



## SEED DIVERSITY IN NATIVE MEXICAN *Jatropha curcas* L. AND THEIR ENVIRONMENTAL CONDITIONS<sup>1</sup>

### [DIVERSIDAD EN SEMILLAS NATIVAS MEXICANAS DE *Jatropha curcas* L. Y SUS CONDICIONES AMBIENTALES]

Ofelia Andrea Valdés-Rodríguez<sup>1</sup>, Arturo Pérez-Vázquez<sup>2</sup>,  
Olivia Palacios-Wassenaar<sup>3\*</sup> and Odilón Sánchez-Sánchez<sup>4</sup>

<sup>1</sup>El Colegio de Veracruz. Carrillo Puerto 26, Zona Centro, Centro, 91000 Xalapa Enríquez, Ver. México. 01 228 841 5100. Email: andrea.valdes@gmail.com

<sup>2</sup>Colegio de Postgraduados-Campus Veracruz. km 88.5 carretera federal Xalapa-Veracruz, 91700 Ver. México. 01 229 201 0770. parturo@colpos.mx

<sup>3</sup>Instituto de Ecología A.C. Red de Biodiversidad y Sistemática. Carretera antigua a Coatepec 351, El Haya, Xalapa 91070, Ver., México. 01 228 8421800. Email: olivia.palacios@gmail.com

<sup>4</sup>Centro de Investigaciones Tropicales. Universidad Veracruzana. José María Morelos 44, Zona Centro, Centro, 91000 Xalapa Enríquez, Ver. 01 228 167 3051. Email: sasodil@yahoo.com.mx

\*Corresponding author

#### SUMMARY

This study presents an assessment of 51 provenances of *Jatropha curcas* L. collected in Mexico during a four-year period. Morphological (weight, length, width and thickness) and chemical characteristics (oil, protein and phorbol ester contents) were evaluated. Correlation and cluster analysis were performed to identify similarities among provenance's characteristics. Long term climatic data (30-year), and accumulative rain and average temperature during the previous 12 months were collected to determine climatic implications on seed characteristics by a principal component analysis. The results indicated that Mexican seeds are heavier and have greater morphological and chemical variability than those reported in other countries, with positive correlation between seed weight and accumulative rain during the previous seven months. The highest correlation between seed parameters was found among length and weight, while protein and oil contents had no correlation with morphological data. Principal component analysis showed that phorbol ester content was negatively correlated with temperature, rain and seed weight. It was concluded that due to their high seed weight, oil and protein contents, native Mexican provenances are valuable germplasm for genetic breeding programs with commercial purposes.

**Keywords:** non-toxic *Jatropha curcas*; seed characteristics; climatic influence.

#### RESUMEN

Este estudio presenta una evaluación de 51 procedencias *Jatropha curcas* L. colectadas en México durante un periodo de cuatro años. Se evaluaron características morfológicas (pesos, longitud, ancho y grosor) y químicas (contenido de aceites, proteínas y ésteres de forbol). Se realizaron análisis de conglomerados y correlación para identificar similitudes entre las características de las procedencias. Se tomaron datos climáticos de largo plazo (30 años), y precipitación acumulada y temperatura promedio durante los 12 meses previos, para determinar, mediante un análisis de componentes principales, las implicaciones climáticas sobre las características de las semillas. Los resultados indicaron que las semillas mexicanas son más pesadas y tienen mayor variabilidad morfológica y química que las reportadas en otros países, y una correlación positiva entre el peso y la lluvia acumulada en los siete meses previos. La mayor correlación entre parámetros se encontró entre longitud y peso, mientras que el contenido de proteínas y la toxicidad no estuvieron correlacionadas con la morfología de las semillas. El análisis de componentes

<sup>1</sup> Submitted February 06, 2018 – Accepted August 23, 2018. This work is licensed under a CC-BY 4.0 International License

principales indicó que el contenido de ésteres de forbol tuvo correlación negativa con la temperatura, la lluvia y el peso de la semilla. Se concluye que, dado el elevado peso y contenido de aceite y proteínas, las procedencias mexicanas constituyen un germoplasma valioso para programas de mejoramiento genético con fines comerciales.

**Palabras clave:** *Jatropha curcas* no tóxica; características de semillas; influencia climática.

## INTRODUCTION

*Jatropha curcas* L. is a subtropical species, considered native from Mexico and Central America, where it has a greater genetic diversity than in other parts of the world (Ovando-Medina *et al.*, 2011; Salvador-Figueroa *et al.*, 2015). Due to its multiple uses, *J. curcas* can be found nowadays in tropical and subtropical regions of Africa and Asia. The species is considered as toxic due to the presence of terpenoids (phorbol esters), which cause vomit and diarrhea (Makkar *et al.*, 1997; Makkar *et al.*, 1998). Ecotypes with very low or not detected phorbol ester contents, and therefore considered non-toxic, can be found only in Mexico (Martínez-Herrera *et al.*, 2006; Becker and Makkar, 2008). Research conducted at the end of the last century found that these ecotypes were located mainly in the eastern part of the country, where local people consume the seeds after cooking or roasting them (Makkar *et al.*, 1997; Makkar *et al.*, 1998). Currently, non-toxic ecotypes are extended to other parts of the country and even abroad due to their internationally recognized potential for food purposes (Francis *et al.*, 2013; Valdes-Rodriguez *et al.*, 2013b).

Notwithstanding its great potential as food and energy resource, the native Mexican *J. curcas* provenances have not yet been thoroughly researched. Considering this fact and the variability of their seeds, research for breeding purposes on these provenances has huge importance. This wide genetic diversity also needs to be evaluated under their natural conditions to determine their potential. Several authors have reported the phenotypical (Yi *et al.*, 2014; Rao *et al.*, 2015; Saadaoui *et al.*, 2015), chemical (Ginwal *et al.*, 2004) and genetic (Montes *et al.*, 2014; Rao *et al.*, 2015; Salvador-Figueroa *et al.*, 2015; Zavala del Angel *et al.*, 2016) variability of *J. curcas*. However, there is little information about the relationship between these characters and their native environmental factors; although in other countries some studies have found significant interaction among genotype and environment (Larcher, 2004; Xu *et al.*, 2012; Rao *et al.*, 2015).

In order to preserve the diversity of its native resources, the Mexican government implemented the National System of Phylogenetic Resources (SINAREFI) in 2002 (Córdova *et al.*, 2015). Part of this national program consisted in setting up research networks; one of these was the *Jatropha* spp. Group. Members of this network collected seed samples of toxic and non-toxic *J. curcas* in their native locations. However, a detailed evaluation of seed characters and agroecological factors has not been achieved yet. Learning and understanding how the soils and climatic characteristics affect the seed characters of these native germplasm is important to define the natural behavior of this species. Besides, it is also important to identify ecotypes with morphological and chemical advantages to select promising genotypes for commercial purposes. Therefore, the main purposes of this research consisted in evaluating the seed characteristics of native Mexican *J. curcas* provenances and the climate and soils of the sites where they were collected in order to provide an assessment of seed diversity and their native soils and climatic conditions.

## MATERIALS AND METHODS

The seek for *Jatropha* mature trees was performed from August to November (harvesting season for this species in the southeast of Mexico) during 2009, 2010, 2011 and 2012, inside the region comprised by the coordinates 21° 34' N and 98° 35' W and 19° 19' N and 96° 20' W. The following conventions were adopted: 1) every plant or group of plants separated by a distance longer than 100 m was identified as one provenance or site, and was geographically referenced using a GPS Garmin eTrex H; 2) every provenance collected each year was considered as one collection. For every site, all the available ripe fruits of the trees were harvested and transported to the facilities of El Colegio de Postgraduados where the seeds were obtained as in Valdes *et al.* (2011). Considering only fruiting trees, fifty-one provenances with ripe fruits were located during this four-year period. However, due to budget cuts and that some trees were no longer on the site during the following years, most

provenances were sampled for only one year. At the end of the four-year period, we obtained 65 different collections, of which three provenances were sampled three consecutive years, and eight provenances were sampled two consecutive years. Data from each provenance and its year of collection appears in Supplementary Table S1.

During the first two years, phorbol esters and percentage of oil content analysis were performed to 49 collections; additionally, we determined protein content to 13 collections. To prepare the samples for chemical analysis, seeds were cracked, subsequently the testa was removed and kernels were grounded using an electric mill for grain and vegetable samples. Oil content was determined through the ultrasound technique, using hexane as solvent. Quantitative determination of proteins by total nitrogen was performed using Kjeldahl method, according to standard REF AOAC 945.18-18 (AOAC, 2012). Phorbol ester content was determined by the technique proposed by Makkar *et al.* (1998), using 12-myristate-13-acetateforbol (PMA) at a concentration of 0.5 mg/ml as the standard reference. Seeds were considered toxic when its phorbol ester content was higher than 0.2 mg/g, based in previous information from the local people who usually consume the seeds, and chemical studies that analyzed those seeds (Valdes-Rodriguez *et al.*, 2013b).

The soil and climate information at the study region was obtained from the National Institute of Statistics and Geography (INEGI, 2012). Climate data is based in a modified Köppen classification for Mexico (García, 1973) within a 30-year period analysis, while soil classification was adapted from the FAO/UNESCO 1968 system by the Geography department of INEGI with a 1:250,000 scale. In order to analyze more specific data for each site, temperature and accumulative rain from the previous 12 months before harvesting were obtained from the nearest meteorological station of the National Meteorological System (SMN).

