

## Effect of fertilizers on the biomass production of *Moringa oleifera* and on some soil indicators during the establishment

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The performance of some morphological and productive indicators of *Moringa oleifera* related to compost application, chemical fertilization and bio-fertilizers, was determined, and the effect of this crop during its establishment on soil fertility was evaluated. The research was carried out in the Institute of Animal Science. A random block design was used, with four replications and seven treatments, in a red ferrallitic soil. The fertilization treatments were: T1 = control; T2 = bovine manure (25 t ha<sup>-1</sup>); T3 = bovine manure (25 t ha<sup>-1</sup>) + Fitomas E; T4 = bovine manure (25 t ha<sup>-1</sup>) + EcoMic; T5 = bovine manure (25 t ha<sup>-1</sup>) + EcoMic® + Fitomas E; T6 = NPK (60:90:160; 0.6 t ha<sup>-1</sup>) and T7 = NPK (60:90:160; 0.3 t ha<sup>-1</sup>) + EcoMic + Fitomas E. Height, number of branches, number of leaves, stem diameter, population and yield of DM ha<sup>-1</sup> of plants were determined. In the soil, the contents of N, P, Ca, Mg, pH and OM were evaluated. The Supergenius accession under edaphoclimatic and handling conditions, similar to those of the research, should reach a height of 1 m and 35 leaves per branch at 60 days, approximately. Treatment T4 showed the best biomass production (6.61 t DM ha<sup>-1</sup>) and the highest contribution to the improvement of nutrient contents of soil (P: 136.56 p.p.m.; Ca: 1.89%; Mg: 0.38% and OM: 4.84%). It can be concluded that the application of 25 t ha<sup>-1</sup> of bovine manure and EcoMic showed the highest yield of moringa and the best contribution to nutrient contents of soil, while the application of compost, with bio-fertilizers or without them, had better effect on soil fertility than the application of inorganic fertilizer, alone or combined, because it influenced positively on all the nutrients of soil. It is recommended to continue research related to the application of fertilizers on moringa sowing, as well as other related to the effect of this plant on soil fertility, with different times of their exploitation and for other accessions.

Key words: *productivity, forage, biomass, soil fertility, fertilization*

The production of forages with high biomass productivity per surface unit and appropriate quality for animal intake is essential to achieve sustainability of husbandry. Therefore, recent studies have evaluated different promising tropical species, *Moringa oleifera* among them. This plant can be cultivated as green manure, for human consumption or for cattle. According to Pérez *et al.* (2010) and Bonal *et al.* (2014), it can be cultivated intensively, with yields between 78 and 259 t GM ha<sup>-1</sup>, when densities between 1 and 16 million plants ha<sup>-1</sup> were used and it was cut every 45 days. Reyes (2004) assured that it provides a high amount of nutrients but, consequently, their extractions to the soil are also high.

Under the edaphoclimatic conditions of Cuba, there has not been information about studies that indicate the performance of *Moringa oleifera*, its contributions or extractions, neither its response to different fertilizations. That is why the objectives of this research were to determine the performance of some morphological and productive indicators of *Moringa oleifera* related to the application of compost, chemical fertilization and bio-fertilizer, as well as to evaluate the effect of this crop in the soil fertility.

### Materials and Methods

The research was carried out during the dry season of 2012, in the "Estación Experimental Miguel Sistachs Naya", of the Institute of Animal Science, located in San José de las Lajas, Mayabeque province, between 22° 53' N and 82° 02' W and at 80 m o.s.l. (Anon 1989).

The soil was prepared by the conventional method: plowing and crossing with alternating harrows. A tractor 60 Hp Belarus was used, having a plow ADIS 3 and a mean harrow of 600 kg. The sections were labeled with rotobactor. The density of 1,000,000 plants ha<sup>-1</sup> was used, so plants were sowed at the distance of 10 x 10 cm, which was signaled with a wooden marker calibrated at this distance. Gamic seeds of *Moringa oleifera* of the Supergenius accession from India were used and two seeds were sown with a dibble. Each section had an area of 5 m<sup>2</sup> and the distance between sections was 2 m. The plants for the experiment were sown on March 20th, 2012 and the establishment cut and evaluation of indicators was carried out at 60 days.

