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## Initial growth of two varieties of pigeon pea (*Cajanus cajan*) in the tropics of Ecuador

Crecimiento inicial de dos variedades de gandul (*Cajanus cajan*) en el trópico de Ecuador

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### ABSTRACT

The objective was to assess the production and efficiency of the initial growth of pigeon pea or guandul (*Cajanus cajan*) var. Negro and EGV22 with and without nitrogen fertilizing. The plants were planted in plastic bags during summer 2012. Two nitrogen fertilization treatments were applied at 90 d of germination (+N and -N). The variables were: number of nodules (NN), length of root (LR), weight of nodules (WN), height of plant (HP), total dry matter (TDM), leaf area index (LAI), specific leaf area (SLA), and rate of assimilation (RA), relative rate of growth (RRG) and rate of crop growth (RCG). A full-randomized experimental design was used with a factorial arrangement 2 x 2 and regression analysis. Differences were observed ( $P < 0.05$ ) among varieties for NN, HP, so and RCG, where the highest average HP was obtained by EGV22 variety. For the nitrogen factor, its application decreased ( $P < 0.05$ ) NN, WN, and HP. It is concluded that Negro variety can be used in the production systems since it has great TDM and the best establishment for the environmental conditions of Ecuador tropic.

**Keywords:** nitrogen fertilization, legumes, growth rates.

### RESUMEN

El objetivo fue evaluar la producción y eficiencia del crecimiento inicial de dos variedades de Gandul (*Cajanus cajan* var. Negro y EGV22) con y sin fertilización nitrogenada. Las plantas fueron sembradas en bolsas de plástico durante el verano del 2012. Se aplicó a los 90 d de germinación dos tratamientos de fertilización nitrogenada (+N y -N). Las variables fueron: número de nódulos (NN), largo de raíz (LR), peso de nódulos (PN), altura de planta (AP), materia seca total (MST), índice de área foliar (IAF), área foliar específica (AFE), tasa de asimilación neta (TAN), tasa relativa de crecimiento (TRC) y tasa de crecimiento del cultivo (TCC). Se utilizó un diseño experimental completo al azar con arreglo factorial 2x2 y análisis de regresión. Se observó diferencia ( $P < 0.05$ ) entre variedades para NN, AP, TAN y TCC, donde el mayor promedio de AP lo obtuvo la variedad EGV22. Para el factor nitrógeno, su aplicación provocó disminución ( $P < 0.05$ ) en NN, PN y AP. Se concluye que la variedad Negro, puede ser utilizada en los sistemas de producción pecuarios, ya que tiene una buena producción de materia seca y un mejor establecimiento, adaptándose mejor a las condiciones ambientales del trópico de Ecuador.

**Palabras clave:** fertilización nitrogenada, leguminosas, tasas de crecimiento.

## INTRODUCTION

Worldwide, the constant growth of the human population has increased the demand for food. In Ecuador, according to data from the III National Agricultural Census (SINAGAP, 2012), 11.0% of the land planted corresponds to perennial crops, 27.2% to cultivated pastures and 9.1% to natural pastures; the same ones that can be used for the production of animal protein (De la Fuente *et al.*, 2008).

However, the opening of land for human settlements deprives the soil of protection, loses its fertility and, since it is not used for agricultural or livestock purposes, it prevents the development of new agricultural production systems (Petit *et al.*, 2009).

In addition to the above, in Ecuador as in most of the tropical regions, the production and availability of quality fodder is limited during the dry season: so the planting of legumes can help the recovery and conservation of the soil (Olivera *et al.*, 2005; Nieuwenhuys *et al.*, 2008; Ruiz *et al.*, 2015) to fix atmospheric nitrogen, being tolerant to drought and have the capacity to produce forage with a high value of proteins and minerals that when consumed by animals increases their productivity.

Initial growth is important because it is the most vulnerable stage of a plant, indicates its capacity to accumulate biomass, compete for resources, and its ability to adapt to a specific site (Villar *et al.*, 2008).

The pigeon pea (*Cajanus cajan*), also known as palo bean, pigeon pea, among others, is a shrub legume, tolerant to drought and soils with low nutrient content. In addition, to serve for human food; the forage can be used for animal feed, because it has high levels of protein (16 to 22%) and a digestibility of dry matter of approximately 59% (Peters *et al.*, 2003; Carvajal-Tapia *et al.*, 2016), being able to be supplied in green material in grazing or as dry fodder in the corral (Trómpiz *et al.*, 2001).