For each site and each year of collection, 100 seeds were randomly taken for morphological characterization: weight (with precision balance, 0.001 g), length, width and thickness (digital caliper, 0.01 mm). The sphericity and volume of seeds were calculated as in (Karaj and Müller, 2010), where the

higher the sphericity value for a seed, the closer is the shape to a sphere.

Statistical descriptors (maximum, minimum, average, standard deviation, and coefficient of variation CV) of seed characters were estimated for each collection. The relationship between the morphological variables of each collection (every site and year) was estimated by a Pearson *r* correlation analysis, and a *t* test with a significance level of 0.01. With the average data of each collection, a frequency distribution for the whole population (all sampled data) for each character was obtained.

The similarity among collections was estimated through a hierarchical cluster analysis (CA) by the square Euclidean distance as interval and the Ward as cluster method, with the help of the software SPSS v 23. The relationship among climate and soil type with seed morphological characters was determined by an analysis of variance of the sets of provenances that were grouped in each climate and soil classification, with a level of significance of 0.05 and the help of the software Sigmaplot 10.0.

The relationship among seed characters of each provenance by year of collection and its local environmental conditions was determined by correlating average precipitation, temperature and altitude from three data periods: a 30 year, a twelve-month and a seven-month period before harvesting. The climate data set with the highest correlation was used to perform a Principal Components Analysis (PCA) with the correlation matrix of this climate data and the average values of weight, size and phorbol ester contents of each provenance per year of collection.

To determine the relationship between average temperature and accumulative rain of the site and seed morphological characters, a regression analysis was performed for each provenance that had more than one year of collection and the seven-month period of climate data before harvesting.

## RESULTS

The average seed frequency distribution for the measured parameters of the whole data shows that 86% of seed weights are above 0.65 g (72% between 0.65 and 0.80 g); 87% between 17.5 and 19.5 mm

length; 65% between 7.5 and 9.0 mm width; 67% between 9 and 10.5 mm thickness; 63% have a volume between 5.5 and 7.1 cm<sup>3</sup>; and 69% have a sphericity between 0.61 and 0.65 (Fig. 1).

Considering the morphological characteristics of the seeds, three main groups were found in the dendrogram (Fig. 2). The cluster number 1 contained 51% of the provenances, mainly collected in 2009; these seeds were smaller and lighter than the ones in the other two clusters. Cluster 2 contained 23% of the provenances, grouping the longer and thicker seeds.

Cluster 3 contained 26% of the provenances, all of them collected in 2012, with the widest and more spherical seeds. Clusters 2 and 3 differ only 2% in weight, being not statistically different (P=0.55) for this character.

A high percentage of positive significant correlations (P<0.01) were found between weight and length, volume and thickness of the seeds from the 51 provenances (Table 1). Sphericity was the parameter with the lowest number of significant correlations between this data and weight.

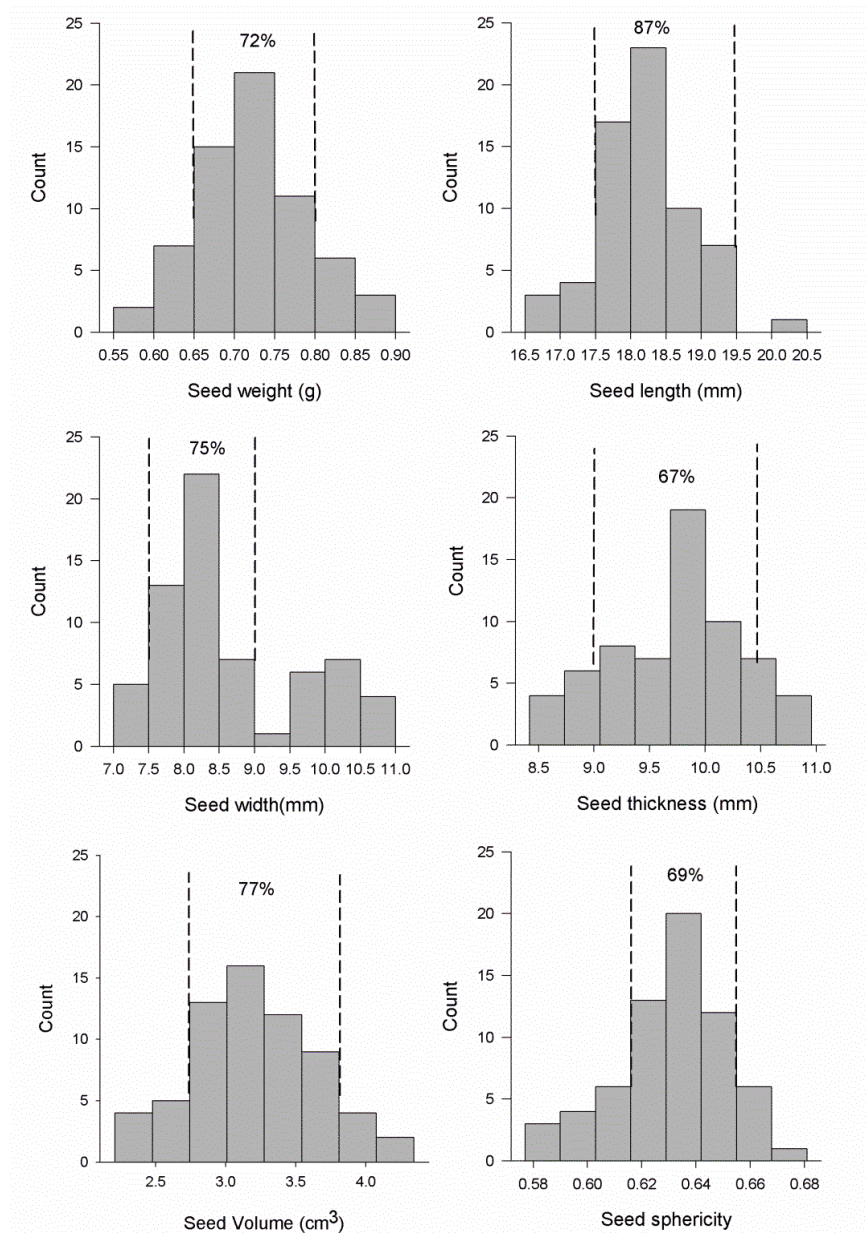


Figure 1. Frequency distribution of seed morphological characteristics from 51 native Mexican *J. curcas* provenances.

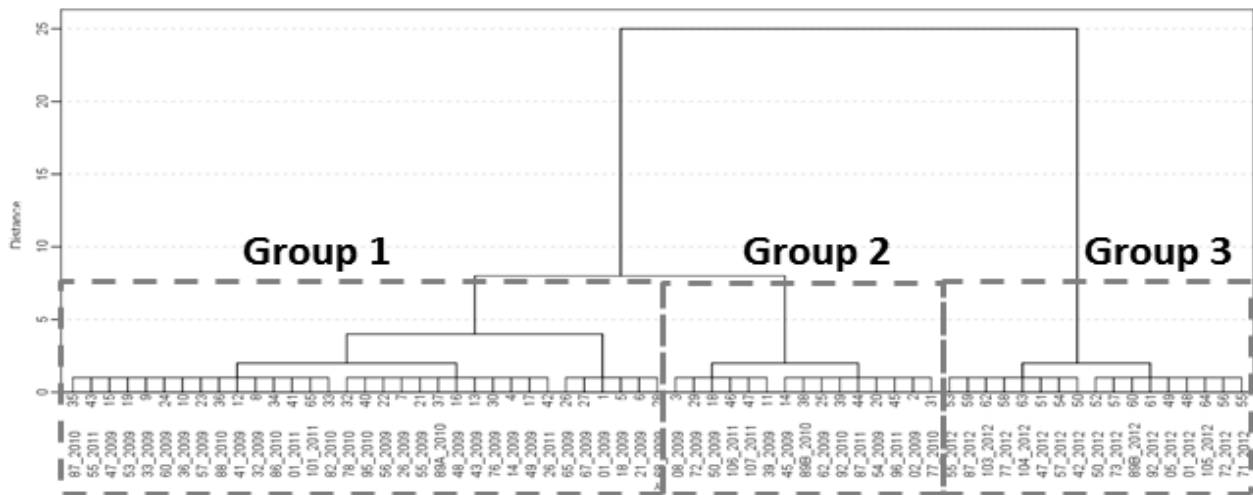


Figure 2. Dendrogram obtained from the cluster analysis for 51 native Mexican *Jatropha curcas* provenances in southeast Mexico.

Table 1. Percentage of positive significant correlations between weight, size and shape found among 51 native Mexican *Jatropha curcas* provenances

Parameter	Length	Width	Thickness	Sphericity	Volume
Weight	91	55	85	6	95
Length		40	52	82	95
Width			77	94	98
Thickness				83	95
Sphericity					58

All the provenances were found only in three of the eight existing climatic zones in the study area (Fig. 3, Table 2). Native *J. curcas* was found growing only in warm-humid (Am), warm-sub humid (Aw2) and semi-warm humid (Acf) climates, but not in drier or cooler climates. According to the cluster analysis, the heaviest seeds were mainly located in cluster 2, corresponding to a warm humid climate (Am). Most of the plants were found growing on Vertisol (33%) and Regosol (20%) soils, followed by Phaeozem (12%) soils. In these lands, 45% of the textures are considered fine (clayey), and 27% are coarse (sandy) or medium (loam) in equal proportions. Most provenances (88%) were collected in sites located at altitudes below 600 m above sea level (asl) and 70% of the sites had an annual rainfall between 1000 and 2000 mm.