A random block design was used, with four replications and seven treatments for a total of 28 plots in a typical red ferrallitic soil (Hernández *et al.* 1999). The treatments were: T1 = control, T2 = bovine manure (25t ha<sup>-1</sup>), T3 = bovine manure (25t ha<sup>-1</sup>) + Fitomas E, T4 = bovine manure (25t ha<sup>-1</sup>) + EcoMic, T5 = bovine manure (25t ha<sup>-1</sup>) + EcoMic + FitomasE; T6 = NPK (60:90:160; 0.6 t ha<sup>-1</sup>) and T7 = NPK (60:90:160; 0.3 t ha<sup>-1</sup>) + EcoMic + Fitomas E.

EcoMic was applied at a rate of 10% of seed weight, so 12 g of EcoMic were used in each replication, to inoculate 120g of gamic seed. This is a bio-fertilizer that allows to increase the capture of atmospheric nitrogen and include it to the inoculate plants. This way nutrition is favored and increases productivity. It contains arbuscular mycorrhizal fungi (AMF), with a high degree of purity and biological stability, and they

live in symbiosis with the roots of higher plants.

Fitomas E was applied in the foliar area, at 20 and 40 days after sowing, in doses of 500 mL ha<sup>-1</sup>. This is a natural bio-fertilizer, derived from sugar industry, with an anti-stress effect that helps to overcome the problems caused by nutritional deficit, climatic changes, plagues and phyto-toxicity produced by agrochemical substances, among other effects. Besides, it stimulates growth and development of roots, stems and leaves, and favors the development of nutrition and reduction of the crop cycle.

The evaluated indicators in plants were: height (m), number of branches, number of leaves, stem diameter (mm), plant population m<sup>-2</sup> and yield of DM ha<sup>-1</sup>. To determine height, number of branches and leaves, and stem diameter, ten plants were measured by section. The density of plants was determined when taking two samples in a linear meter each one, corresponding to two furrows of the section and the border effect was not considered. The yield was calculated by means of the cut of the whole section without the border effect. It was cut at 20 cm height. The area remained free of weeds using manual cleaning and irrigation was carried out (200 m<sup>3</sup> ha<sup>-1</sup>) every three days during the first month of cultivation and 300 m<sup>3</sup> ha<sup>-1</sup> every nine days until the moment of the cut.

In the soil, the initial evaluation of its fertility was carried out, previous to the application of treatments, by taking eight compound samples per ha with the helicoidal drill. After carrying out the cut at 60 days, two compound samples per plot were taken at random with helicoidal drill. The soil was air dried, milled and sieved (0.2 mm), to take 50 g for the chemical analysis. The pH (potenciometric), organic matter (Walkley and

Black, cited by Jackson 1970), nitrogen (AOAC 1995), phosphorus (Oniani 1964), calcium and magnesium (Maslova, cited by Paneque 1965) were quantified.

For the statistical analysis, the statistical package InfoStat (2008) was used. The theoretical suppositions of analysis of variance and variance homogeneity were analyzed by the test of Levene (1960) and normality of errors by the test of Shapiro and Wilk (1965) for the counting variables. The variables number of branches and number of leaves did not fulfilled the suppositions, so they were transformed according to  $\sqrt{x}$ . In the case of density, the transformation was not necessary, because the suppositions for the original variable were fulfilled, and therefore, an analysis of variance according to a random block design was carried out. The analysis of variance of the evaluated indicators was carried out and the test of Duncan (1955) was applied.

### Results and Discussion

Table 1 shows the performance of some morphological and productive indicators of *Moringa oleifera*. The height and the quantity of leaves per branches did not have differences among treatments. The results indicated that the Supergenius accession, under edaphoclimatic and handling conditions, similar to those of the research, should reach a height of 1m and 35 leaves per branch after 60 d, approximately.

