

Response to bocashi and vermicompost of *Moringa oleifera* Lam. after pruning

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Abstract

Moringa oleifera Lam. due to its multiple uses, it has an enormous potential as an alternative for sustainable cultivation to be implemented in the central area of the state of Veracruz. The present work evaluates the vegetative and reproductive response of the species to organic fertilization after pruning in this region. The experimental design was completely randomized with three-year-old moringa trees, pruned only once at a height of 1.5 m, to which two organic fertilizers were applied: vermicompost and bokashi, and one control. The fertilizers were applied at the beginning of the experiment and later every four months. The variables: 1) number of new branches; 2) survival of branches; 3) length of branches; 4) thickness of branches; 5) number of inflorescences per plant; 6) number of buttons per inflorescence; 7) flowers per inflorescence; 8) fruits per inflorescence; 9) number of fruits per plant; 10) length of fruits; 11) number of seeds per fruit; and 12) seed dimension, were measured every month for 10 months. Treatments were evaluated by one-way analysis of variance ($p < 0.05$). The results showed that the application of organic fertilizers did not influence variables 3, 4, 5, 6, 7 and 8 as there were no significant statistical differences in these ones; except for the total number of fruits per plant, where plants with bocashi achieved the highest productivity. It is concluded that the best fertilizer for fruit production was bocashi, while with vermicompost no significant differences were observed.

Keywords: *Moringa oleifera* Lam., organic fertilizers, vegetative response.

Reception date: January 2019

Acceptance date: March 2019

Introduction

Moringa (*Moringa oleifera* Lam.) is a tree native to northwestern India (Zayed, 2012), the species arrived at Mexico probably by Filipino ships traveling from Manila, Philippines to Acapulco, Guerrero (Olson and Fahey, 2011). Moringa is very appreciated for its nutritional and medicinal characteristics, present in most of the plant (leaves, flowers and fruits, all edible). The versatility of environmental conditions to which moringa plant adapts, including tropical zones of low soil humidity high temperatures, high evaporation and large variations in precipitation (Alves *et al.*, 2010), turn it into a species with great potential to grow in different areas of our country.

Although this plant, in particular, develops better under 500 masl and not above 1 500 masl (Olson and Fahey, 2011). Some authors indicate that Moringa requires a minimum of 250 mm of annual rainfall, and it can tolerate clay and sandy soils (Valdes-Rodríguez *et al.*, 2014), but poorly drained soils impair its growth (Velázquez *et al.*, 2016). With respect to their nutritional requirements, there are studies related to organic fertilization on moringa, which show their positive impact on the production of foliar biomass mainly for human and animal consumption (cattle) (Pérez *et al.*, 2010; Suárez, 2014).

Organic fertilization is important to establish a sustainable crop or with low environmental impacts. An organic fertilizer originates through the aerobic and anaerobic decomposition of organic waste mainly of animal and vegetable origin. This action occurs through microorganisms that exist in the waste itself, under controlled conditions, which produces a partially stable material of slow decomposition under favorable conditions (Restrepo, 2001).

Two organic fertilizers widely known in Mexico and much of Latin America are bocashi and vermicompost, their simple way of processing them and low production cost, make them a viable option for farmers who use them. The word bocashi comes from the Japanese language and, in the case of fermented organic fertilizers, means to pre-steam the organic materials of the fertilizer through the heat generated by the decomposition (Restrepo and Hensel, 2009). Likewise, vermicompost is a product resulting from microbial transformation of organic waste in controlled environments and is obtained when several species of earthworms participate in its elaboration (Olivares *et al.*, 2012).

The positive impact of organic fertilizers in degraded environments is an alternative that is important to promote and increase, since with its application in the soil the growth of the plants is stimulated, the fertility, nutrition and the structure of the soil is gradually improved, as well the water retention capacity and the activity of microorganisms are increased, which contributes to reduce the loss of nutrients by leaching (Restrepo and Hensel, 2009). Therefore, it is necessary to promote the application of organic fertilizers in places and crops with productive potential, as is the case of moringa, and thus avoid the application of agrochemicals, which are one of the main causes of degradation and pollution of natural resources and biodiversity (Altieri, 1999; Ramos and Terry, 2014).

Currently there are few studies on moringa reported in the state of Veracruz. One of them was performed by Valdes *et al.* (2014) and is focused on the registration of moringa allometric measurements (growth, flowering and fruit production) without receiving organic fertilization. Another work done by Quintas *et al.* (2016) analyzed the vegetative response of moringa to two organic fertilizers. However, this last one only comprised a period of seven months and did not address the response of such fertilizers over the reproductive variables. Therefore, an evaluation of the productive variables would allow estimating the effects of organic fertilizers on the productivity of moringa plants, in order to determine if their application represents an efficient and profitable agronomic practice for this crop.