Seed weight was similar among climate and soil types ( $P=0.55$  climate;  $P=0.58$  soil). No statistical differences were found between seed weight, length, width, thickness, volume and sphericity related to soil types ( $P>0.05$ ). However, seeds were longer and

thicker in the warm humid climate, indicating that places with higher annual rainfall produced bigger seeds.

There was no significant correlation between seed weight or size, and temperature or accumulative rainfall for the 30 year and 12 months period before collection. However, for the seven months before harvesting, seed weight had a positive nonlinear correlation when accumulative rainfall was lower than 2000 mm (Fig. 4a). An example can be seen at seed weight distribution in Medellin provenance, which moves towards heavier classes when accumulative rainfall increases (Fig. 4b). In addition, principal component analysis resulted in two components which explained 99.9 % of the total variance (Table 3a and 3b) and showed that seed weight and length were positively correlated with rainfall, while toxicity (phorbol esters) was negatively correlated with temperature and rainfall, whereas thickness had no correlation with climatic conditions (Fig. 5).

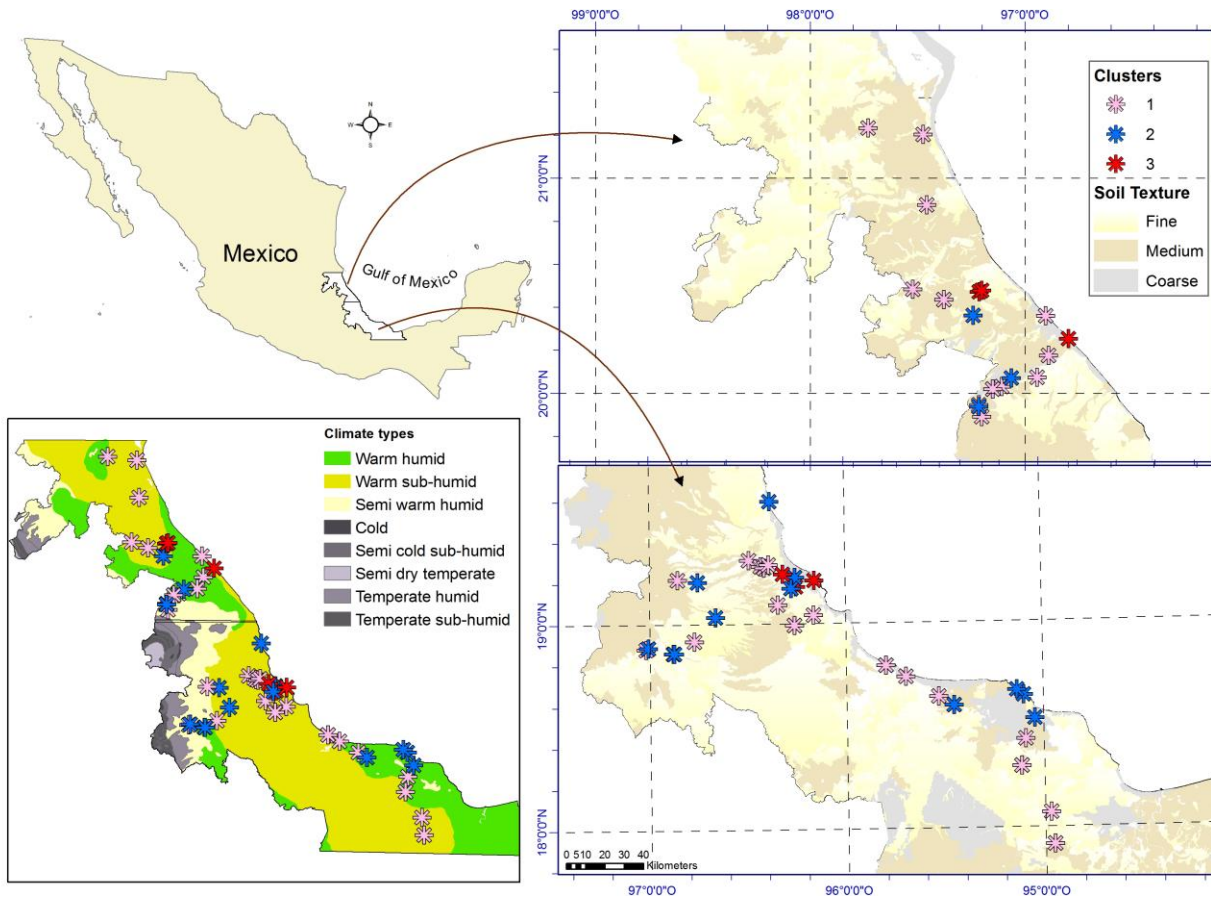


Figure 3. Geographic location, soil textures and climatic conditions from sites where native Mexican *J. curcas* provenances were collected.

Table 2. Percentage of native Mexican *Jatropha curcas* provenances found per climatic type, soil classification and soil use in southeastern Mexico.

Climate	Soil use	Soil Classification								
		Acrisol	Andosol	Cambisol	Feozem	Gleysol	Luvisol	Regosol	Rendzina	Vertisol
Warm humid	Wetland			100.0%						
	Cultivated pasture		57.1%			14.3%		28.6%		
	Evergreen forest						100.0%			
	Rainfeed agriculture				42.9%			14.3%		42.9%
	Urban				100.0%					
Warm sub-humid	Cultivated pasture			8.3%				50.0%		41.7%
	Irrigated agriculture				50.0%			50.0%		
	Evergreen forest								100.0%	
	Rainfeed agriculture			8.3%	8.3%			16.7%		66.7%
	Urban							100.0%		
Semi-warm humid	Evergreen forest									100.0%
	Rainfeed agriculture	60.0%	20.0%							20.0%

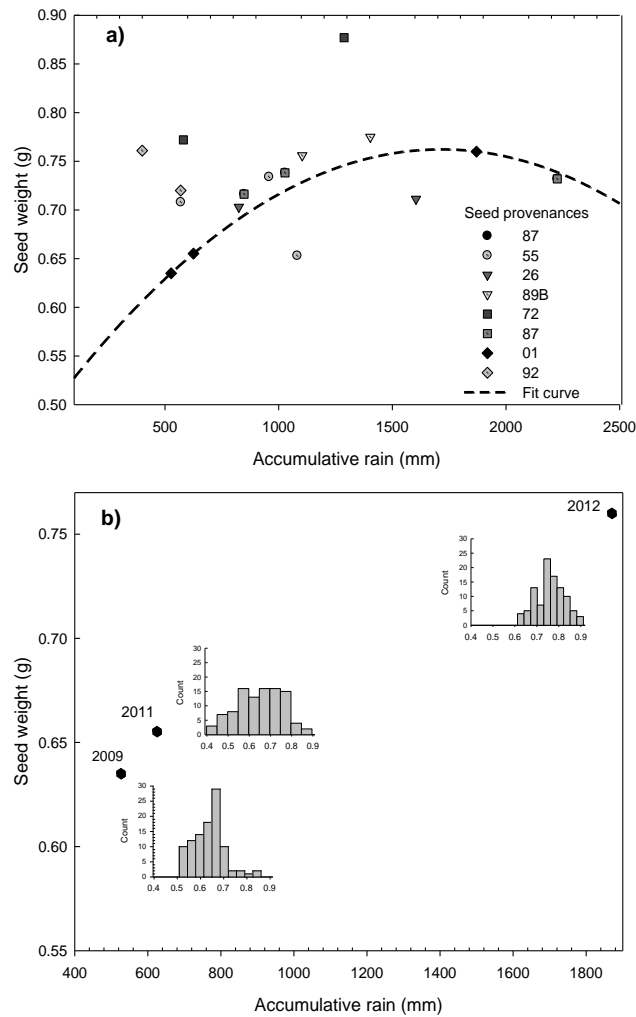


Figure 4. a) Seed weight versus accumulative rainfall seven months before collection. b) Seed weight distribution versus accumulative rainfall during a three years period in a native Mexican *J. curcas* provenance.

Table 3a. Total variance explained by the principal component analysis.

Component	Total	% of variance	% accumulated
1	456056.952	87.983	87.983
2	62285.153	12.016	99.999
3	1.610	.000	100.000

Table 3b. Principal component analysis values obtained by the Varimax method and Kaiser normalization.