Therefore, the objective of the present study was to evaluate the initial growth of two varieties of gandul (Negro and EGV22) with and without nitrogen fertilization on some productive and agronomic characteristics in the tropics of Ecuador.

## MATERIAL AND METHODS

The research was carried out at the Experimental Farm "El Oasis", of the Equinoctial Technological University, Campus Santo Domingo, Ecuador; during the summer of 2012. The experimental site is located at 00° 13 '37 "LS and 79° 15' 04" LO; the prevailing climate is humid tropical with an annual average temperature of 23.5 °C and annual precipitation of 2700 mm distributed mainly in the summer months (Miranda, 2010).

### Evaluated material

The sowing of Gandul varieties (Negro and EGV22), was carried out in black plastic bags with a capacity of approximately 4 kg, two seeds were placed per bag to later leave

a plant as an experimental unit. The soil used was of volcanic origin (Andisol-USDA) that was collected at a depth of 0 to 20 cm (Table 1).

**Table 1. Chemical characteristics of the soil used in the experiment.**

pH	OM	NH <sub>4</sub>	P	S	Fe	Cu	Zn	Mn	B	K	Ca	Mg
	%	mg kg <sup>-1</sup>								cmol kg <sup>-1</sup>		
5.9	2.2	41.0	6.5	6.3	42.0	5.6	1.9	2.8	0.3	0.3	8.3	2.9

OM = Organic material, NH<sub>4</sub> = Ammonium nitrate, P = phosphorus, S = sulfur, Fe = iron, Cu=copper, Zn = zinc, Mn = magnesium, B = boron, K = potassium, Ca = calcium, Mg = manganese. Laboratory of Chemistry, Equinoctial Technological University from Santo Domingo Campus

### Description of treatments

Two types of fertilization with nitrogen (+N) were considered, which in the early stages of the growth of legumes requires nitrogen fertilization, because the symbiosis between legumes and rhizobium has not been developed. While the other treatment was without nitrogen (-N), soil fertilization was performed at the time of preparing the planting. For the -N treatment, 54.0 kg ha<sup>-1</sup> of phosphorus (P, Super simple phosphate, FERTISA, Guayaquil, Ecuador) and 194.0, 86.0 and 44.0 kg ha<sup>-1</sup> of potassium (K, potassium chloride), sulfur were incorporated into the soil. (S) and magnesium (Mg) (K-mag, FERTISA, Guayaquil, Ecuador).

While for the treatment +N, 137.0 kg ha<sup>-1</sup> of N (Urea, FERTISA, Guayaquil, Ecuador), 81.0 kg ha<sup>-1</sup> of P (Diammonium Phosphate, FERTISA, Guayaquil, Ecuador), 137.0, 53.0 and 44.0 kg ha<sup>-1</sup> were applied of K, S and Mg (K-mag), respectively, according to the recommendations of the manual of nutrition and pasture fertilization (Bernal and Espinosa, 2003).

In addition, during the period of plant growth, fertilizations were carried out at the foot of the plants, fifteen days after germination and then every 30 days; for -N they were applied 1.0 g of K-mag fertilizer, which corresponded to doses of 297.0, 243.0 and 297.0 kg ha<sup>-1</sup> of K, S and Mg, respectively. While for the +N treatment, 1.0 g of ammonium sulfate was added, which corresponded to a dose of 284.0 kg ha<sup>-1</sup> of N.

### Study variables

In the present work we measured: the length of the root (LR), from the neck to the cap; number of nodules (NN), healthy (pink color) and vain (gray); the nodules were extracted in water to avoid damage; nodule weight (WN); plant height (HP), from the base of the stem to the apex; number of branches (NR); number of leaves (NL); length of branches (Lr) and total dry matter weight (TDMW). The samples were dried in an oven at 65 °C until constant weight was obtained; leaf area (LA); using the methodology suggested by Rincon *et al.* (2012).