The objective of this research was to evaluate the use of organic fertilizers (vermicompost and bocashi) on the vegetative and reproductive response of moringa after pruning, in the central region of the state of Veracruz.

Materials and methods

Study area

This research was conducted at the Postgraduate College (COLPOS), Campus Veracruz, which is located in the municipality of Manlio Fabio Altamirano, in the central area of the state of Veracruz. This municipality is located at the coordinates 19°18' North latitude and 96° 32' West longitude. It has an altitude of 25 m and its soil type is loam-clay-sandy. The climate is warm with rains in summer and winter; the average annual rainfall is 1 239.5 mm and the average annual temperature is 25 °C (Cruz *et al.*, 2015).

Experimental procedure

Three-year-old moringa plants, planted at a density of 1 m by 1 m, with an average height of 6 m were evaluated. 24 individuals were randomly selected to apply the treatments, with eight individuals per block. The first treatment consisted in the application of vermicompost made with 30% coffee pulp and 70% sugarcane bagasse. The second treatment consisted in the application of bocashi, made with cattle manure (50 kg), maize stubble (25 kg), litter (25 kg), ash (10 kg), wheat bran (25 kg), molasses (4 L), bread yeast (250 gr) and soil of the region (50 kg) and finally the control, to which no fertilizer was applied.

In order to standardize the size of the plants and facilitate the taking of measurements, a single pruning was performed one day before starting the treatments, the trees were in a period of vegetative growth without evidence of floral clusters and fruits. The pruning consisted in the cutting of the main trunk at a height of 1.5 m and the elimination of the lateral branches and sprouts of the plant, leaving only the main trunk. The next day the first application of organic fertilizers was made. Three applications of four kilograms per plant were made every four months distributed during the ten months that the work lasted.

The fertilizer was applied in a radius of 40 cm of the main trunk of the plant, it was buried 10 cm and later it was covered with the soil removed from the area. After the application of the organic fertilizer a single irrigation was practiced using 19 L of water to irrigate each plant. Between the application periods no irrigation was done and weed control was done manually.

Additionally, soil sampling of the experimental site was carried out as follows: 15 sub-samples of 500 g were randomly collected, each one at a depth of 10-15 cm from the soil, the samples were combined in a homogeneous manner and a 500 g portion was subsequently taken. Likewise, for organic fertilizers (vermicompost and bocashi) a representative sample of (500 g) was taken. The soil samples of the region and the organic fertilizers were sent for analysis to the soil laboratory of the Institute of Ecology, AC (INECOL).

The methodology used for the sample of soil and organic fertilizers was the one indicated by NOM-021SEMARNAT-2000. The variables evaluated were divided into A) vegetative, which were analyzed weekly during four months: 1) number of new branches (branches); 2) survival of branches; 3) length of branches; and 4) branch thickness; and B) reproductive: 5) number of inflorescences per plant; 6) number of buttons per inflorescence; 7) number of flowers per inflorescence; and 8) number of fruits per inflorescence; 9) number of fruits per plant; 10) length of fruits; 11) number of seeds per fruit; and 12) seed dimension. These last variables were evaluated during a period of ten months starting from March to December 2016.

Vegetative variables

The number of sprouts and their survival per plant was determined by means of the total accounting of these on their initial registration and was carried out weekly during four months. The measurement that corresponds to the variables of length and thickness of the sprouts was taken with the help of a flexometer (1 mm precision) and a digital vernier (0.01 mm precision) respectively, for which six sprouts were selected per plant, because this was the minimum number of sprouts surviving per plant the first two months after pruning. During the first five months, these two variables were evaluated weekly; however, when observing that moringa was growing slowly measurements were made monthly from the sixth month onwards.

Reproductive variables

The total number of floral clusters and fruits per plant was counted weekly. Of the total floral clusters 10 were selected for each plant to be monitored weekly. From each floral cluster selected, the number of buttons, flowers and fruits was counted. Also, the fruits were measured in length, using a flexometer (1 mm) to measure each pod from one end to the other. To determine the total number of seeds per pod all the fruits of the harvest were opened, and all the seeds were extracted in order to be counted, later they were measured in length and width with a digital vernier (0.01 mm precision).