Variable	Re-scaled component	
	1	2
Altitude	.978	-.211
Temperature	-.823	.087
Rain	.785	.619
Phorbol_esters	-.535	-.024
Width	-.030	.477
Volume	-.003	.464
Length	.070	.458
Sphericity	-.080	.243
Weight	.113	.221
Thickness	-.004	.025

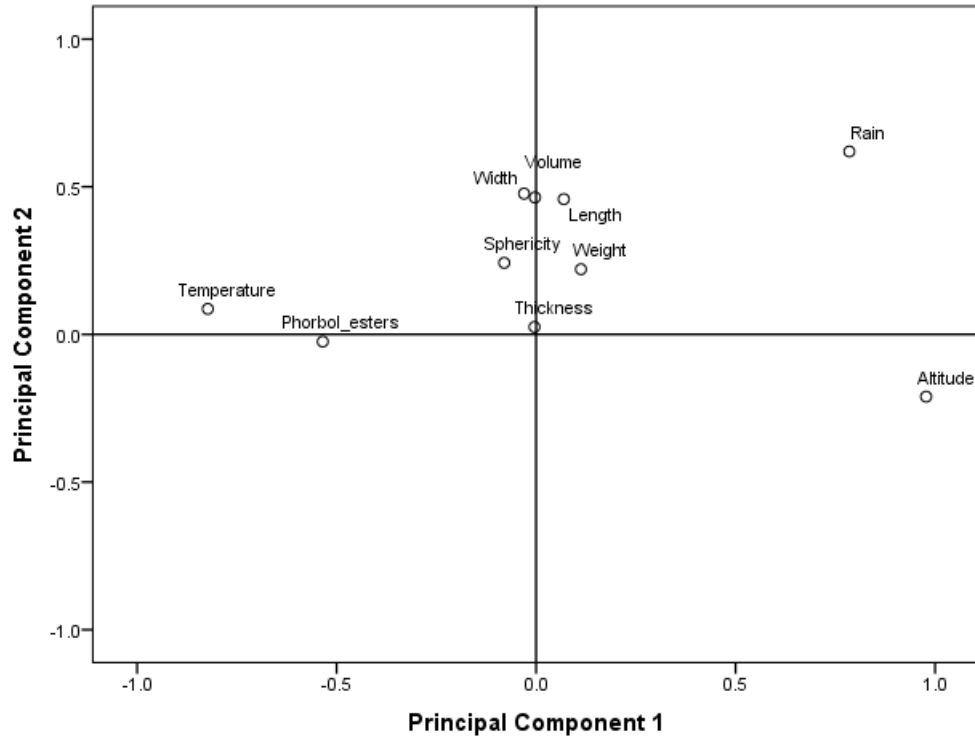


Figure 5. Principal component analysis (PCA) for seed characteristics of native Mexican *J. curcas* provenances and environmental factors.

In relation to seed toxicity, 60% of the provenances were non-toxic, and these were mostly concentrated at the north side of the study area (Fig. 6), while the toxic ones were found at the southern part. There was a positive linear correlation ( $r=0.17$ ) between phorbol esters and protein contents, but this was not statistically significant ( $P=0.54$ ). Average  $\pm$  SD (Min-

Max) phorbol ester content was  $0.22\pm0.45$  (0 – 2.69 mg/g); oil content (%) was  $47.42\pm6.58$  (32 – 61); and protein content (%) was  $25.89\pm5.02$  (18.34 – 34.43). No significant correlation between weight and the chemical characteristics of the seeds was found neither between protein nor between oil contents ( $P=0.22$ ).

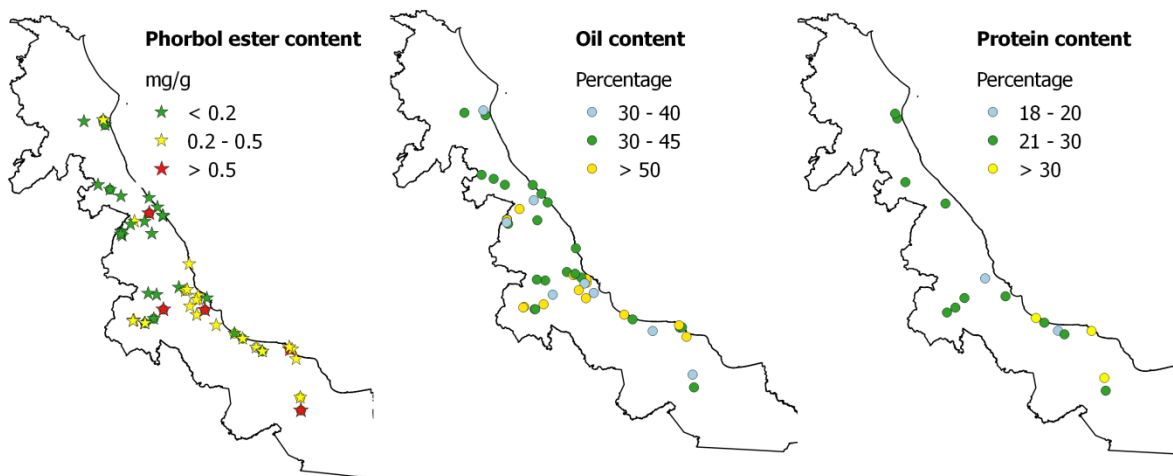


Figure 6. Toxicity, protein and oil percentage in native Mexican *J. curcas* provenances.



## DISCUSSION

Average seed size ranges, including sphericity and volume, registered more extended ranges than seeds from southeast and west provenances of Mexico (Table 4), India and South America (Karaj and

Müller, 2010; Montes *et al.*, 2011; Shabanimofrad *et al.*, 2013; Yi *et al.*, 2014) indicating a higher morphology diversity of these native seeds; which is consistent with plants from the centers of origin (Salvador-Figueroa *et al.*, 2015).

Table 4. Average and ranges of seed morphological data and their corresponding climate reported for *Jatropha curcas* in different regions of Mexico

Character	Southeast Mexico (1)	Southeast Mexico (2)	Southeast Mexico (3)	Southeast Mexico (4)	Pacific coast Mexico (5)
Seed weight (g)	0.72	0.76	0.87	0.66	0.79
Average					
Seed weight (g) (min – max)	0.57 - 0.88				
Seed length (mm)	18.2	18.4	20.3	18.3	18.7
Average					
Seed length (mm) (min – max)	16.5 - 20.1				
Seed width (mm)	8.6	10.5	10.6	10.7	11.5
Average					
Seed width (mm) (min – max)	7.1 - 10.7				
Seed thickness (mm)	9.7	8.8	9	8.7	9
Average					
Seed thickness (mm) (min – max)	8.4 – 11.0				
Volume (cm <sup>3</sup> )	3.2	3.6	4.1	3.6	4.0
Average					
Volume (cm <sup>3</sup> ) (min – max)	4.4 – 8.7				
Sphericity (%)	0.63	0.65	0.61	0.65	0.67
Average					
Sphericity (%) (min – max)	0.58 - 0.68				
Seed oil content (%)	48		46	37	
Average					
Seed oil content (%) (min – max)	32 - 61				
Seed protein content (%)	18 - 34		30	22	
(min – max)					
Seed phorbol ester content (mg/g) (min – max)	0 - 5.21	0.11	0.18	2.65	0.25
Rain (mm) (min – max)	936 - 3716	1186	1900 – 2600	1400 - 1600	450
Temperature (°C) (min – max)	19 - 25	24	24	28	26.4
Climate type	Aw2, Am(f), C(f)	<b>Aw1</b>	Aw2	Am(f)	Aw0
Altitude (m.a.s.l.) (min – max)	1 - 1011	71	690	73	251
Soil texture	Sandy, loamy, clayey	Sandy loam	Sandy clay loam	Sandy lime loam	Sandy loam
Agronomic management	No fertilization or irrigation applied	No fertilization or irrigation applied	No fertilization or irrigation applied	No fertilization or irrigation applied	Fertilization and irrigation applied
Number of accessions	51	<b>1</b>	1	1	1

Note: (1) Current work; (2, 3) (Valdés-Rodríguez *et al.* 2013a); (4, 5) (Cruz *et al.* 2015)

With a normal distribution, sphericity and length were the less variable parameters among the 51 provenances, with the lowest CV values (4.0% and 4.4%, respectively). Volume was the most disperse variable, because width and thickness had not a normal distribution ( $P < 0.01$ ) and had high CV (5.0% and 7.2% respectively). This indicates that width and thickness are the highest source of variation in *Jatropha* seed morphology. Although, seed weight had the highest variation, with a CV of 10.8%, which is similar to Indian seeds, whose weight variation ranges between 8.96 to 10.13% (Karaj and Müller, 2010; Wani *et al.*, 2012). Studies including other regions in Africa, Asia and Central and South America reported a seed weight CV of 13.48% (Montes *et al.*, 2014), being still lower than the seeds assessed here. Besides being more diverse, the higher variation in seeds from the same provenance is an indication of a low seed selection. Fig. 7 allows to identify two regions where seeds have the lowest morphological variation, located in the north and center-west part of the study area. While most seeds

in the north part belong to cluster 1 and have the lowest weights, seeds located in the center-west belong to clusters 2 and 3, containing the heaviest seeds. Therefore, it can be considered that these provenances are valuable for breeding purposes due to their greater size and low variation.

In regard to the relationships between size and weight, significant but lower correlations between weight and length were also found in seeds from China, Laos and Thailand (0.48), and Malaysia (0.57) (Guan *et al.*, 2013; Shabanimofrad *et al.*, 2013), and in other Mexican collections of seeds from the same study area where correlations were similar (0.73) (Cruz *et al.*, 2015). However, seeds from India had no significant correlation between these parameters; instead the highest correlation (0.44) was found among weight and width (Rao *et al.*, 2008). This means that southeastern native Mexican provenances are heavier and longer.