The physiological efficiency of the plant (EFP) was determined through growth as a function of time (weeks). To make this type of analysis, two basic measurements were used, the dry matter production of the aerial fraction of the plant material and the foliar area (Carranza *et al.*, 2009). The parameters used to estimate the physiological

efficiency of the plant were: leaf area index (LAI), specific leaf area (SLA) and rate of the crop growth (RCG); determined by the formulas established by Hunt (1990); rate of net assimilation (RNA) and rate of relative growth (RRG) determined by the formulas described by Tayeb (2012).

### Regression analysis and experimental design

To estimate the growth curves or trend lines over time of each of the two nitrogenous fertilization treatments, a regression analysis was used with 12 destructive samplings (one sampling per week after germination) and five repetitions within each sampling (SAS, 2012).

For the analysis of continuous variables, a randomized complete block design with factorial arrangement (2 x 2) was used and a comparison test of means was carried out with the Tukey test at 5% (SAS, 2012).

## RESULTS

In the present work, no differences were observed ( $P > 0.05$ ) in number of nodules (NN), root length (LR), weight of nodules (WN), stem diameter (DT), plant height (HP), number of branches (NR) and number of leaves (NL) in the two varieties of Guandul or pigeon pea; except ( $P < 0.05$ ) for length of branches (Lr, Table 2).

Similar situation was observed for nitrogen treatment; the plants that did not receive fertilization (-N) showed a greater number of nodules (82.0) than the plants that received nitrogen (+ N), where 60.8 nodules were counted ( $P < 0.05$ , Table 2). Another variable that showed significant effects ( $P < 0.05$ ) of the fertilization was the length of branches (Table 2).

The pigeon pea (*Cajanus cajan*) showed a major root growth in the treatment without fertilization in the Negro variety (66.4 cm); while the LR in the treatment with nitrogen (+ N) growth was similar ( $P > 0.05$ ), in both varieties (Negro and EGV22) with 60.2 and 61.0 cm, respectively. Similarly, for WN and NN it was observed that the nodules present in the Negro variety with -N, were in greater number (86) and heavier (4 g); but equal ( $P > 0.05$ ) to that found for the variety EGV22 with + N and -N, which were in a range of 57 to 77 nodules with a weight of 2 to 3 g, respectively.

**Table 2. Means  $\pm$  standard deviation of number of nodules (NN), root length (LR), weight of nodules (WN), stem diameter (DT), plant height (HA), number of branches (NR), length of branches (Lr) and number of leaves (NL) in two varieties of pigeon pea and two levels of fertilization in Santo Domingo, Ecuador.**

Variety	NN	LR (cm)	WN (g)	DT (mm)	HP (cm)	NR	Lr (cm)	NL
Negro	75.6 $\pm$ 41.7	63.3 $\pm$ 28.4	3.0 $\pm$ 2.5	6.1 $\pm$ 2.3	85.2 $\pm$ 49.0	0.7 $\pm$ 1.6	4.8 $\pm$ 9.2 <sup>a</sup>	18.0 $\pm$ 11.6
EGV22	67.1 $\pm$ 34.0	60.9 $\pm$ 22.6	2.7 $\pm$ 1.8	6.1 $\pm$ 2.1	91.7 $\pm$ 49.7	1.1 $\pm$ 1.8	9.0 $\pm$ 13.7 <sup>b</sup>	21.6 $\pm$ 15.3
Nitrogen								
-N	82.0 $\pm$ 38.3 <sup>a</sup>	64.1 $\pm$ 27.0	3.7 $\pm$ 2.1	6.1 $\pm$ 2.1	93.9 $\pm$ 51.4	0.8 $\pm$ 1.4	5.4 $\pm$ 9.7 <sup>a</sup>	18.9 $\pm$ 11.1
+N	60.8 $\pm$ 35.2 <sup>b</sup>	60.1 $\pm$ 24.1	2.0 $\pm$ 1.8	6.0 $\pm$ 2.4	83.0 $\pm$ 46.9	1.1 $\pm$ 2.0	8.3 $\pm$ 13.6 <sup>b</sup>	20.6 $\pm$ 15.9

-N = without nitrogen; + N = with nitrogen; \* Averages with a, b are different statistically significant ( $P < 0.05$ )

