Technical Note

Collection and characterization of Jatropha curcas *L. provenances*

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ABSTRACT: The objective of this study was to characterize nine *Jatropha curcas* provenances, collected in several provinces of Cuba, during the nursery and establishment stages. To characterize these materials the methodology for the collection and characterization of germplasm was used. In the nursery, the sprouting of the propagules happened since 7 or 14 days after planting; while seedling emergence, when seeds were used, occurred 14 days after sowing. The survival percentage of the propagules fluctuated between 20 and 86,6 %, and from seed, from 86,6 to 90 %. In the field, survival varied between 66,6 and 100 % in the provenances transplanted by propagules and, in general, it was higher than the one detected in the nursery. In the provenances transplanted by seedlings no variation was detected in survival (90,9 % for the two accessions: CSSS-5 and CSS-6). It is concluded that the percentage of sprouting, emergence, rooting and survival were very variable indicators, under nursery as well as under field conditions, which was closely linked to the genotype, age and seed quality of the collected materials (propagules as well as seed). It is recommended to collect *J. curcas* in other zones, in order to contribute to the increase of the germplasm of this important species, which constitutes an alternative genetic resource for biofuel production.

Key words: nurseries, plant establishment, propagules, seedlings, survival

INTRODUCTION

The morphological and agronomic characterization of plant genetic resources allows to describe and differentiate the qualitative and quantitative attributes of several individuals of a species (Ramos *et al*., cited by Suárez *et al*., 2012). In addition, it allows the identification and selection of outstanding types for their future multiplication and use (Machado *et al*., 2012).

These techniques have been applied by researchers and specialists in many countries, in collections of different crops of interest, and have been mainly based on qualitative descriptors: color, texture and pigmentation; as well as quantitative ones: growth rate, height, number of nodes, number of leaves and their dimensions, stem diameter, number of branches of different orders, branch diameter, fruit number, dimensions and weight, and emergence, among others (Lezcano *et al*., 2012; Ramírez *et al*., 2012).

In the particular case of *Jatropha curcas* L., activities of collection and morphological and productive characterization of several provenances have been conducted (Machado, 2011a; 2011b), which has allowed to describe and identify the most prominent materials.

The objective of this work was to characterize a group of *J. curcas* provenances, collected in several provinces of Cuba, during the nursery and establishment stages.

MATERIALS AND METHODS

Zone and procedure for collection. The collection was made in different localities of three eastern provinces of Cuba (Victoria de las Tunas, Santiago de Cuba and Guantánamo) through the methodology proposed by Machado *et al*. (1999). A material was also obtained from seed, of the Pasture and Forage Research Station of Sancti Spiritus (CSSS-5) and another one, collected this way, from Santiago de Cuba (CSS-6). To characterize the provenances all the collected plant material (propagules and seed) was transferred to the Pasture and Forage Research Station Indio Hatuey.

The acquisition of propagules (cuttings) from preferably isolated, well-branched, vigorous plants, with abundant branches and leaves, with little or no affectation by pathogen microorganisms or phytophagous insects was an essential requisite, as long as the conditions allowed it.

Nevertheless, material was also collected from individuals which were part of more or less numerous wild populations, and others that were in limiting fences of cattle paddocks or crop fields, as well as in hedges; but with the condition that the deep effect of successive previous prunings was not observed, which could have altered the number of branches that these genotypes normally produced (table 1).

The propagules were cut from branches that could have higher or lower degree of caducity. In all cases the apical portions, in which the tissue and the pods are still immature, were discarded, because this can limit the rooting and appearance of growths, according to the observations previously made in cuttings of other species such as *Morus alba* (Boschini and Rodríguez, 2002). Fifteen stakes were cut in each of the donor plants, with a length from 30 to 40 cm and diameter of 1-4 cm, approximately. They were tied in a bundle, conveniently identified and wrapped in paper, which was maintained moist until arriving at their destination.

The provenances were identified with a key formed by the acronyms belonging to: the collection (C), site of origin (T: Victoria de las Tunas, G: Guantánamo, S: Santiago de Cuba) and seed type (E: stakes and S: seeds), which is shown in table 1.

Nursery stage. Sowing in the nursery was carried out in September, in a 3 x 1 m plot prepared for such purpose. Nylon bags of 28 x 13 cm and a substratum that consisted in a mixture of soil (70 %) and organic matter (30 %) were used. For the provenances CSE 3, CSE 4, CSE 5, CGE 2, CGE 1 and CTE 4, 15 replications were sown, and for CTE 1, 11 replications, due to the low availability of genetic material. In the case of CSSS-5 and CSS-6, collected by seed, 30 replications were planted for each one.