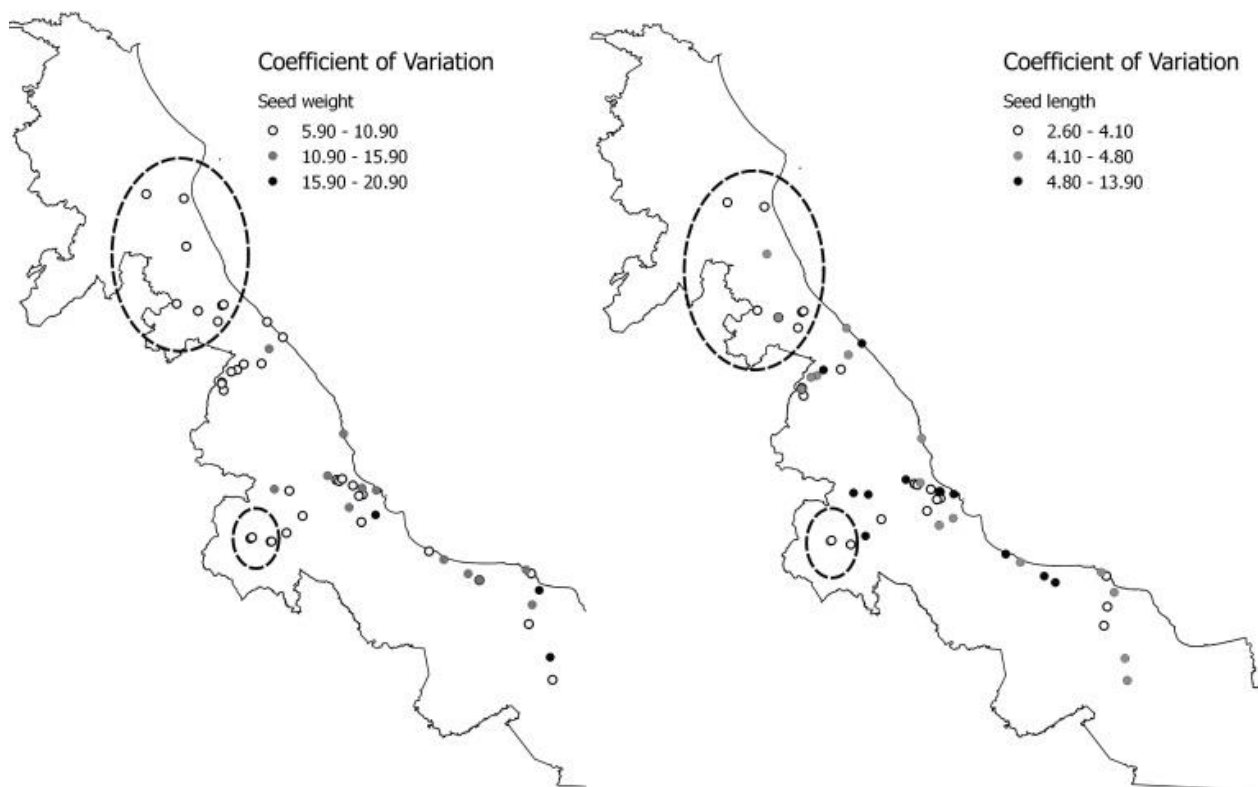


Figure. 7. Coefficients of variation of seed weight and length distribution of 51 Mexican *Jatropha curcas* provenances.

The dendrogram showed that seeds with an average weight of 0.68 g are the most abundant (Cluster 1); while 49% of the seeds have average weights over 0.75 g. Here, it is important to point out that these native Mexican provenances have superior seed weights (>0.70 g) compared to other from locations around the world that were grown under agronomic management with irrigation and fertilization (Karaj and Müller, 2010; Montes *et al.*, 2011; Yi *et al.*, 2014). This means that native Mexican seeds are naturally bigger, especially considering the fact that all of them came from non-commercial plantations, and did not receive any agronomic or genetic management.

Dendrogram also showed that morphology varied among the year of collection, by grouping seeds from the same provenances in different clusters. This result highlights a high variability within provenances. It was found that this variation is highly related to climatic conditions, with water availability as a key factor. The analysis shows that the amount of accumulative rain during the previous seven months was a crucial factor to increase seed weight (Figs. 4 and 5). Therefore, it is recommended to invest in irrigation when trying to increase yield or improve seed characters for commercial purposes.

By analyzing climatic conditions from the collected provenances, it was found that these native plants are more adapted to higher temperatures and more humid conditions than the ones reported in other subtropical places around the world where non-native *J. curcas* have been cultivated (Wen *et al.*, 2012; Adil *et al.*, 2015). Climatic growing conditions found in the current research are very similar to the ones reported by Maes *et al.* (2009) and Córdova *et al.* (2015).

Around 86% of the land use where the provenances were collected was rainfed agriculture or cultivated pasture. Although, it is important to remark that the researched plants were not found as a part of commercial plantations, but, as life fences or garden plants. The high rate of land conversion obeys to the fact that 90 % of this region of Mexico was changed from primary forest to extensive livestock (Medina *et al.*, 2010), therefore *J. curcas* was used as live fence in many parts.

Most of the plants were located in Vertisol and Regosol, according to the soil maps (INEGI, 2012),

because these are the most abundant and representative soils in Veracruz. Both Vertisol and Regosol soils are known by their clayed texture and their slightly acid to alkaline pH (Medina *et al.*, 2010).

Principal component analyses showed no correlation between seed size or weight and soil type. Hence it is considered that soil type is not a key factor for the development of seed characters in these native provenances as irrigation, fertilization, or the genetics of the plant. However, considering the soil types (Vertisols and Regosols), it was noticed that these native *J. curcas* provenances have evolved in rich soils, being naturally not fitted to poor soils, as opposed to what is commonly expressed by other authors (e.g. Heller, 1996; Openshaw, 2000; Martínez-Herrera *et al.*, 2006). Another important finding is the high percentage of native *J. curcas* plants growing in clayey soils, which are reported as not suitable for this species (FACT, 2010; Trabucco *et al.*, 2010). On this regard, it can be considered that these plants survive in this type of soil because the region has a long dry season (about five to seven months), with heavy rains concentrated mainly during one or two months, separated by a period of one month during summer to autumn. Consequently, their roots are exposed to excess of water only during a brief period. Therefore, considering that previous studies demonstrated that *J. curcas* could develop well in clay-loam soils if the water is not stagnated (Valdés-Rodríguez *et al.*, 2011), it can be stated that *J. curcas* can survive well on clay soils under these conditions.

The high number of seeds considered as non-toxic found in the southeast of Mexico could be explained by the fact that, in this region *Jatropha* seeds are used as food by Totonaca descendants, an ethnic group that has inhabited this territory for centuries (Aguilera, 2004). Previous research found that the Totonacas dispersed the non-toxic plants by seeds and cuttings (Valdés-Rodríguez *et al.*, 2013; Valdés-Rodríguez *et al.*, 2013b), which could contribute to the wide variation found in seed toxicity degree, ranging from not detected up to 2.7 mg/g phorbol esters. Although these values are still lower than in other parts of the country, where seeds with up to 4.04 mg/g of phorbol esters contents have been found (Martínez-Herrera *et al.*, 2010).

Protein in kernels (16 - 34%) had similar ranges than in provenances from other parts of Mexico (18 – 32%) (Martínez *et al.*, 2007; Martínez-Herrera *et al.*, 2010). Studies about the protein contents in *Jatropha* kernels found that its digestibility could be higher than 70% after pressure cooking (Martínez-Herrera *et al.*, 2010). This could be a very important character that is still not commercially exploited. Therefore, the non-toxic provenances represent an added value in these cases in which seeds are used for biofuel purposes.

Oil content in these seeds also have a wide variation (32 – 61%), with 59% of the provenances having 50% or more oil in kernels. *Jatropha*'s seed oil content in other ecotypes of the world varies mainly between 30 to 50% (Gubitz *et al.*, 1999; Kaushik *et al.*, 2007; Wang *et al.*, 2008; Basha *et al.*, 2009; Nzikou *et al.*, 2009; He *et al.*, 2011) and between 50 to 63% in the kernel (Akhbar *et al.*, 2009; Senger *et al.*, 2016). In this regard, seed collectors in Mexico have identified very oily seeds, with up to 65% oil content (SINAREFI, *Jatropha* spp. Network), but a breeding program of these varieties has not yet been developed in the country. Therefore, we highlight the importance to promote the development of a breeding program of these elite plants for commercial purposes.

Other important application from this research is that this climatic and soil information can be a useful input to locate regions where *J. curcas* can be successfully cultivated.

## CONCLUSIONS

*Jatropha curcas* seeds in their native environment show a very high diversity intra and inter provenance, from seed shape to chemical characters that only diversification centers show. Therefore, we consider that the location and characterization of the plants provided in the current study could be used to increase the available data for this valuable germplasm. It can finally be remarked that, due to the nature of the soils where the native provenances were found, it is considered that they are not well adapted to poor soils and dry climates. It is important to consider this fact in *Jatropha* cultivating programs for commercial purposes.

## Acknowledgements

The Priority Research Line 3 (Alternate Energy and Biomaterials) from El Colegio de Postgraduados Campus Veracruz is acknowledged.