Petit *et al.* (2010), when studying the performance of moringa, alone or associated, founded similar results to those referred in this study, concerning height of plants. However, Padilla *et al.* (2014), when evaluating the cut height in forage production of this species, found irregular performance in this indicator. Medina *et al.* (2007) obtained lower results, with values of 0.55 m of

Table 1. Behavior of the production *Moringa oleifera* biomass, with the use of different fertilizer and biofertilizer

Treatment	Height, m	Branch/ plant	Leaf/ branch	Diameter of the stem, mm	Population, plants m <sup>-2</sup>	Performance, t DN ha <sup>-1</sup>
T1	1.01	2.57 <sup>a</sup> (6.60)	5.67 (32.45)	25.52 <sup>a</sup>	60.0 <sup>a</sup>	5.77 <sup>a</sup>
T2	1.02	2.62 <sup>a</sup> (6.85)	5.92 (35.10)	27.42 <sup>a</sup>	65.0 <sup>a</sup>	5.51 <sup>a</sup>
T3	1.05	2.56 <sup>a</sup> (6.55)	6.12 (37.70)	21.98 <sup>a</sup>	63.8 <sup>a</sup>	5.52 <sup>a</sup>
T4	1.04	2.76 <sup>c</sup> (7.55)	5.90 (34.80)	31.01 <sup>c</sup>	92.0 <sup>c</sup>	6.61 <sup>c</sup>
T5	0.98	2.52 <sup>a</sup> (6.85)	5.72 (32.80)	26.36 <sup>a</sup>	82.5 <sup>b</sup>	5.57 <sup>a</sup>
T6	1.00	2.65 <sup>b</sup> (7.00)	6.14 (37.85)	29.65 <sup>b</sup>	80.0 <sup>b</sup>	6.28 <sup>b</sup>
T7	1.05	2.63 <sup>b</sup> (6.93)	5.98 (35.80)	32.22 <sup>c</sup>	82.5 <sup>b</sup>	6.13 <sup>b</sup>
SE ± and Sign	3.28	0.03 *	0.17	0.21*	0.52***	0.01*

( ) Original means; data transformed according to  $\sqrt{x}$

<sup>a,b,c,d</sup> Values with common letters do not differ at P<0.05 (Duncan, 1955).

\* P<0.05, \*\*\* P<0.001

height and only 16 leaves per branch, after 60 days of culture, in a nursery.

The number of branches varied among treatments. T1, T2, T3 and T5 had the lowest values, with figures between 6.55 and 6.85 branches per plant, followed by T6 and T7 (7.0 and 6.93 branches per plant). T4 had the highest value, with 7.55 branches per plant. In the case of stem diameter, T4 and T7 had the highest values, without significant differences among them.

As for the number of branches, the values obtained were lower to those informed by Medina *et al.* (2007), who, with same cultivation time but different density, obtained between 10 and 12 branches per plant, which indicated the effect that could have had the density used in this indicator.

The population of plants decreased from 100 sowed plants,  $m^{-2}$ , to values between 92.0 and 60.0 plants  $m^{-2}$ . The lower losses were in T4, and the highest in T1, which reaffirmed the superiority of T4 regarding the remainder treatments used in this study. Treatments T5, T6 and T7 followed T4, according to this indicator.

The generalized decrease of population could also be related to the attack of *Atta insularis* (leaf-cutting ants) to the small plantlets, which produced defoliations leading to the death of many of them. Reyes (2004) agrees that high densities create competition among plants for nutrients, light and vital space. This causes plantlet losses that can go from 20 to 30% per cut, and it produces great decreases of productive material per area, which could also be decisive in the results reached in this study.

In the case of yield per surface unit, significant differences were obtained among treatments. Treatment T4 also had the best results, followed by T6 and T7. Pérez *et al.* (2010) assured that this plant can reach yield values of 8.3 t DMha<sup>-1</sup>, with cuts every 45 days, which is superior to the results of this research. Castillo *et al.* (2013), when evaluating the influence of density (10,000; 20,000 and 40,000 plants ha<sup>-1</sup>) and cut frequency (60 and 90 days), obtained yields between 9.99 and 10.83 t ha<sup>-1</sup>. Although these are studies with different densities, they evidenced that moringa can reach superior yield values, according to the used density, cut frequency and edaphoclimatic characteristics were it develops.