Manual irrigation was applied two or three times per week, to maintain the necessary moisture. During the first 95 days of this period, and every seven days, the number of cuttings with sprouts and the emerged seedlings (from seeds) were counted, in order to determine the survival percentage.

Establishment stage. The provenances were transferred to the field when they were 160 days old and had reached around 55-60 cm. They were planted in holes, separated at 2 m between plants and 3 m between provenances, at 30 cm of depth, and they were weekly irrigated. The number of

Table 1. Zones of collection and characteristics of the donor plants chosen for the harvest.

Key of the provenance	Zone of collection	Characteristics of the donor plants	Province
CSE-3	Alto Songo	Wild, individual plant, with abundant primary and secondary branches.	Santiago de Cuba
CSE-4	Alto Songo	Wild, individual plant, with abundant primary, secondary and tertiary branches.	Santiago de Cuba
CSE-5	Mayarí Arriba	Plant located in a living fence, with clusters of leaf buds (5-6 leaves) and abundant secondary and tertiary branches.	Santiago de Cuba
CGE-2	Farm El Mamoncillo	Wild, individual plant, with abundant secondary and tertiary branches.	Guantánamo
CGE-1	Farm Media Luna	Plant collected in a living fence, with a high branching degree.	Guantánamo
CTE-1	Farm Las Catalinas	Plant collected in a living fence, with a high branching degree.	Victoria de las Tunas
CTE-4	Farm Gallego	Plant collected in a hedge, with a high branching degree.	Victoria de las Tunas
CSSS-5	Seed donated by the Pasture Research Station of Sancti Spiritus	––	Sancti Spíritus
CSS-6	Mayarí Arriba	Seed collected in the living fence of a farm. Individual plant with high branching degree.	Santiago de Cuba

replications per provenance was related to the survival reached in the nursery stage.

The plants were considered established when the bud production phase (pre-flowering) began. During the establishment manual weeding was done, with a monthly or bimonthly frequency, depending on the weed-invasion degree. Observations were also made every 15 days on plants with sprouts and live seedlings, to calculate the survival percentage.

RESULTS AND DISCUSSION

Table 2 shows data of the eight collected provenances and their environment.

The group of collected provenances was relatively small. This was mainly related to the short period dedicated to the collection activities (four days) and the preference given to the specific location of plants with many branches and morphologically adequate for seed production, which required much time.

Nevertheless, it is valid to state that a higher quantity of donors was sampled, compared with the collection made in Sancti Spiritus (Machado, 2011a), which is related to the higher abundance of *J. curcas* in the eastern provinces (Toral *et al*., 2008). For such reason more attention should be dedicated to this aspect, in future collections of materials of this species.

As can be observed in table 2, the collection was conducted on plants that remained in wild or cultivated state.

However, in this last case, as the precaution was taken to select individuals which did not have the effect of successive prunings (table 1), their branches showed a natural morphological pattern and not the one which can be observed when a high production of branches is recorded on plants subject to successive prunings of the foliage (Alfonso, 2008; Dias *et al*., 2009). These requisites allowed to choose potentially adequate donors for seed production, which also showed a high differentiation degree in the habitats where they were selected, in correspondence with their genotypes.

During the nursery stage a remarkable variation was observed in the days that passed for the sprouting of the propagules or the seed emergence (table 3). Such performance is normally associated to the age of the donor plants, their individual characteristics and the branch sections from which the propagules were selected; as well as to the seed quality in the types collected this way.

In this sense, in the provenances collected by propagules, the donor individuals were regularly characterized by being vigorous and old, except in some cases in which they were relatively young, taking into consideration the height and branch vigor. Yet, in all these provenances the sprouts began to be observed since 7 or 14 days after being planted in the bags. This time can be considered precocious if it is compared with the results obtained by Machado (2011a), who detected the presence of sprouts since 35 days, in those propagules whose donor plants (3) were only two years old.

The highest survival values (80-86,6 %) were obtained in two of the Santiago de Cuba provenances, particularly in CSE-3, which can be considered acceptable; moderate values were found in those collected in Guantánamo (40-53 %); and the lowest in the materials collected in Victoria de las Tunas, especially in CTE-4 (20 %).

This contrasting response, for sprouting and emergence as well as for survival, could be associated to the heterogeneity of the genetic structures of the populations of this species, still undomesticated (Ríos, 2012).