In Table 3 it can be seen that no significant differences were found for the variety interaction by fertilization for NN ( $P = 0.90$ ), LR ( $P = 0.40$ ), WN ( $P = 0.18$ ) and stem diameter ( $P = 0.37$ ). The growth trend for LR was quadratic with a lower development for the variety EGV22 with -N from week five of growth after germination. The NN and WN with +N had a linear trend of growth, therefore the amount of NN and its weight was gradually increasing throughout the study weeks. While for the plants with -N the NN and WN showed a quadratic growth, with a stabilization of the number and weight of nodules from the eighth week; this reveals that under the conditions in which the experiment was conducted, the WN had a relationship directly proportional to the NN, since as its quantity was reduced, its weight also did (Figure 1). On average, NN and WN showed a decrease ( $P < 0.05$ ) when applying nitrogen. It is important to mention that the trend observed for LR after the eighth week of growth may have been due to space restriction, which prevented further development and growth of the root system.

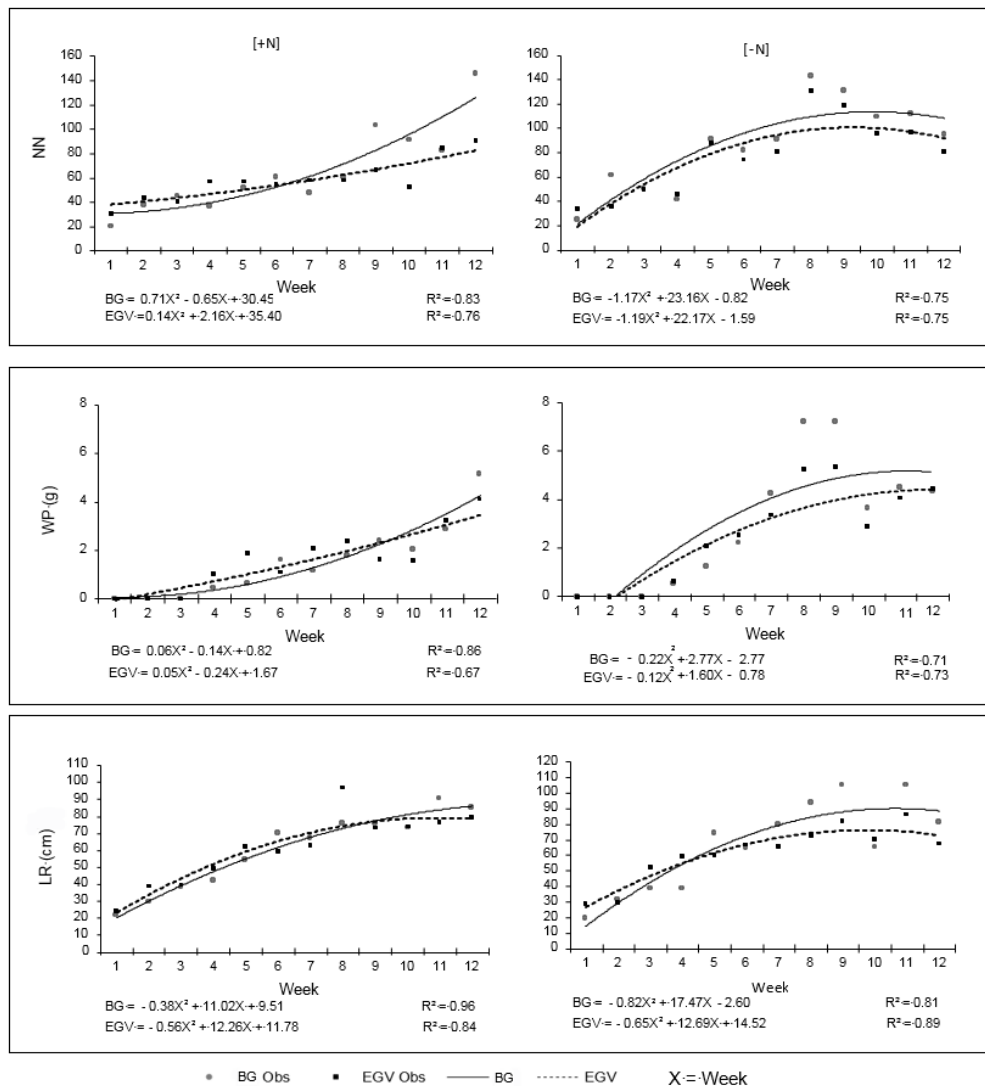


Figure 1. Length of root (LR), number of nodules (NN) and weight of nodules (WN) for two varieties of pigeon pea (*Cajanus cajan*; Negro and EGV22) with (+N) and without (-N) nitrogenated fertilization in Santo Domingo, Ecuador.

