Morphological characterization of eight Moringa oleifera *(Lam.) provenances under nursery conditions*

Odalys Toral[1], Y.Cerezo[2], J. Reino[1], and H. Santana[1]
Estación Experimental de Pastos y Forrajes Indio Hatuey,
Universidad de Matanzas Camilo Cienfuegos, Ministerio de Educación Superior
Central España Republicana, CP 44280, Matanzas, Cuba
[2]Universidad de Matanzas Camilo Cienfuegos, Cuba
E-mail: odalys.toral@ihatuey.cu

ABSTRACT

A study was conducted at the Pasture and Forage Research Station Indio Hatuey, in order to morphologically characterize eight *Moringa oleifera* (Lam.) provenances under nursery conditions. A completely randomized design was used and the data were statistically analyzed through a Simple Classification Variance analysis. The following indicators were evaluated: germination, emergence, survival, plant height, stem diameter and number of leaves. Five replications of 25 seeds per provenance were used for the germination test (21 days); and 30 bags were planted for each provenance at the nursery. The provenance Holguín-Mayarí had the best performance regarding height (17,55 cm) growth rate (0,43 cm/day), number of leaves (seven) and stem diameter (0,37 cm). Plain, Holguín-Mayarí and Paraguay were the ones with the best emergence and survival (100 %). The lowest values were shown by Guatemala (average height: 14 cm, emergence: 76 %, germination: 49 %). None of the provenances exceeded 85 % of germination. There were no lesions due to the attack by insects or diseases. It is concluded that these provenances have morphological characteristics which differentiate them from each other, and that Holguín-Mayarí showed the best performance in the evaluated indicators. To perform further morphoagronomic studies of the different *M. oleifera* provenances is recommended.

Key words: emergence, germination, *Moringa oleifera*, survival

INTRODUCTION

In the last decade, *Moringa oleifera* (Lam.) has stood out –within a group of non-leguminous trees– as a promising plant for cut and carry, grazing/browsing systems, as well as in the formation of windbreaks and living fences (Martín *et al*., 2010).

M. oleifera (Lam.) –known in Cuba as tilo americano, moringa, tila and palo de jeringa; and in other countries as marango, palo jeringa, ben, acacia and jazmín francés– is a tree up to 9 m tall. Its leaves are compound, alternate and tripinnate; they have a length of 30-70 cm and are disposed in foliole groups, with five pairs arranged on the main petiole and a foliole on the terminal part (García Roa, 2003; Comisión Técnica de Fitomed, 2010).

This plant is mainly valued for its fruits, leaves, flowers and roots –all of them edible–; as well as its oil –also edible–, which is obtained from the seeds. In addition, it is used in traditional medicine in the areas where it is native or has been introduced (Fuglie, 2001).

In Cuba, moringa is used in the agricultural context only as a living post in fences (Bosch, 2009), for which there is not enough information about its response when it is sown by seeds or seedlings. For such reason, the objective of this work was to characterize the performance of eight *M. oleifera* (Lam.) accessions under nursery conditions.

MATERIALS AND METHODS

Geographic location, soil and climate. The study was conducted at the Pasture and Forage Research Station Indio Hatuey, which is located at 20º 50' North latitude and 79º 32' West longitude, in the Perico municipality (Matanzas province, Cuba), at an altitude of 19 m.a.s.l.

The soil has plain topography and is classified as lixiviated Ferralitic Red (Hernández *et al*.,

2003). It shows moderate fertility –with 80 % of clay–, as well as good aeration conditions and a slightly acid pH (6,3). Mean annual rainfall is 1 200 mm. The average temperature is 25 ºC, with relative humidity of 60-70 % during the day and 80-90 % at night.

Treatments. The treatments were eight moringa accessions introduced from abroad or collected in the country (table 1).

Experimental procedure. To determine the germination percentage, 125 seeds per provenance were taken and five replications –of 25 seeds each– were set on Petri dishes, with moisturized filter paper. The germination test concluded 21 days after its setting up.

At the nursery, perforated polyethylene bags, 28 x 13 cm, were used, in which a substratum composed by 75 % of Ferralitic Red soil and 25 % of decomposed cattle manure (organic matter source) was deposited, for seedling planting.