## REFERENCES

- Adil E., S., Kant D., R., Tripathi, V., Bakshi, M., Srivastava, P., Jamil, S., Singh, H.B., Singh, N. and Abhilash, P. C. 2015. *Jatropha curcas* L.: A crucified plant waiting for resurgence. *Renewable and Sustainable Energy Reviews*. 41:855–862. <http://doi.org/10.1016/j.rser.2014.08.082>
- Aguilera, M. R. 2004. *Recetario Totonaco de la costa de Veracruz*. F. Iturraga, J; Marcelli, A; Guevara, Ed. 2nd ed. Mexico, D.F., CONACULTA.
- Akhbar, E., Yaakob, Z., Kamaruddin, S.K., Ismail, M. and Salimon, J. 2009. Characteristics and composition of *Jatropha curcas* oil seed from Malaysia and its potential as biodiesel feedstock. *European Journal of Scientific Research*. 29(3):396-403. <https://www.scopus.com/record/display.uri?eid=2-s2.0-67449103136&origin=inward&txGid=4647bc6696380fa5398e1074891eb4f>
- AOAC. 2012. *Official Methods of Analysis of AOAC International 19th ed.*, Gaithersburg, MD, USA: AOAC 50 International. Available online at: [www.eoma.aoac.org](http://www.eoma.aoac.org) (Verified 14 jun 2015)
- Basha, S. D., G. Francis, H. P. S. Makkar, K. Becker and Sujatha, M. 2009. A comparative study of biochemical traits and molecular markers for assessment of genetic relationships between *Jatropha curcas* L. germplasm from different countries. *Plant Science*. 176: 812-823. <https://doi.org/10.1016/j.plantsci.2009.03.008>
- Becker, K. and Makkar, H. P. S. 2008. *Jatropha curcas*: A potential source for tomorrow's oil and biodiesel. *Lipid Technology*. 20(5): 104–107. <http://doi.org/10.1002/lite.200800023>
- Córdova T., L., Bautista R., E., Zamarripa C., A., Rivera L., J., Pérez V., A., Sánchez S., O., Martínez H., J. and Cuevas S., J. 2015. *Diagnóstico y Plan Estratégico de la Red Jatropha spp. en México*. México: SNICS, SINAREFI.
- Cruz R., B. A., Pérez-Vázquez, A., García-Pérez, E., Gallardo L., F. and Soto H., R. M. 2015.

- Análisis químico-morfológico comparativo de accesiones de *Jatropha curcas* L. del estado de Veracruz. *Revista Mexicana de Ciencias Agrícolas*. 6(3):589–601. [http://www.scielo.org.mx/scielo.php?script=sci\\_arttext&pid=S2007-09342015000300012&lng=es&nrm=iso](http://www.scielo.org.mx/scielo.php?script=sci_arttext&pid=S2007-09342015000300012&lng=es&nrm=iso)
- FACT. 2010. From Cultivation to Application. The *Jatropha Handbook*. Fuels from Agriculture in Communal Technology foundation, Eindhoven. [http://www.globalbioenergy.org/uploads/media/0603\\_FACT\\_Foundation\\_-\\_Jatropha\\_Handbook.pdf](http://www.globalbioenergy.org/uploads/media/0603_FACT_Foundation_-_Jatropha_Handbook.pdf)
- Francis, G., Oliver, J. and Sujatha, M. 2013. Non-toxic *Jatropha* plants as a potential multipurpose multi-use oilseed crop. *Industrial Crops and Products*. 42(1):397–401. <http://doi.org/10.1016/j.indcrop.2012.06.015>
- García, E. 1973. Modificaciones al sistema de clasificación climática de Köppen (Vol. 1). México, D. F.: Universidad Autónoma de México.
- Ginwal, H., Rawat, P. and Srivastava, R. 2004. Seed source variation in growth performance and oil yield in *Jatropha curcas* Linn. in central India. *Silvae Genetica*. 53:186–192. <https://doi.org/10.1515/sg-2004-0034>
- Guan, J., Yu, H., Zhang, J., Yang, R. and Fan, Y. Hong. 2013. Study on seed morphology and genetic diversity of *Jatropha curcas* L. from different provenances. *Advance Journal of Food Science and Technology*. 5(2):169–173. [maxwellsci.com/print/ajfst/v5-169-173.pdf](http://maxwellsci.com/print/ajfst/v5-169-173.pdf)
- Gübitz, G. M., Mittelbach, M. and Trabi, M. 1999. Exploitation of the tropical oil seed plant *Jatropha curcas* L. *Bioresource Technology*. 67(1):73–82. [http://doi.org/10.1016/S0960-8524\(99\)00069-3](http://doi.org/10.1016/S0960-8524(99)00069-3)
- He, W., King, A. J., Khan, M. A., Cuevas, J. A., Ramiaramanana, D. and Graham, I. A. 2011. Analysis of seed phorbol-ester and curcumin content together with genetic diversity in multiple provenances of *Jatropha curcas* L. from Madagascar and Mexico. *Plant Physiology and Biochemistry*. 49(10):1183–1190. <http://doi.org/10.1016/j.plaphy.2011.07.006>
- Heller, J. 1996. Physic nut. *Jatropha curcas* L. Promoting the conservation and the use of underutilized and neglected crops. Gatersleben, Germany-Rome, Italy: Institute of plant genetics and crop plant-international plant genetic resources institute.
- INEGI. 2012. Cartas edafológicas y Climáticas. Escala 1:250.000. Veracruz, México: Instituto Nacional de Estadística, Geografía e Informática. <http://www.beta.inegi.org.mx/app/mapas/>
- Karaj, S. and Müller, J. 2010. Determination of physical, mechanical and chemical properties of seeds and kernels of *Jatropha curcas* L. *Industrial Crops and Products*. 32(2):129–138. <http://doi.org/10.1016/j.indcrop.2010.04.001>
- Kaushik, N., Kumar, K., Kumar, S., Kaushik, N. and Roy, S. 2007. Genetic variability and divergence studies in seed traits and oil content of *Jatropha (Jatropha curcas* L.) accessions. *Biomass and Bioenergy*. 31(7):497–502. <http://doi.org/10.1016/j.biombioe.2007.01.021>
- Larcher, W. 2004. *Ecofisiología vegetal*. São Carlos: RiMa.
- Maes, W. H., Trabucco, A., Achten, W. M. J. and Muys, B. 2009. Climatic growing conditions of *Jatropha curcas* L. *Biomass and Bioenergy*. 33(10):1481–1485. <https://doi.org/10.1016/j.biombioe.2009.06.001>
- Makkar, H. P. S., Aderibigbe, A. O. and Becker, K. 1998. Comparative evaluation of non-toxic and toxic varieties of *Jatropha curcas* for chemical composition, digestibility, protein degradability and toxic factors. *Food Chemistry*. 62(2):207–215. [http://doi.org/10.1016/S0308-8146\(97\)00183-0](http://doi.org/10.1016/S0308-8146(97)00183-0)
- Makkar, H. P. S., Becker, K., Sporer, F. and Wink, M. 1997. Studies on Nutritive Potential and Toxic Constituents of Different Provenances of *Jatropha curcas*. *Journal of Agricultural and Food Chemistry*. 45(8):3152–3157. <http://doi.org/10.1021/jf970036j>
- Martínez H., J., Evangelista, L. S. and Martínez, A. A. 2007. Perfil nutricional al de semillas de *Jatropha curcas* L. provenientes de Michoacán. *Alimentos Ciencia e Ingeniería*. 16 (2):313–314.
- Martínez H., J., Martínez A., A., Makkar, H., Francis, G. and Becker, K. 2010. Agroclimatic conditions, chemical and nutritional characterization of different provenances of *Jatropha curcas* L. from Mexico. *European*