**Table 3. Means ± standard deviation for the following variables, number of nodules, length of root, weight of nodules and diameter of stem according to the interaction of pigeon pea varieties and fertilization level in Santo Domingo, Ecuador**

Variety	Nitrogen	Number of nodules	Length of root (cm)	Weight of nodules (g)	Diameter of stem (cm)
Negro	-N	86.2±40.5	66.5±31.2	3.9±2.5	6.3±2.4
	+N	65.0±40.4	60.2±25.2	2.0±2.0	5.9±2.3
EGV 22	-N	77.7±35.8	61.8±22.1	3.4±1.7	6.0±1.7
	+N	56.5±28.6	60.0±23.2	2.1±1.6	6.2±2.4

-N = without nitrogen; +N = with nitrogen.

### Effect of nitrogen in the aerial part of the pigeon pea

Statistical significance was found ( $P < 0.05$ ) for HP between the varieties and effect of fertilization, surpassing the variety EGV22 in  $7.3 \text{ cm p}^{-1}$  to the Negro variety; and for the effect of fertilization the -N treatment, allowed to obtain taller plants with an approximate average of  $10.0 \text{ cm p}^{-1}$  in comparison with the treatment + N; conversely, for the interaction of treatments, no statistical significance was found ( $P > 0.05$ ).

Regarding the TDM, statistical significance was observed ( $P < 0.05$ ) in the variety interaction by fertilization, where the Negro -N variety with  $6181.6 \text{ kg ha}^{-1}$  showed to have greater production of biomass; although statistically similar to the variety EGV22 with + N ( $6129.0 \text{ kg ha}^{-1}$ ) and -N ( $5502.2 \text{ kg ha}^{-1}$ ), being able to overcome with  $1223.0 \text{ kg ha}^{-1}$  the variety Negro +N (Table 4). The same behavior and result of TDM was obtained in LAI, since both the variety EGV22 with +N and Negro with -N, showed the highest LAI with 6.29 and 5.76, respectively. In Figure 2, the initial growth of the aerial part of the legumes is observed, which showed a positive growth behavior over time (weeks).

**Table 4. Mean ± standard deviation of leaf weight in dry matter, weight of stem in dry matter and weight of root in dry matter in two varieties of pigeon pea and in two levels of fertilization in Santo Domingo, Ecuador.**

	Weight of leaves in dry matter (g)	Weight of stem in dry matter (g)	Weight of root in dry matter (g)
Variedad			
Negro	5.41±5.16	0.74±1.60	4.07±4.50
EGV22	5.60±4.93	1.14±1.80	4.33±4.72
Nitrogen			
-N	5.57±4.69	6.10±6.41	4.52±5.05
+N	5.44±5.39	5.54±6.62	3.88±4.10

-N = without nitrogen; +N = with nitrogen.

In HP throughout the experiment the variety EGV22 was positioned above the Negro variety; while the TDM of the legume varieties was affected by the incorporation or restriction of N, from the sixth week. One of the moments in which the fertilization treatment was applied, it was observed that in the +N treatment, the EGV22 variety had a greater increase in TDM; conversely, the Negro variety proved to have more biomass with the -N treatment, a similar tendency observed in TDM was presented in the LAI,

where the variety EGV22 required N to express a greater leaf area, since in the absence of N as shown in figure 2, from the sixth week the LAI began to decline; while in the Negro variety the application or restriction of N did not infer in the manifestation of leaf area ( $P > 0.05$ ).