Before sowing, the surface crust of the soil was removed and the top of the bag was pressed until loosening it. The seeds (one per bag), without undergoing any previous treatment, were placed in the middle of the bags.

Thirty bags were sown for each provenance and only the 20 ones in the middle were evaluated, while the other 10 were used to avoid the edge effect of the neighboring species. The material sown in the nursery received daily irrigation (from 8:30 to 10:00 a.m.), in order to preserve the necessary humidity for seedling emergence and growth.

The time passed between sowing and the transfer of the seedlings to field conditions or evaluation stage was considered as the nursery period, which ended at 40 days.

Characterization. In the characterization of the different provenances the following indicators were considered: a) germination and emergence (%), which were evaluated every three days; b) survival (%), since 10 days after planting and with a seven-day frequency; c) height (cm), which was measured every ten days since sowing; d) number of leaves, from the tenth day after sowing and every ten days; e) stem diameter (cm), every ten days, since 20 days after sowing. From the appearance of the first two leaves, and with a seven-day frequency, phytosanitary observations of insect presence and lesions (%) and the infestation by pathogen microorganisms (%) were made, according to the scale of values from I to V:

I. very high degree of affectation (100 % of the leaf area affected).
II. high degree of affectation (50 % of the leaf area affected).
III. moderate degree of affectation (25 % of the leaf area affected).
IV. slight degree of affectation (10 % of the leaf area affected).
V. Incipient degree of affectation (1 % of the leaf area affected).

Design and statistical analysis. A completely randomized design with 30 repetitions was used. For interpreting the results a simple classification variance analysis was used, and the means were compared through the multiple range test (Duncan, 1955). The statistical pack used was Startgraphics version 5.1.

RESULTS AND DISCUSSION

Figure 1 shows the results of the germination test performed on the *M. oleifera* seeds under controlled conditions. The provenances Plain and Holguín-Mayarí showed better values (84 and 80 %, respectively); while Guatemala and Paraguay, with 49 and 60 %, showed the lowest germination rates.

Table 1. Evaluated accessions.

Number	Provenance	Observations
1	PKM- 1	Introduced from India
2	Supergenius	Introduced from India
3	Plain	Introduced from India
4	Guatemala	Introduced from Guatemala
5	Criolla-Granma	Collected in Cuba
6	Paraguay	Introduced from Paraguay
7	Holguín-Mayarí	Collected in Cuba
8	Matanzas-Ciudad	Collected in Cuba

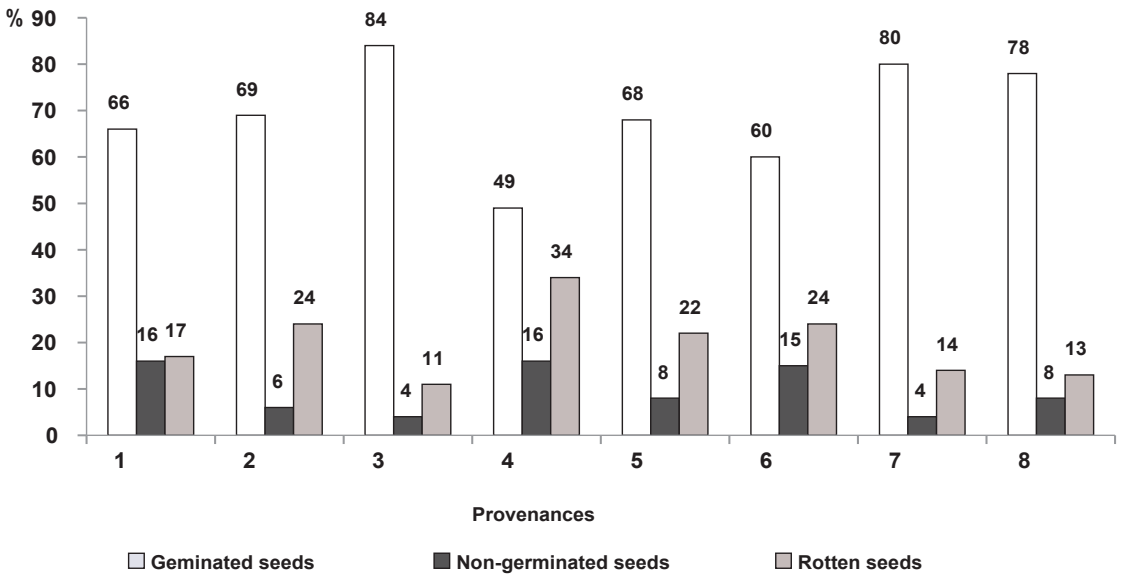


Figure 1. Germination test (%).