Emergence started at 14 days for the two provenances planted by seed and the survival

Table 2. Data of the collected provenances and their environment.

Number of provenances	State of the sample	Type of vegetation	Land use	Specific habitat	Soil texture
8	Cultivated (3)	Grassland (2)	Paddock (4)	Hedge (4)	Clayey (5)
	Wild (5)	Scrub (4)	Cultivated area (1)	Clearing (1)	Loamy (2)
		Scrubland (2)	Road edge (3)	Mixed with pastures (1)	Sandy (1)
				Mixed with trees (1)	
				Living fence (1)	

The number of provenances is indicated between parentheses

Table 3. Number of sprouted or emerged plants and survival percentage in the nursery stage.

Provenance	No. of replications	Days passed after sowing										Survival (%)
		7	14	21	28	35	42	49	56	77	95	
CSE-3	15	3	3	11	14	14	13	13	13	13	13	86,6
CSE-4	15	2	4	4	7	8	8	8	6	7	7	46,6
CSE-5	15	6	9	10	11	13	13	13	13	13	12	80,0
CGE-2	15	7	9	9	9	8	7	7	7	6	6	40,0
CGE-1	15	7	7	7	7	8	8	8	8	8	8	53,3
CTE-1	11	1	4	4	6	6	6	6	5	5	4	36,3
CTE-4	15	0	1	2	2	3	3	3	3	3	3	20,0
CSSS-5$^{\Delta}$	30	0	24	24	25	26	26	26	26	26	26	86,6
CSS-6$^{\Delta}$	30	0	27	28	28	28	28	27	27	27	27	90,0

$^{\Delta}$Sowing by seed.

percentage was high in both. This could be related to seed quality because they were newly-harvested, which normally have a high germination percentage. In this sense, in the technical information of mexican *J. curcas* seeds it is referred that the fresh seed shows high germination percentages (around 80 %), although this indicator can vary between 60 and 90 % (SENASICA, 2012).

Table 4 shows the results of the number of rooted plants and their survival percentage under field conditions, since 25 days after sowing until 100 days. A noticeable variation was detected for the last indicator, with values that fluctuated from 66,6 to 100 % for the provenances obtained from propagules in the nursery and of 90,9 % for the seedlings obtained from the seed, which can be considered acceptable (Heller, 1996).

It is remarkable that the survival percentage in the field stage in some cases was higher –or much higher– than the one detected under nursery conditions (table 3). This response could have been due to the fact that their transference was done when they were 160 days old, for which the reserves in the propagules should have been high, so that these materials did not suffer, markedly, the stress that normally occurs in the plants transplanted from the nursery (Machado, 2011a). In addition, they received moisture through irrigation. The lowest values were observed in one of the materials collected in Guantánamo (CGE-2) and the highest ones, in the provenances collected in Santiago de Cuba and Victoria de las Tunas (CSE-4 and CTE-4).

In the accessions that were taken to the field in the form of seedlings (CSSS-5 and CSS-6) the

Table 4. Number of rooted plants and survival under field conditions.

Provenance	No. of replications	Days passed after planting						Survival (%)
		25	40	55	70	85	100	
CSE-3	13	13	13	12	10	10	10	76,9
CSE-4	7	7	7	7	7	7	7	100,0
CSE-5	12	10	9	9	9	9	9	75,0
CGE-2	6	6	5	5	4	4	4	66,6
CGE-1	8	8	8	6	6	6	6	75,0
CTE-1	4	4	4	3	3	3	3	75,0
CTE-4	3	3	3	3	3	3	3	100,0
CSSS-5	11	11	10	10	10	10	10	90,9
CSS-6	11	11	10	10	10	10	10	90,9

survival was high; this can be related to the nature of the propagation material, which is capable of developing a very strong root system of fast and efficient anchoring to the soil (Sunil *et al*., 2008).

It is concluded that the percentages of sprouting, emergence, rooting and survival were very variable indicators under nursery, as well as field conditions, which is closely related to the genotype, age and quality of the seed of the collected materials (propagules as well as seed). In addition, there was a similar fluctuation pattern in terms of rooted plants, but with a higher survival percentage in the field, to which the higher age of the material and the propitious moisture conditions due to the use of irrigation, contributed.

To continue the collection of *J. curcas* in other zones of the country is recommended, in order to have access to materials with adequate characteristics for seed production and thus contribute to the increase of the germplasm of this important species, which constitutes an alternative genetic resource for biofuel production.

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