Statistical analysis

For each variable, the corresponding descriptive statistics were obtained (average, standard deviation, maximum, minimum and coefficient of variation). The analysis of each variable between treatments was carried out using a one-way analysis of variance test with a level of significance of 5%. The post-hoc tests were performed using the Tukey method, both analyzes supported by the SigmaPlot 10.0 software.

Results and discussion

The results of the physical-chemical analysis of the soil of the region and the two organic fertilizers showed a remarkable difference in three P, N and Mg macronutrients, with the vermicompost obtaining the highest magnitudes. According to González-González and Crespo-López (2016), percentages of N from 1.5% in organic fertilizers favor the production of foliar biomass up to 100%. Thus, this vermicompost turned out to be the fertilizer with the greatest potential for foliage production. While Sarwar *et al.* (2017) established that doses less than 1% of N-P-K can increase moringa stems and foliage by 50% or more in a loamy soil without fertilization.

Therefore, the doses found in both vermicompost and bocashi had the ability to increase the growth of plants above the control. Additionally, it is considered that the pH of bocashi would tend to improve the alkalinity of the soil of the site, which was found to be acidic (Table 1).

Table 1. Physical and chemical composition of the soil of the region and the organic fertilizers used.

Component	Control	Bocashi	Vermicompost
pH	5.63	8.23	5.12
P available (mg kg ⁻¹)	7	26	292
P total (mg kg ⁻¹)	254	1867	2788
N (%)	0.17	0.95	2.79
K (cmol kg ⁻¹)	0.41	18.68	9.76
MO (%)	3.03	17.08	41.51
C/N	12	16	10
Ca (cmol kg ⁻¹)	10.28	34.47	31.28
Mg (cmol kg ⁻¹)	4.93	14.5	18.74
Fe (mg kg ⁻¹)	32.3	149.5	342.8
Mn (mg kg)	23.7	23.6	61.8
Cu (mg kg ⁻¹)	0.31	2.5	6.2
Zn (mg kg ⁻¹)	0.88	17.84	19.87
Carbon organic (%)	1.76	9.91	24.08
Density (g cm ⁻³)	1.51	0.98	0.68
Clay (%)	51.6	21.6	27.6
Slime (%)	26.4	33.6	19.6
Sand (%)	22	44.7	52.7

While the high levels of Fe in bocashi and vermicompost may favor the synthesis of chlorophyll in moringa plants. Likewise, the high contents of Mn will favor the photosynthesis and assimilation of nitrogen (Azcon and Talon, 2013).

Vegetative variables

After pruning and at the end of a period of 16 weeks it was observed that the treatment with bocashi had an average of 12.87 sprouts per plant, vermicompost 8.87 and control 13.12. In this variable, no statistical differences were found between treatments ($p= 0.079$). This result could be due that trunk diameters of the subjects fertilized with vermicompost were 18% lower than those of the control, while in bocashi they were 15% lower than the control, which is related to the increase in the number of sprouts, which was proportional to the diameter of the stems.

In this regard, Medina *et al.* (2007) obtained an average of 11 to 12 branches per plant without the application of any type of fertilizer, which is similar to what was obtained in this experiment. The percentage of surviving branches per plant was 73% for bocashi, 66% for vermicompost and 77% for control and no significant statistical differences were found between treatments ($p= 0.867$). No statistically significant differences were found between treatments in the length of branches ($p= 0.174$) and thickness of branches ($p= 0.514$). While the average number of surviving branches per plant until day 283 of monitoring was 3.87, a result lower than that found by Swati *et al.* (2013), which was of eight branches with application of organic fertilization.

The results obtained indicate that the low survival of the branches could be due to the distance in which the moringa plants were planted, which was only 1 m between each of them, being a factor that could negatively affect this variable, the main cause observed was the fall of the branches due to wilting. Previous research mentions that the height of the plants and the number of branches is related to the distance of planting, being increased if it is greater between plant and plant (Sosa *et al.*, 2016), thus reaching higher yields according to the density in which the plants are planted (Lok and Suárez, 2014). From the previous results it is deduced that vegetative response to the treatments was not significant for a period of 10 months.

Therefore, if moringa is to be cultivated under similar conditions in a similar period of time, it is not possible to recommend the application of these fertilizers in the quantities applied in this experiment for a greater production of biomass. This is explained because both vermicompost and bocashi are slow release fertilizers and require periods of up to one to three years of constant application to observe results and effectiveness (Restrepo and Hensel, 2006; Ramos and Terry, 2014); this way, with this characteristic they could contribute substantially to the improvement of the biological activity of the soil and therefore of the plant (Usman *et al.*, 2003), but in a period of time of at least two years.