The germination rate reported in literature (Jahn *et al*., 1986; Nautiyal and Venhataraman, 1987) varies between 60 and 90 % for fresh seeds, and it must be stressed that most of the studied accessions are within that range. The storage time is known to influence the germination rate. For example, in India rates of 60, 48 and 7,5 % were reported for the seeds sown one, two and three months after being harvested, respectively (Sharma and Raina, 1982); although Moroto *et al*. (2000) reported lower values (between 10 and 60 %) in seeds which had been harvested a month before. The germination rate which was recorded in the accessions Guatemala and Paraguay can be related to the harvest age, because the seeds used were sown three months after their maturation.

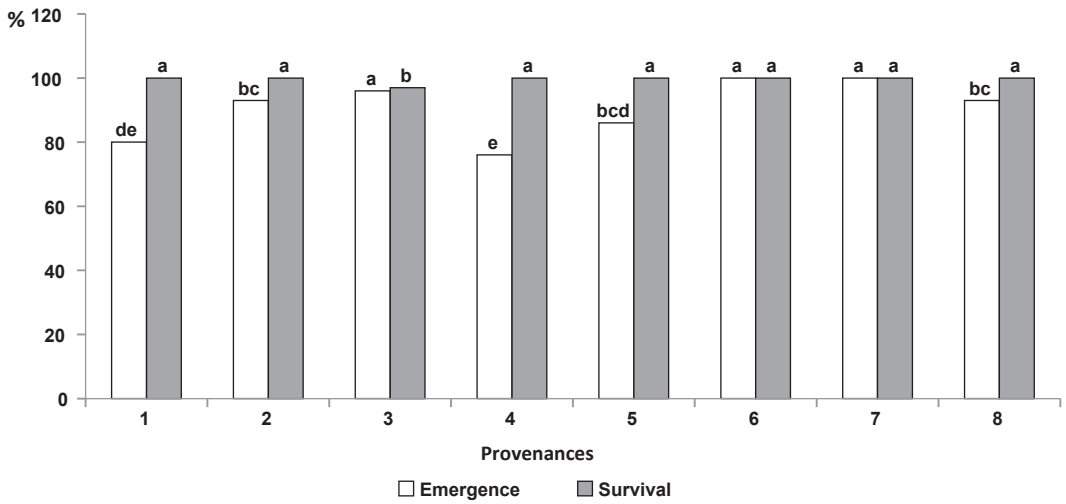
On the other hand, the seeds did not receive any pre-germination treatment, and it has been proven that these treatments help to break dormancy and, thus, increase germination rate. In this sense, González and Navarro (2001) found values of 96,8 % germination in *Albizia lebbeck*, with a cut of the seed coat; this showed that the use of this treatment favors the faster contact of embryonic structures with the environmental water and oxygen, which increases germination.

A high number of rotten seeds was observed (fig. 1), varying between 11 and 34 %, which could have been brought about by the non-ideal storage conditions that propitiated a high degree of infestation by pathogens. In this sense, Navarro (2009) reported that the loss of viability during storage can be caused by attacks of insects or fungi, which destroy the physical integrity of the seed, or due to the seed natural deterioration because of their ageing.

The emergence and survival values of moringa under nursery conditions are shown in figure 2. The provenances Paraguay and Holguín-Mayarí had excellent emergence percentage (100 %), just like Plain (97 %), and differed significantly from the others. These results are similar to the ones reported by Medina *et al*. (2007), who evaluated *M. oleifera* and *Leucaena leucocephala* in nursery stage, until 30 days after sowing, and obtained 95 and 100 %, respectively.