- Journal of Scientific Research. 39(3):396–407. <http://www.eurojournals.com/ejsr.htm>
- Martínez-Herrera, J., Siddhuraju, P., Francis, G., Dávila-Ortiz, G. and Becker, K. 2006. Chemical composition, toxic/antimetabolic constituents, and effects of different treatments on their levels, in four provenances of *Jatropha curcas* L. from Mexico. Food Chemistry. 96(1):80–89. <http://doi.org/10.1016/j.foodchem.2005.01.059>
- Medina C., A., Salazar C., T. and Álvarez P., J. L. 2010. Fisiografía y suelos. In: Atlas del patrimonio natural, histórico y cultural de Veracruz (Eds Florescano and Ortiz), pp. 30–43. Veracruz, Mexico: Gobierno del Estado de Veracruz.
- Montes O., L. R., Torres S., A. F., Jongschaap, R. E. E., Azurdía P., C. A., Berduo S., J. E., Trindade, L. M., Visser, R. G. F. and Van Loo, E. N. 2014. High level of molecular and phenotypic biodiversity in *Jatropha curcas* from Central America compared to Africa, Asia and South America. BMC Plant Biology. 14(1):77. <http://doi.org/10.1186/1471-2229-14-77>
- Montes, J. M., Rodríguez A., M., Vaca Chávez, J., Guzmán, C. and Calandri, E. 2011. Characterization of *Jatropha curcas* L. seed and its oil, from Argentina and Paraguay. Journal of the Argentine Chemical Society. 98:1–9. <https://www.aqa.org.ar/index.php/volumenes-publicados/78-vol-98-2011>
- Montes, J. M., Technow, F., Martin, M. and Becker, K. 2014. Genetic diversity in *Jatropha curcas* L. assessed with SSR and SNP markers. Diversity. 6:551–566. <http://doi.org/10.3390/d6030551>
- Nzikou, J. M., L. Matos, F. Mbemba, C. B. Ndangui, N. P. G. Pambou-Tobi, A. Kimbonguila, Th. Silou, M. Linder and S. Desobry. 2009. Characteristics and composition of *Jatropha curcas* oils, variety Congo-Brazzaville. Research Journal of Applied Sciences, Engineering and Technology. 1(3):154-159. [http://maxwellsci.com/print/rjaset\(3\)154-159.pdf](http://maxwellsci.com/print/rjaset(3)154-159.pdf)
- Openshaw, K. 2000. A review of *Jatropha curcas*: an oil plant of unfulfilled promise. Biomass and Bioenergy. 19(1):1–15. [http://doi.org/10.1016/S0961-9534\(00\)00019-2](http://doi.org/10.1016/S0961-9534(00)00019-2)
- Ovando-Medina, I., Sánchez-Gutiérrez, A., Adriano-Anaya, L., Espinosa-García, F., Núñez-Farfán, J. and Salvador-Figueroa, M. 2011. Genetic diversity in *Jatropha curcas* populations in the State of Chiapas, Mexico. Diversity. 3(4):641–659. <http://doi.org/10.3390/d3040641>
- Rao, G. R., Korwar, G. R., Shanker, A. K. and Ramakrishna, Y. S. 2008. Genetic associations, variability and diversity in seed characters, growth, reproductive phenology and yield in *Jatropha curcas* (L.) accessions. Trees - Structure and Function. 22(5):697–709. <http://doi.org/10.1007/s00468-008-0229-4>
- Rao, G. R., Sarkar, B., Raju, B. M. K., Reddy, P. S. and Kumar, P. S. 2015. Genetic association, divergence and genotype × environment interaction in *Jatropha* (*Jatropha curcas* L.). Biofuels. 6(5–6):323–329. <http://doi.org/http://dx.doi.org/10.1080/1759-7269.2015.1110773>
- Saadaoui, E., Martín, J. J., Bouazizi, R., Ben Romdhane, C., Grira, M., Abdelkadir, S., Larbi K., M. and Cervantes, E. 2015. Phenotypic variability and seed yield of *Jatropha curcas* L. introduced to Tunisia. Acta Botanica Mexicana. 110:119–134. <http://www.redalyc.org/articulo.oa?id=57432981006>
- Salvador-Figueroa, M., Magaña-Ramos, J., Vázquez-Ovando, A., Adriano-Anaya, M. L. and Ovando-Medina, I. 2015. Genetic diversity and structure of *Jatropha curcas* L. in its centre of origin. Plant Genetic Resources. 13(1):9–17. <http://doi.org/10.1017/S1479262114000550>
- Senger, E., Bohlinger, B., Esgaib, S., Hernández-Cubero, L. C., Montes, J. M. and Becker, K. 2016. Chuta (edible *Jatropha curcas* L.), the newcomer among underutilized crops: a rich source of vegetable oil and protein for human consumption. European Food Research and Technology. 243(6):987–997. <http://doi.org/10.1007/s00217-016-2814-x>
- Shabanimofrad, M., Rafii, M. Y., Megat Wahab, P. E., Biabani, A. R. and Latif, M. A. 2013. Phenotypic, genotypic and genetic divergence found in 48 newly collected Malaysian accessions of *Jatropha curcas* L. Industrial Crops and Products. 42:543–551. <http://doi.org/10.1016/j.indcrop.2012.06.023>
- SINAREFI. Red *Jatropha*. Available online: [http://www.sinarefi.org.mx/redes/red\\_jatropha.html](http://www.sinarefi.org.mx/redes/red_jatropha.html) (accessed on 16 April 2014).

- Trabucco, A., Achten, W. M. J., Bowe, C., Rafaerts, Van Orshoven, J., Norgrove, L. and Muys, B. 2010. Global mapping of *Jatropha curcas* yield based on response of fitness to present and future climate. *GCB Bioenergy*. 2:139–151. <https://doi.org/10.1111/j.1757-1707.2010.01049.x>
- Valdés-Rodríguez, O. A., Pérez V., A., García P., E. and Inurreta A., H. D. 2013a. Condiciones agroecológicas de procedencias nativas de *Jatropha curcas* L. In *Energía Alternativa y Biocombustibles* (Eds. A. Pérez V. and E. García P.) pp. 143–152. Mexico: Colegio de Postgraduados.
- Valdes-Rodriguez, O. A., Sanchez-Sanchez, O., Perez-Vazquez, A. and Caplan, J. 2013b. The Mexican Non-toxic *Jatropha curcas* L., Food Resource or Biofuel? *Ethnobotany Research and Applications*. 11:1–7. <http://doi.org/10.11547-3465-11-001>
- Valdes-Rodriguez, O. A., Sánchez-Sánchez, O., Pérez-Vázquez, A. and Ruiz-Bello, R. 2011. Soil texture effects on the development of *Jatropha* seedlings – Mexican variety “piñón manso”. *Biomass and Bioenergy*. 35(8):3529–3536. <http://doi.org/10.1016/j.biombioe.2011.05.003>
- Wang, Z.Y., Lin, J.M. and Xu, Z.F. 2008. Oil contents and fatty acid composition in *Jatropha curcas* seeds collected from different regions. *Nan Fang Yi Ke Da Xue Xue Bao*. 28(6):1045-1046. <https://www.ncbi.nlm.nih.gov/pubmed/18583260>
- Wani, T. A., Kitchlu, S. and Ram, G. 2012. Genetic variability studies for morphological and qualitative attributes among *Jatropha curcas* L. accessions grown under subtropical conditions of North India. *South African Journal of Botany* 79:102–105. <http://doi.org/10.1016/j.sajb.2011.10.009>
- Wen, Y., Tang, M., Sun, D., Zhu, H., Wei, J., Chen, F. and Tang, L. 2012. Influence of Climatic Factors and Soil Types on Seed Weight and Oil Content of *Jatropha curcas* in Guangxi, China. *Procedia Environmental Sciences*. 12(Icese 2011):439–444. <http://doi.org/10.1016/j.proenv.2012.01.301>
- Xu, W., Mulpuri, S. and Liu, A. 2012. Genetic diversity in the *Jatropha* genus and its potential application. *CAB Reviews: Perspectives in Agriculture, Veterinary Science, Nutrition and Natural Resources*. 7(59):1–15. <http://doi.org/10.1079/PAVSNNR20127059>
- Yi, C., Reddy, C., Varghese, K., Ha Bui, T. N., Zhang, S., Kallath, M., Kunjachen, B., Ramachandran, S. and Hong, Y. 2014. A new *Jatropha curcas* variety (JO S2) with improved seed productivity. *Sustainability*. 6(7):4355–4368. <http://doi.org/10.3390/su6074355>
- Zavala Del Angel, I., García-Pérez, E., González-Hernández, D., Pérez-Vázquez, A. and Ávila Reséndiz, C. 2016. Genetic Diversity of *Jatropha curcas* L. in Veracruz State, Mexico and its Relationships with the Content of Phorbol Esters. *Global Advanced Research Journal of Agricultural Science*. 5(5):149–158. <http://garj.org/full-articles/genetic-diversity-of-jatropha-curcas-l-in-veracruz-state-mexico-and-its-relationships-with-the-content-of-phorbol-esters.pdf?view=inline>

**Supplementary Table S1.** Morphological mean±SD values in 51 native Mexican provenances by year of collection (SD=Standard Deviation)

Provenance	Year	Weight ± SD (g)	Length ± SD (mm)	Width ± SD (mm)	Thickness ± SD (mm)	Volume ± SD (mm <sup>3</sup> )	Sphericity ± SD
01	2009	0.635 ±0.068	17.472 ±0.844	7.427 ±0.445	8.874 ±0.470	0.483 ±0.052	0.600 ±0.023
02	2009	0.751 ±0.090	18.037 ±0.787	8.931 ±0.645	10.645 ±0.556	0.721 ±0.091	0.664 ±0.026
08	2009	0.803 ±0.065	19.410 ±0.722	7.439 ±0.448	10.107 ±0.282	0.612 ±0.055	0.584 ±0.016
14	2009	0.686 ±0.059	17.790 ±0.754	7.517 ±0.568	9.353 ±0.375	0.525 ±0.062	0.606 ±0.024
18	2009	0.590 ±0.054	16.844 ±0.641	7.790 ±0.578	9.704 ±0.261	0.534 ±0.049	0.643 ±0.022
21	2009	0.633 ±0.048	16.587 ±0.473	7.641 ±0.474	9.423 ±0.400	0.501 ±0.046	0.640 ±0.020
26	2009	0.703 ±0.054	18.321 ±0.665	7.893 ±0.493	9.853 ±0.382	0.598 ±0.066	0.614 ±0.019
32	2009	0.708 ±0.073	17.681 ±0.806	8.006 ±0.639	9.851 ±0.446	0.586 ±0.076	0.632 ±0.023