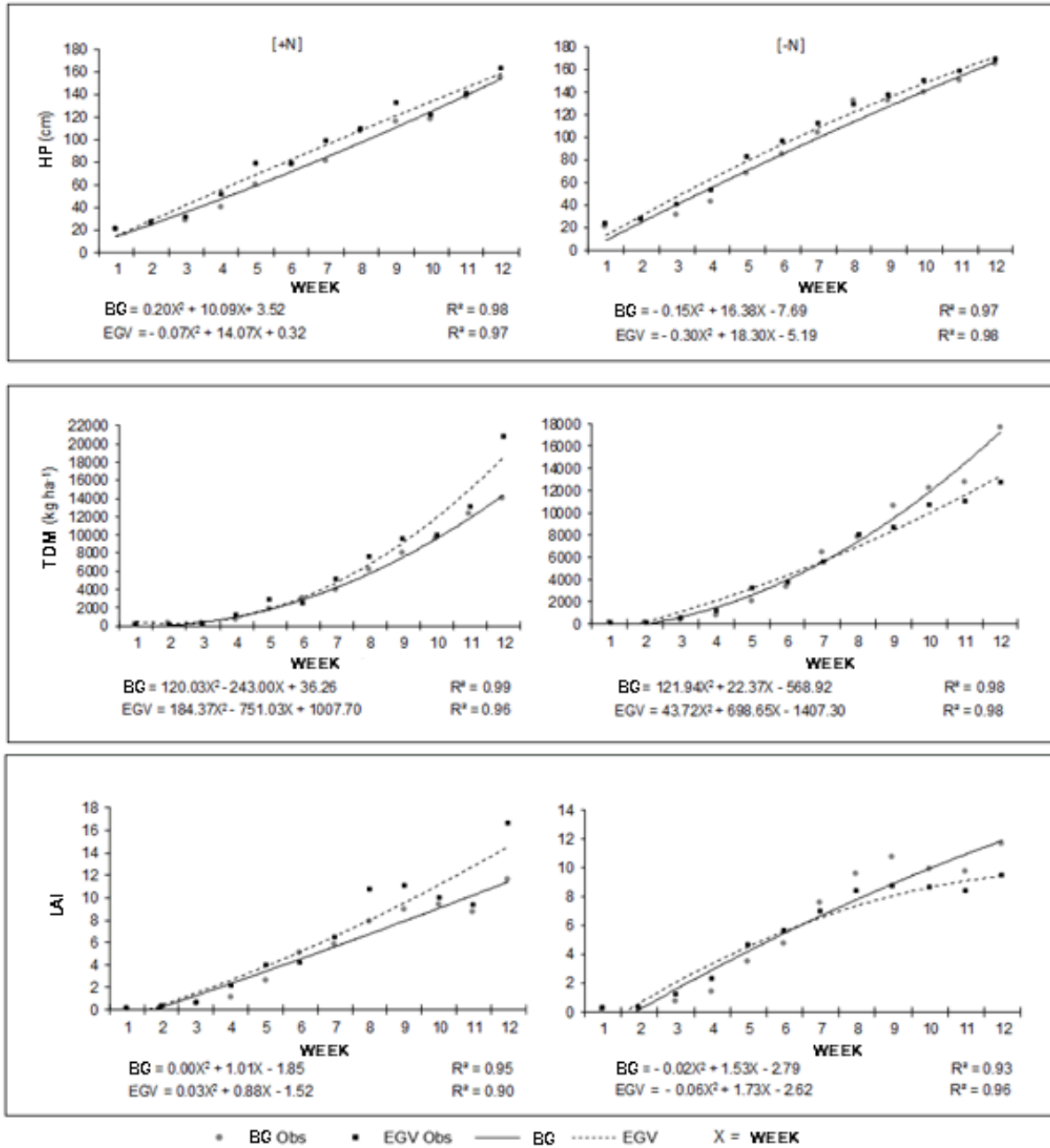


Figure 2. Height of plant (HP), total dry matter (TDM) and leaf area index (LAI), for two varieties of pigeon pea (*Cajanus cajan*, Negro and EGV22) with (+N) and without (-N) nitrogenated fertilization in Santo Domingo, Ecuador.

## Effect of Nitrogen for the physiological efficiency of the pigeon pea

In SLA, no statistical significance was found ( $P > 0.05$ ) for varieties, fertilization treatments and their interaction; that is, they developed the same leaf diameter and area; which were not affected by the application or restriction of N, although their trend over time was negative. In the treatment with +N the Negro variety after the first two weeks of the study until the seventh week, presented a greater decrease of the SLA, to later claim and surpass the EGV22 variety; while in the -N treatment, the two varieties presented a similar trend.

In the case of RNA, only statistical significance ( $P < 0.05$ ) between varieties was observed; being the Negro variety ( $0.00043 \text{ g cm}^2 \text{ d}^{-1}$ ), which showed the highest photosynthetic efficiency since the beginning of the biological cycle, independently of the application or N restriction (Figure 3).