The lowest emergence values were found in the accessions Guatemala and PKM-1 (76 and 80 %, respectively), which could have been due to the above-explained elements related to seed storage, and because the seeds were harvested between three and four months before sowing. Thus, the report by Croess and Villalobos (2008), regarding the fact that the germination capacity of moringa seeds decreases when they are stored for more than two months, is corroborated.

On the other hand, Nonagaki (2006) stated that all seeds have sensors, which detect environmental changes, and that all the conditions that surround them directly determine the success of germination and, consequently, the success of seedling emergence and establishment.



a, b, c, d, e: Values with different superscripts differ at p < 0,05 (Duncan, 1955).

Figure 2. Performance of emergence and survival in nursery (%).

Emergence occurred since six days after the seeds were sown. The emergence index was lower than the one reported by Bezerra *et al*. (2004) when evaluating the effect and type of substratum on the germination and growth of this species in the nursery stage. These authors stated that the first seedlings emerged between three and four days after sowing.

However, Oliveira (2000) reached an average emergence time of eight days, in intact seeds as well as in seeds without integuments, which is similar to the result obtained in this study.

Likewise, when evaluating other tree species under similar trial conditions, Wencomo (2004) found a marked dormancy effect caused by the seed hardness and the presence of water-impermeable seed coats, which restrict germination and emergence.

Regarding seedling survival, no significant differences were found among the provenances; they had 100 %, with the exception of Plain (97 %). These percentages are similar to the ones reported by Medina *et al*. (2007).

According to Moroto *et al*. (2000), in spite of its differences in the taxonomic location with regards to legumes, *M. oleifera* shows a similar architecture with relation to the pattern of branch formation and arrangement in its initial growth stage; the branches are scattered, of higher constitution and with many leaves.

In this sense, the provenance Holguín-Mayarí had a progressive increase of the number of leaves throughout the experiment and differed ($p < 0,05$) from the others (fig. 3), in which this indicator did not increase during the first 20 days. Since that moment, Plain and Criolla-Granma were better than the rest ($p < 0,05$), although they neither showed significant differences among themselves at 30 and 40 days after being sown, nor exceeded six leaves per plant.

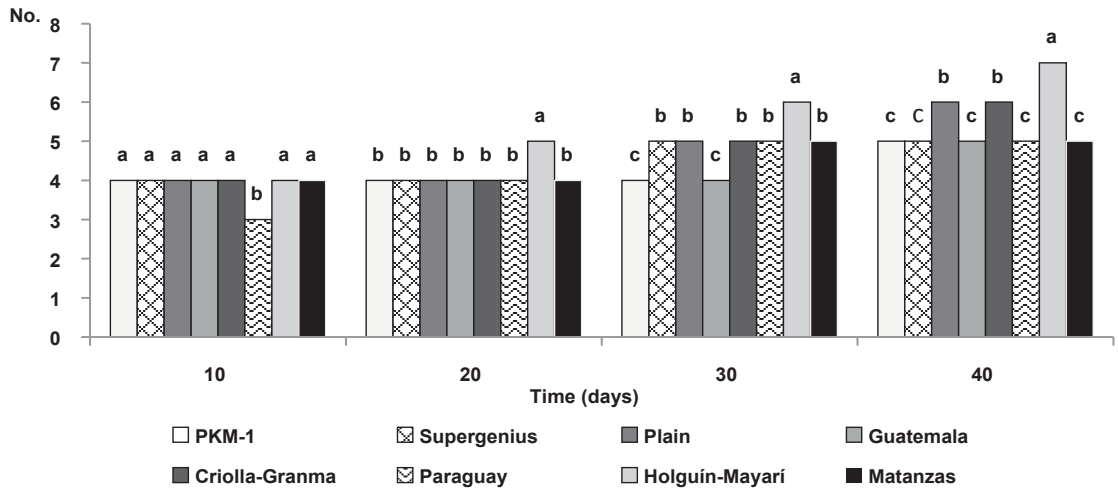
Medina *et al*. (2007), when comparing *M. oleifera* and *L. leucocephala* in the initial growth stage, found significant increases regarding the number of leaves until 42 days after germination in *M. oleifera*; yet, substantial increases were observed in *L. leucocephala* until 63 days.

