b	64.7 ^{ab}
<i>Crotalaria juncea</i>	99.9 ^b	73.5 ^b	5.6 ^b	144.3 ^{ab}	43.7 ^b
<i>Mucuna deeringiana</i>	55.4 ^c	37.1 ^b	5.2 ^b	79.5 ^b	20.4 ^b
<i>Crotalaria spectabilis</i>	43.7 ^c	26.7 ^b	2.8 ^b	31.8 ^c	16.5 ^b
CV (%)	0.44	0.18	0.6	0.27	0.14

Means followed by the same letters in the same column do not differ by Duncan test ($p > 0.05$).

¹Coefficient of variation.

Table 3. Vegetative growth (cm) of the main branches of pepper plans, 30 and 60 days after cutting the leguminous plants.

Treatments	Branches growth (cm)	
	30 Days	60 Days
Control	9.2 ^b	12.9 ^e
<i>Canavalia ensiformis</i>	16.8 ^a	22.4 ^a
<i>Crotalaria juncea</i>	14.9 ^a	18.9 ^b
<i>Cajanus cajan</i>	14.4 ^a	18.0 ^{bc}
<i>Crotalaria spectabilis</i>	11.4 ^b	15.8 ^{cd}
<i>Mucuna deeringiana</i>	11.6 ^b	14.1 ^{de}
<i>Mucuna pruriens</i>	10.9 ^b	13.0 ^e
CV ¹ (%)	18.93	13.43

Means followed by the same letters in the same column do not differ by Duncan test ($p > 0.05$). ¹Coefficient of variation.

juncea had the highest measured accumulation of nutrients and dry biomass production among the legume species considered in this study. Of these, *C. ensiformis* apparently contributes most to the growth of black pepper plants at 60 days after cutting.

Conflict of interest

The authors did not declare any conflict of interest.

ACKNOWLEDGEMENTS

The authors thank Coordination for the Improvement of Higher Education Personnel (CAPES), National Research Council (CNPq) and Foundation to Research Support in the State of Espírito Santo (FAPES) for the grants, project resources and for the scholarships. The corresponding author thank Maranhão Foundation for the Protection of Research and Scientific and Technological Development (FAPEMA) for Fellow of Productivity in Research.

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