While for the RRG no statistical significance was found ( $P > 0.05$ ) in any of the sources of variation, the Negro variety without -N and with + N had RRG of  $0.058$  and  $0.052 \text{ g g}^{-1} \text{ d}^{-1}$ , respectively.

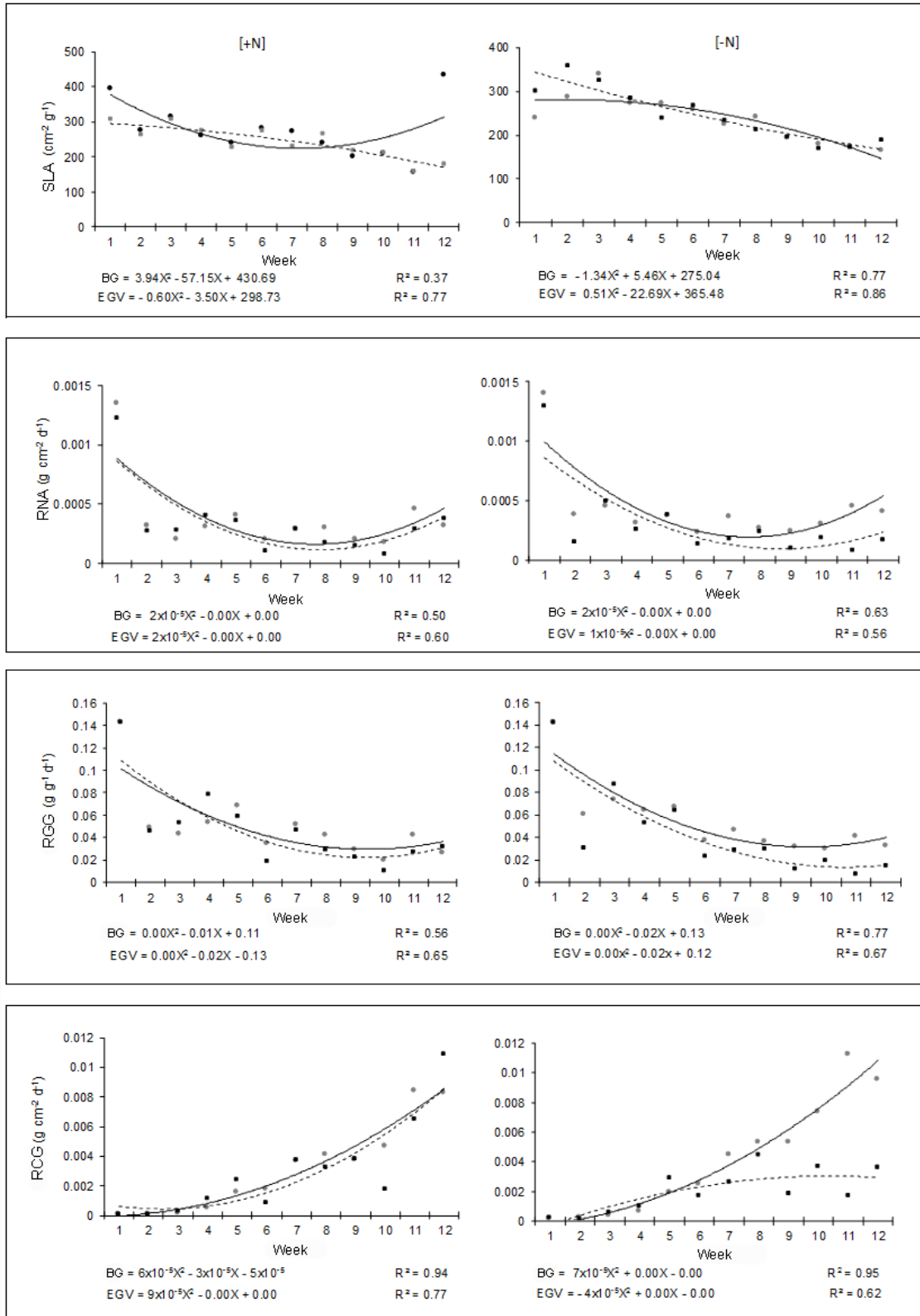
Similarly, in RCG the Negro variety obtained in the -N treatment the highest growth ( $0.0036 \text{ g g}^{-1} \text{ d}^{-1}$ ), statistically different ( $P < 0.05$ ) to the variety EGV22 -N ( $0.0020 \text{ g g}^{-1} \text{ d}^{-1}$ ). The highest RNA and RRG was observed in the first week of the crop cycle, and later declined gradually during the rest of the investigation; this corroborates the inversely proportional relationship that they have with RCG, as a result of the possible shading present in the leaf blades between them (Figure 3).