However, the longest branches were obtained with vermicompost, being 13% longer than the control and 18% longer than with bocashi; in addition, in the thickness of the branches, vermicompost achieved 14% and 16% more thickness than the control and bocashi, respectively (Figure 1 and 2).

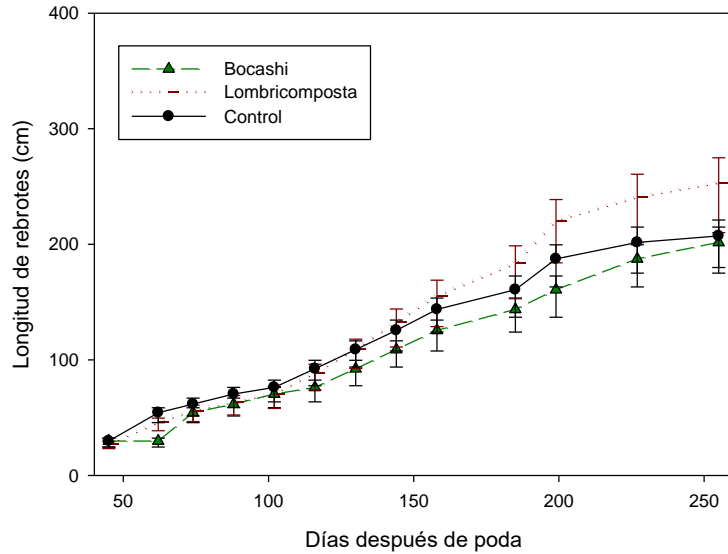


Figure 1. Growth of length of sprouts of *M. oleifera* by treatment. The sidebars represent the standard error of the mean.

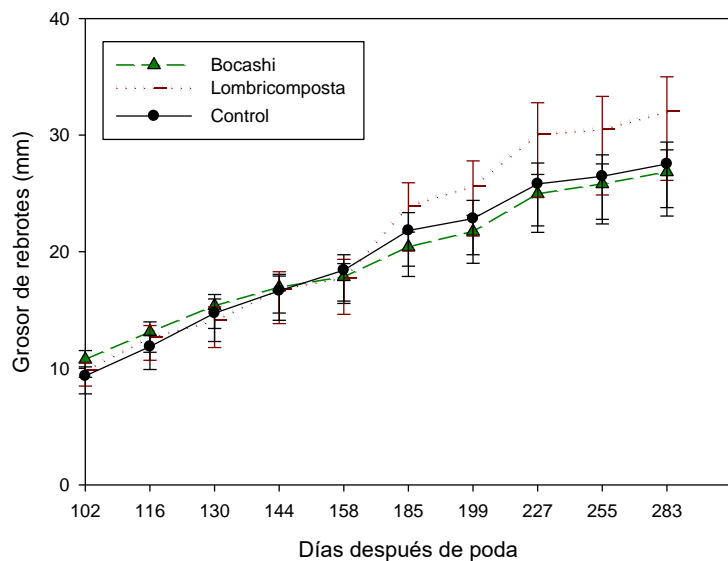


Figure 2. Growth of thickness of *M. oleifera* by treatment. The sidebars represent the standard error of the mean.

This behavior, although not yet significant, could be due to the fact that organic fertilizers indicated a higher percentage of Nitrogen (N) in vermicompost, which was 66% higher than bocashi and 94% higher than control. In this regard, it is known that N is the nutrient with greater importance for the development of the plant, along with P and potassium (K) (Azcon and Talon, 2013).

Reproductive variables

There were no significant statistical differences in the number of floral clusters per plant ($p=0.183$), number of buttons per floral cluster ($p=0.511$), flowers per floral cluster ($p=0.605$) and fruits per floral cluster ($p=0.664$). However, bocashi was the only treatment that

consolidated mature fruits to be harvested (Table 2) and the total number of fruits per plant ($p=0.037$) was the only variable that had significant statistical difference, with the treatment with bocashi giving the greatest number.

Table 2. Average values and standard deviation (dev) of the variables evaluated in the different treatments applied to *Moringa oleifera* plants.

Variable	Control		Vermicompost		Bocashi				
	Average	±desv	Average	±desv	Average	±desv			
Length of new branches	218.34	±96.93	a	253.28	±108	a	207.28	±77.41	a
Thickness of branches	27.53	±10.59	a	32.04	±15.1	a	26.84	±10.69	a
Total number of inflorescences per plant	14.43	±15.47	a	8.73	±9.99	a	14.3	±18.21	a
Buttons by inflorescence	18.21	±33.85	a	7.71	±19.48	a	16.12	±32.08	a
Flowers per inflorescence	3.1	±6.99	a	1.61	±6.23	a	2.64	±7.26	a
Fruits per inflorescence	0.46	±1.4	a	0.61	±1.3	a	0.85	±1.56	a
Total number of fruits per plant	1.89	±3.07	ab	0.94	±2.18	b	3.32	±7.35	a

Different letters in the columns represent statistical differences between treatments ($p < 0.05$).