Figure 4 shows the results of plant diameter 40 days after sowing. Similar values were obtained in all the provenances, which fluctuated between 0,33 and 0,40 cm.

On the other hand, Pérez (2011) achieved a better response when evaluating moringa under nursery conditions, using different substrata. Forty-two days after sowing, the seedlings reached a diameter of 0,58 cm with the combination of red soil plus earthworm humus.

Medina *et al*. (2007) and Medina *et al*. (2011) also obtained better results than the ones in this work, when evaluating moringa and other tree species under nursery conditions, with substrata of alkaline soils. In this study, *M. oleifera* was superior, because it reached 0,45 cm of diameter 35 days after being sown.

Regarding height (fig. 5), there was a progressive growth throughout the study. The highest values



a, b, c: Values with different superscripts differ at $p < 0,05$ (Duncan, 1955).

Figure 3. Number of leaves in the seedlings.

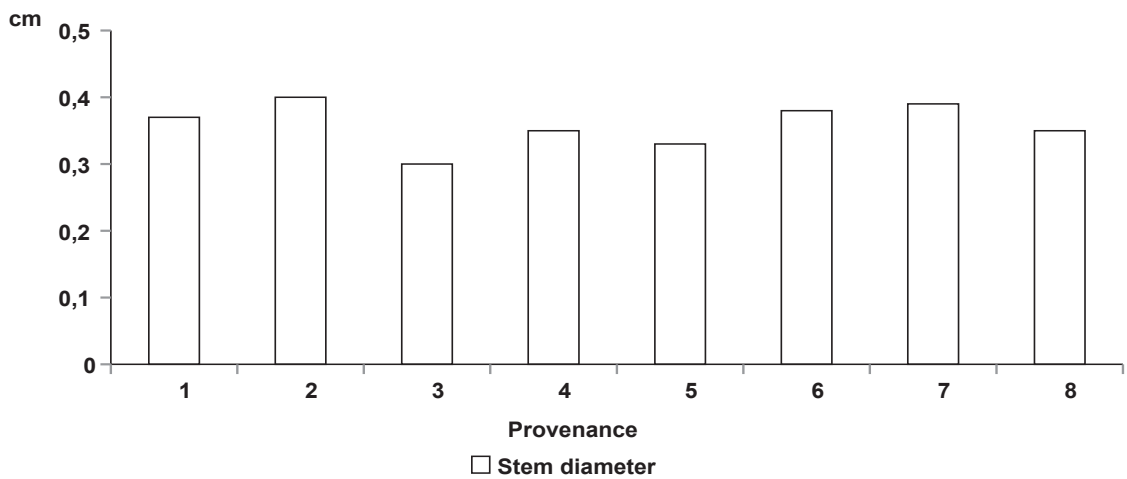


Figure. 4. Stem diameter of the seedlings (cm).

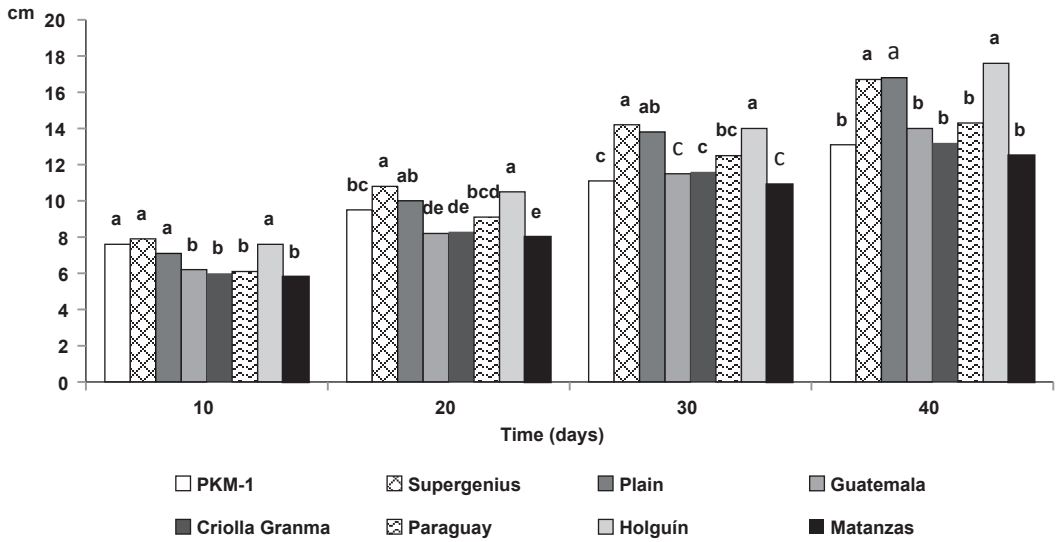
were observed at the end of the stage (40 days after sowing) in the provenances Holguín-Mayarí, Plain and Supergenius (17,5; 16,8 and 16,7 cm, respectively). Matanzas-Ciudad, Criolla-Granma, PKM-1, Guatemala and Paraguay showed the lowest values (12,45; 13,1; 13,1; 14,0 and 14,2 cm, respectively).