The results of productive and morphological indicators of moringa showed that T4 obtained the best performance, followed by T6 and T7. It is significant to state that these last treatments included the application of inorganic NPK fertilizers, alone and combined with bio-fertilizers, respectively. This showed the productive, ecological and economical value that can have the alternative of combining compost with bio-fertilizers, like in T4. Likewise, this evidenced the effectiveness of the use of combinations of mineral fertilizer with bio-fertilizers, since there were no differences in the yields of T6 and T7, and suggested that when reducing the dose of the first, similar productive results to those when optimal

doses are used, can be expected. It can lead to high yields per surface unit, with possible decrease of expenditures and being also friendlier with the environment.

The results pointed out that the Supergenius accession varies its performance under our edaphoclimatic conditions, due to the decrease of its yields and other growth and development parameters: number of branches, leaves per branches and height, compared to the information given by Foidl *et al.* (2001) in Central America.

The performance of the agrochemical indicators is shown in table 2. The total nitrogen (Nt) did not have differences in the initial sampling, previous to the application of the fertilization treatments and the treatment control. In spite of it, this indicator, according to the interpretation tables (Crespo *et al.* 2006), went to a mean value (1.18%) in T0, to a low value (1.15%) in T1. This indicated that moringa, in the first 60 days of sowing, varied the content of this nutrient in the soil, and suggested that this plant can carry out high extractions that, without the application of fertilizers, they are possibly able to contribute to the gradual deterioration of the Nt tenor.

This macronutrient showed an increase ( $P < 0.001$ ) in the remainder treatments, regarding the control, which demonstrated the effectiveness of the used fertilizations that allowed to compensate the extractions and improve the stability of tN in the soil. Chicowo *et al.* (2006) and Salazar-Sosa *et al.* (2003) explained that the minimum farming and the application of organic fertilizers contribute to the increase of residues and, consequently, to the increase of organic matter, which can be reflected on the highest concentration of tN in the soil.

The P did not have significant differences between its initial value and the treatment control at 60 days of moringa sowing. Meanwhile, their highest values were reached in the treatments where EcoMic was applied (T4, T5 and T7). The combination of bovine manure with this biofertilizer (T4 and T5) produced similar result in the phosphorus content in the soil to the results when combining it with inorganic fertilizer.

The performance of P in the treatments where EcoMic was present, showed the contributions of this biofertilizer to the dissolution of P in the soil, due to the action of mycorrhizae (Martin 2009 and Pentón *et al.* 2013). In case of P, besides the incidence of the biofertilizer, another cause for its improvement could be in the increase of organic matter content, although T2 and T3 also showed increase of the organic matter, but the content of P did not increased. Moron (1994) and Sileshi and Mafongoya (2007) assure that P concentrates, especially, in the organic matter, which contains high levels of P, and that the sowing of enhanced pastures of grasses in previously sowed soils, usually determines an increase in the balance of organic matter and, consequently, increases the content of organic P.

Calcium did not have differences between the initial

Table 2. Agrochemical characteristics of the soil, previous to the application of the treatment and 60 days of crop, for each treatment

Treatment	N (%)	P (mg 100g <sup>-1</sup> )	Ca (cmolckg <sup>-1</sup> )	Mg (cmolckg <sup>-1</sup> )	pH (KCl)	MO (%)
Inicio						
T0	1.18 <sup>a</sup>	7.53 <sup>a</sup>	8.10 <sup>b</sup>	1.81 <sup>a</sup>	5.80 <sup>b</sup>	3.44 <sup>b</sup>
60 días de cultivo						
T1	0.15 <sup>a</sup>	6.65 <sup>a</sup>	6.55 <sup>a</sup>	1.73 <sup>a</sup>	5.35 <sup>a</sup>	2.91 <sup>a</sup>
T2	0.27 <sup>b</sup>	10.39 <sup>c</sup>	8.95 <sup>b</sup>	3.22 <sup>bc</sup>	5.58 <sup>b</sup>	4.36 <sup>bc</sup>
T3	0.23 <sup>b</sup>	10.60 <sup>c</sup>	8.90 <sup>b</sup>	3.05 <sup>bc</sup>	5.55 <sup>b</sup>	4.49 <sup>c</sup>
T4	0.26 <sup>b</sup>	13.65 <sup>d</sup>	9.45 <sup>b</sup>	3.14 <sup>bc</sup>	5.88 <sup>c</sup>	4.84 <sup>c</sup>
T5	0.23 <sup>b</sup>	12.89 <sup>d</sup>	9.60 <sup>b</sup>	3.72 <sup>c</sup>	5.90 <sup>c</sup>	4.63 <sup>c</sup>
T6	0.25 <sup>b</sup>	9.18 <sup>b</sup>	6.05 <sup>a</sup>	2.06 <sup>a</sup>	5.38 <sup>a</sup>	2.77 <sup>a</sup>
T7	0.25 <sup>b</sup>	12.59 <sup>d</sup>	6.00 <sup>a</sup>	2.39 <sup>a</sup>	5.35 <sup>a</sup>	2.80 <sup>a</sup>
SE ± and Sign	0.01***	2.03***	0.04***	0.02***	0.06***	0.09***