This result could be due to the fact that bocashi has microorganisms capable of solubilizing certain minerals such as Ca and phosphates, the latter being determinants for flower growth and fruit ripening (Rodríguez *et al.*, 2005).

It is also possible that the difficulty to detect significant differences in the evaluated parameters was due to the enormous variability found in the response of all the plants, with high coefficients of variation (Table 3), where the climatological conditions and the intrinsic variability of the plants could influenced these results (Table 3) (Toral *et al.*, 2013).

Table 3. Maximum and minimum values of the variables evaluated in the different treatments applied to *Moringa oleifera* plants.

Variable	Control		Vermicompost		Bocashi	
	Max-min	CV	Max-min	CV	Max-min	CV
Length of new branches	385-42	0.44	455-90	0.42	397-70	0.37
Thickness of branches	52-11	0.38	66-12	0.47	55-7	0.39
Total number of inflorescences per plant	77-0	1.07	47-0	1.14	100-0	1.27
Buttons by inflorescence	134-0	1.85	113-0	2.52	156-0	1.99
Flowers per inflorescence	36-0	2.25	58-0	3.86	55-0	2.75
Fruits per inflorescence	9-0	3.04	7-0	2.11	10-0	1.84
Total number of fruits per plant	12-0	1.63	9-0	2.33	30-0	2.21

Length of the fruits

Seven months after the pruning, a single harvest was carried out, 24 fruits were obtained, all from the treatment fertilized with bocashi, since the control and the vermicompost did not present mature fruits.

These results could be due to the higher content of K and Ca, as well as silt and a higher pH present in the bocashi, which facilitated the assimilation of these nutrients strongly related to flower and fruit development (Azcon-Bieto and Talon, 2013). The average length of the moringa pod was 27.39 cm, which is within the parameters reported for this plant, ranging from 10 cm to 50 cm in length (Olson and Fahey, 2011).

Although this length of the pods was 10 cm less than that obtained by other authors who also fertilized with mixtures that included vermicompost, but for a period of one year (Swati *et al.*, 2013). Therefore, it is considered necessary to evaluate organic fertilizers for a period longer than one year in order to determine if there are positive long-term development effects, given the slow release of these biofertilizers. Additionally, it has been documented that moringa develops better in light soils, and in this site the substrate was mostly clayey, which makes the penetration of the roots difficult, since it is a heavy and acid soil, therefore nutrient uptake is slower (Pérez *et al.*, 2010).

Number of seeds

The only treatment that managed to consolidate ripe fruits was the fertilization with bocashi, from which a minimum of eight and a maximum of 23 seeds per fruit were obtained, with an average of 14.7 ± 3.3 seeds per fruit. These values were similar to that reported by Pérez *et al.* (2010), ranging from 12 to 25 seeds, and slightly higher than that found by Pérez *et al.* (2010), ranging from 10 to 20 seeds, who described the typical production of these plants. In a similar study where organic fertilization was also used in moringa plants, a maximum of 14 seeds and a minimum of seven seeds per fruit was found Swati (2013), from which it is inferred that this bocashi substantially improved the number of seeds produced in the place.

Seed measurement

The average size of the seeds was 10.85 mm long and 10.66 mm wide. Data that coincide with moringa seeds before being sown, which were 12.9 ± 1.1 mm in length and 11.4 ± 0.8 mm in width and 268.9 ± 42.9 mg in weight (Valdes *et al.*, 2014). In another work similar measures (11.34 to 15.16 mm and 9.77 to 12.19 mm) were observed (Oloyede *et al.*, 2015); thus, the application of organic fertilizers did not seem to affect the dimensions of the seeds.

Conclusions

Organic bocashi and vermicompost fertilizers applied for ten months in moringa did not show significant effects on the vegetative variables of *Moringa oleifera*. On the other hand, in the reproductive variables, differences were noted in the total number of fruits per plant when fertilized with bocashi. For this reason, the application of bocashi is recommended to improve the production of fruits of the plant. In the same way, when organic fertilizers are used in heavy soils, it is recommended to evaluate the plants for a period greater than a year, to determine if they have better effects over the vegetative variables.

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