In all the efficiency variables, the Negro variety exceeded ( $P < 0.05$ ) the variety EGV22, especially in the -N treatment, where approximately from the sixth week the variety EGV22 decreased NRA, RRG and RCG, observing a differential behavior between the varieties (Figure 3). In the variety EGV22 the application of nitrogen fertilizer improved ( $P < 0.05$ ) the variables of biomass production and crop efficiency, allowing an adequate growth and development of the plants.

## DISCUSSION

The plants, being in restrictive conditions of nitrogen can modify the LR to find nutrients and have a greater survival (Villar *et al.*, 2008). In relation to the increase of NN and WN, the results of the present study are similar to that reported by Mayz (2007), who, when evaluating doses of N and P in pigeon pea, mentioned that a low nitrogen content of less than  $20 \text{ kg ha}^{-1}$  is beneficial for nodulation. According to George and Singleton (1992) they mentioned that, at the beginning of the growth of the legumes, the nodulation tends to slow down and in the absence of N the biomass of the root increases; in this regard, Díaz *et al.* (2011) noted that it is important to know the optimum point of fertilization in legumes.





● BG Obs    ■ EGV Obs    — BG    - - - - EGV    X = Week

Figure 3. Specific leaf area (SLA), net accumulation rate (RNA), relative growth rate (RRG) and crop growth rate (RCG), for two varieties of pigeon pea (*Cajanus cajan*; Negro and EGV22) with (+N) and without (-N) nitrogenous fertilization in Santo Domingo, Ecuador.

### **Effect of nitrogen in the aerial part of the pigeon pea**

Martínez *et al.* (2003) who evaluated 25 lines of *Cajanus cajan*, found that the varieties most recommended for the production of forage are those of high size. In addition, Mayz (2007) observed that legumes treated with nitrogen tend to have higher biomass in initial stages, since according to Pliego *et al.* (2003) the application of N can cover the needs of the plants, increasing their total biomass. The results obtained in this work are contrary to those of Pliego *et al.* (2003), since the EGV22 variety presented higher plants, without obtaining higher TDM.

However, the application of N in the EVG22 variety was essential to improve the IAF, but the development will depend on each species (Gómez-Carabalí *et al.*, 2011). For Higuera *et al.* (2001) the differences in the morphological components of the plants are due to the duration of physiological stages of the legumes.

### **Effect of Nitrogen for the physiological efficiency of the pigeon pea**

For the SLA, the trend was negative over the weeks, which could be due to the fact that with the passage of time the thickness of the leaf increases (Villar *et al.*, 2008). According to Grazia *et al.* (2001) fertilization with nitrogen affects the development of foliar structure (leaf area, leaf area duration and leaf expansion rate), but does not modify the leaf area per unit of mass.

A negative trend was observed in the weeks of study for NRA and RRG these same results were mentioned by Hoyos *et al.* (2009), which could be due to the greater accumulation of dry matter, in relation to the production of photoassimilates during the growth of the crop. While the increase in RCG was the result of constant cell division in meristematic tissues and plant architecture (Hernández *et al.*, 1995).

In the variety interaction by fertilization for RCG, according to Villar *et al.* (2008) commented that RCG is influenced by the environmental conditions and genetic components of each species. Hoyos *et al.* (2009) mentioned that RCG can be altered with the application of nitrogenous fertilizer.

## **CONCLUSION**

The application of nitrogen in *Cajanus cajan* negatively influenced the number and weight of the nodules. For forage purposes, the Negro variety -N is a viable option to be used in livestock production systems; since it has the capacity to develop greater leaf biomass during the initial growth, under conditions of restriction of nitrogen fertilizer, demonstrating its adaptation to the conditions of the humid tropic of Ecuador.

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