T0: sampling previous to the application of treatments. <sup>a,b,c,d</sup> Values with common letters do not differ at P<0.05 (Duncan, 1955). \*\*\* P<0.001

treatment (T0) and T2, T3, T4 and T5, but it decreased between T0 and T1 ( $P < 0.001$ ). This reveals that the crop should carry out high absorption of this nutrient in the first 60 days of forage production, and suggests the necessity of fertilization to achieve stability of this macronutrient. The initial content of Mg did not differ to the content after 60 days for T1, T6 and T7, while it showed an increase for those treatments that used bovine manure.

The pH also had a decrease ( $P < 0.05$ ) from the initial treatment to T1, and showed variances among treatments that, apparently, were determined by the combination of fertilizers applied. This way, T2 and T3 had similar performance that increased the pH regarding the control. After them, with high values, came T4 and T5, while the combination of NPK (60:90:160), alone or with biofertilizer (T6 and T7), had similar performance to T1.

The contents of calcium and magnesium suggest, in general, that the application of bovine manure can improve their contents and counteract the possible absorption of these nutrients by moringa. The performance of these macronutrients was related to the pH.

Cairo and Fundora (1994) stated that there is a directly proportional relationship between the contents of the alkali earth cations ( $\text{Ca}^{2+}$  and  $\text{Mg}^{2+}$ ) and the value of pH, in a way that, when the firsts increase or decrease, the pH also increases or decreases, respectively. Lok *et al.* (2003) found similar performance to the obtained in this study and stated that the pH increased due to the increase of the contents of Ca and Mg, due to the accumulation and quality of legume litter, which had an important contribution to the organic matter in the soil.

Likewise, the increase of organic matter ( $P < 0.01$ ) in the soil of the treatments T2, T3, T4 and T5, could also be one of the causes of the performance found for N, P, Ca and Mg.

The results indicated that the fertilizer application

contributes positively to the contents of soil nutrients. Specifically, N and P can be improved starting from any of the combinations of them, but Ca, Mg and pH depend on the content of the existing OM. In that sense, the organic fertilizers can have the best effect. Besides, when the NPK (60:90:160) was combined with biofertilizer, and the dose of the first was diminished to the half, the performance of the soil nutrients was similar to the results obtained when the inorganic fertilizer was applied, but with the entire dose. Due to the high prices of chemical fertilizers in the international market, and the noxious effects that can have their indiscriminate use on the quality and health of the soil (Rincón and Muñoz 2005, Nicholls and Altieri 2006 and Altieri and Nicholls 2008), they are applied in lower quantities, which suggested that the combination of both types of fertilizers favors, economical and environmentally, the development of the system.

It can be concluded that the application of different manures and biofertilizers contributes to the performance of morphological and productive indicators of *Moringa oleifera* for the biomass production during the establishment, although their yields and indicators of growth and development were below those informed for the Supergenius accession, under tropical conditions. This species seems to decrease the contents of soil nutrients when strategic fertilizations are not applied. The combination of 25 t ha<sup>-1</sup> of bovine manure and EcoMic showed the highest yield of moringa and the best contribution to the contents of soil nutrients in this stage. The application of organic manure, with or without biofertilizers, had better effect on the soil fertility than when applying the inorganic fertilizer, alone or combined, because it influenced positively on all the soil nutrients.

Further studies on the application of fertilizers the moringa are recommended, as well as others that relate



the effect of this plant in the soil fertility with different times of exploitation and for other accessions.

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