The provenances expressed their highest growth since 20 days after sowing, which is related to the principle that seedlings tend to develop the root system, in order to guarantee the subsequent water and nutrient absorption.

This coincides with the results obtained by Medina *et al*. (2007), who stated that moringa –since the beginning of its growth– experiences an accelerated trend because it develops a very deep root system, which makes a better utilization of soil nutrients and available water.

Seemingly, the fast growth of *M. oleifera* is a particularity of the species. In this sense, Toral (2005), when evaluating the establishment in field of 67 forage woody species, observed that moringa surpassed the others –even *L. leucocephala*– regarding establishment rate, because it reached 2 m of height at seven months, which is the prefixed height for the beginning of exploitation.

Table 2 shows the growth dynamics of the provenances in each of the samplings. Holguín-



a, b,:c, d, e: Values with different superscripts differ at p < 0,05 (Duncan, 1955).

Figure 5. Seedling height in the different samplings (cm).

Mayarí, Supergenius and Plain showed the highest increase (0,43; 0,41 and 0,42 cm/day, respectively) 40 days after being sown. Pérez *et al*. (2010) obtained similar values of growth rate in nursery, when using moringa seeds without scarification. Nevertheless, when the seeds were scarified, they observed a higher growth rate (up to 0,57 cm/day), justifying the previous reports about this process, which creates much more favorable conditions for the increase of seedling emergence in a significant way. In this study, the provenance with the lowest growth rate was Matanzas-Ciudad (0,31 cm/day).

With regards to the degree of lesions by pests and the symptoms caused by diseases, no affectations were observed from the scale used in the experiment. Thus, the provenances were considered immune. In Venezuela, Medina *et al*. (2007) found a similar response when studying the performance of moringa in the presence of the attack by pests and diseases in nursery.

However, García Roa (2003) and Reyes (2005) reported that *M. oleifera* plants are affected –under nursery conditions– by a varied number of insects, especially *Atta* sp, *Remigia latipes*, *Coccus* sp. and, to a lower extent, *Acerias beldoni*. Likewise, Alfaro and Martínez (2008) observed remarkable attacks by *Atta* spp. in nursery, mainly on the leaves and fresh shoots of the seedlings.

It is concluded that the studied provenances have morphological characteristics that differentiate them among themselves, and that Holguín-Mayarí showed the best performance regarding emergence, survival, height, stem diameter, number of leaves, growth rate and germination.

It is advisable to conduct further studies about the morphoagronomic performance of the different *M. oleifera* provenances during the establishment, as well as in their exploitation stage in different production systems.

Table 2. Growth dynamics (cm/day).

Day	Provenance							
	1	2	3	4	5	6	7	8
10	0,76	0,79	0,71	0,62	0,60	0,61	0,76	0,58
20	0,48	0,54	0,50	0,41	0,42	0,46	0,53	0,40
30	0,37	0,47	0,46	0,38	0,39	0,42	0,47	0,38
40	0,34	0,41	0,42	0,35	0,33	0,36	0,43	0,31

Received: July 19, 2013
Accepted: September 10, 2013