Provenance	Year	Weight $\pm$ SD (g)	Length $\pm$ SD (mm)	Width $\pm$ SD (mm)	Thickness $\pm$ SD (mm)	Volume $\pm$ SD (mm <sup>3</sup> )	Sphericity $\pm$ SD
33	2009	0.661 $\pm$ 0.073	17.646 $\pm$ 0.847	8.559 $\pm$ 0.625	9.861 $\pm$ 0.501	0.627 $\pm$ 0.086	0.647 $\pm$ 0.024
36	2009	0.752 $\pm$ 0.053	17.994 $\pm$ 0.729	8.291 $\pm$ 0.655	10.325 $\pm$ 0.466	0.647 $\pm$ 0.083	0.642 $\pm$ 0.022
39	2009	0.826 $\pm$ 0.051	20.053 $\pm$ 0.853	8.539 $\pm$ 0.714	10.688 $\pm$ 0.375	0.768 $\pm$ 0.091	0.610 $\pm$ 0.027
41	2009	0.697 $\pm$ 0.095	17.740 $\pm$ 0.716	8.015 $\pm$ 0.648	10.328 $\pm$ 0.606	0.618 $\pm$ 0.089	0.640 $\pm$ 0.024
43	2009	0.619 $\pm$ 0.065	18.358 $\pm$ 0.983	7.803 $\pm$ 0.490	9.206 $\pm$ 0.412	0.554 $\pm$ 0.072	0.598 $\pm$ 0.022
45	2009	0.719 $\pm$ 0.064	18.598 $\pm$ 0.724	8.447 $\pm$ 0.762	10.197 $\pm$ 0.531	0.674 $\pm$ 0.096	0.629 $\pm$ 0.028
47	2009	0.721 $\pm$ 0.094	17.897 $\pm$ 0.911	8.123 $\pm$ 0.625	10.008 $\pm$ 0.592	0.612 $\pm$ 0.088	0.633 $\pm$ 0.026
48	2009	0.745 $\pm$ 0.086	18.494 $\pm$ 0.865	7.847 $\pm$ 0.542	10.271 $\pm$ 0.513	0.626 $\pm$ 0.077	0.618 $\pm$ 0.024
49	2009	0.662 $\pm$ 0.074	17.904 $\pm$ 0.905	7.723 $\pm$ 0.592	9.423 $\pm$ 0.467	0.546 $\pm$ 0.063	0.610 $\pm$ 0.026
50	2009	0.832 $\pm$ 0.076	19.294 $\pm$ 0.850	8.379 $\pm$ 0.517	10.537 $\pm$ 0.531	0.714 $\pm$ 0.066	0.619 $\pm$ 0.022
53	2009	0.660 $\pm$ 0.072	18.026 $\pm$ 0.844	8.359 $\pm$ 0.525	10.041 $\pm$ 0.439	0.635 $\pm$ 0.071	0.637 $\pm$ 0.026
54	2009	0.657 $\pm$ 0.131	18.393 $\pm$ 0.852	8.302 $\pm$ 0.707	10.561 $\pm$ 0.471	0.679 $\pm$ 0.095	0.637 $\pm$ 0.024
55	2009	0.653 $\pm$ 0.111	18.333 $\pm$ 1.083	7.651 $\pm$ 0.613	9.816 $\pm$ 0.494	0.580 $\pm$ 0.089	0.607 $\pm$ 0.028
56	2009	0.696 $\pm$ 0.123	18.199 $\pm$ 0.770	8.039 $\pm$ 0.532	9.978 $\pm$ 0.382	0.613 $\pm$ 0.065	0.623 $\pm$ 0.022
57	2009	0.710 $\pm$ 0.124	18.020 $\pm$ 0.993	8.235 $\pm$ 0.791	10.362 $\pm$ 1.193	0.651 $\pm$ 0.139	0.640 $\pm$ 0.044
60	2009	0.722 $\pm$ 0.082	17.680 $\pm$ 1.225	8.579 $\pm$ 0.834	10.089 $\pm$ 0.566	0.645 $\pm$ 0.110	0.652 $\pm$ 0.034
62	2009	0.875 $\pm$ 0.079	18.521 $\pm$ 0.932	8.427 $\pm$ 0.680	9.842 $\pm$ 0.344	0.646 $\pm$ 0.083	0.623 $\pm$ 0.025
65	2009	0.620 $\pm$ 0.077	17.633 $\pm$ 1.059	7.077 $\pm$ 0.436	8.418 $\pm$ 0.565	0.442 $\pm$ 0.063	0.577 $\pm$ 0.026
67	2009	0.601 $\pm$ 0.046	17.552 $\pm$ 0.577	7.386 $\pm$ 0.329	8.463 $\pm$ 0.348	0.460 $\pm$ 0.042	0.588 $\pm$ 0.017
68	2009	0.633 $\pm$ 0.055	16.511 $\pm$ 0.673	7.362 $\pm$ 0.410	9.055 $\pm$ 0.422	0.462 $\pm$ 0.048	0.626 $\pm$ 0.023
72	2009	0.877 $\pm$ 0.099	19.326 $\pm$ 0.764	8.033 $\pm$ 0.520	9.856 $\pm$ 0.498	0.643 $\pm$ 0.080	0.596 $\pm$ 0.019
76	2009	0.713 $\pm$ 0.067	18.454 $\pm$ 0.882	7.726 $\pm$ 0.603	9.522 $\pm$ 0.796	0.570 $\pm$ 0.082	0.600 $\pm$ 0.033
77	2010	0.858 $\pm$ 0.073	18.925 $\pm$ 0.801	9.218 $\pm$ 0.757	10.956 $\pm$ 0.480	0.803 $\pm$ 0.099	0.656 $\pm$ 0.026
78	2010	0.740 $\pm$ 0.070	18.147 $\pm$ 0.822	7.926 $\pm$ 0.684	9.888 $\pm$ 0.539	0.597 $\pm$ 0.073	0.620 $\pm$ 0.030
82	2010	0.691 $\pm$ 0.045	17.497 $\pm$ 0.463	8.113 $\pm$ 0.621	9.384 $\pm$ 0.469	0.557 $\pm$ 0.048	0.629 $\pm$ 0.029
86	2010	0.703 $\pm$ 0.071	17.660 $\pm$ 0.776	8.050 $\pm$ 0.620	9.925 $\pm$ 0.549	0.592 $\pm$ 0.069	0.635 $\pm$ 0.030
87	2010	0.738 $\pm$ 0.095	17.945 $\pm$ 1.046	8.438 $\pm$ 0.609	9.764 $\pm$ 0.558	0.623 $\pm$ 0.092	0.635 $\pm$ 0.026
88	2010	0.733 $\pm$ 0.070	17.965 $\pm$ 0.599	8.517 $\pm$ 0.675	10.267 $\pm$ 0.481	0.660 $\pm$ 0.078	0.647 $\pm$ 0.023
89A	2010	0.780 $\pm$ 0.062	18.155 $\pm$ 0.727	7.683 $\pm$ 0.522	10.074 $\pm$ 0.531	0.590 $\pm$ 0.068	0.617 $\pm$ 0.024
89B	2010	0.756 $\pm$ 0.069	18.461 $\pm$ 0.756	8.633 $\pm$ 0.568	10.193 $\pm$ 0.431	0.683 $\pm$ 0.083	0.637 $\pm$ 0.021
92	2010	0.761 $\pm$ 0.111	18.398 $\pm$ 0.972	8.316 $\pm$ 0.679	9.998 $\pm$ 0.608	0.646 $\pm$ 0.109	0.626 $\pm$ 0.022
95	2010	0.648 $\pm$ 0.108	18.110 $\pm$ 0.759	7.929 $\pm$ 0.639	9.985 $\pm$ 0.492	0.601 $\pm$ 0.069	0.622 $\pm$ 0.021
01	2011	0.655 $\pm$ 0.112	17.619 $\pm$ 0.920	8.212 $\pm$ 0.674	9.874 $\pm$ 0.425	0.601 $\pm$ 0.081	0.639 $\pm$ 0.023
26	2011	0.711 $\pm$ 0.042	17.683 $\pm$ 0.755	7.588 $\pm$ 0.570	9.786 $\pm$ 0.359	0.551 $\pm$ 0.061	0.619 $\pm$ 0.025
55	2011	0.708 $\pm$ 0.074	17.874 $\pm$ 1.022	8.445 $\pm$ 0.552	9.776 $\pm$ 0.436	0.621 $\pm$ 0.088	0.637 $\pm$ 0.022
87	2011	0.716 $\pm$ 0.140	18.721 $\pm$ 1.065	8.168 $\pm$ 0.718	9.999 $\pm$ 0.550	0.645 $\pm$ 0.106	0.615 $\pm$ 0.025
96	2011	0.700 $\pm$ 0.076	18.497 $\pm$ 0.907	8.484 $\pm$ 0.663	10.591 $\pm$ 0.307	0.697 $\pm$ 0.077	0.641 $\pm$ 0.028
101	2011	0.572 $\pm$ 0.072	17.427 $\pm$ 0.684	8.030 $\pm$ 0.630	9.765 $\pm$ 0.540	0.575 $\pm$ 0.079	0.637 $\pm$ 0.024
106	2011	0.752 $\pm$ 0.070	19.160 $\pm$ 0.737	8.561 $\pm$ 0.667	10.502 $\pm$ 0.465	0.724 $\pm$ 0.091	0.626 $\pm$ 0.022
107	2011	0.821 $\pm$ 0.101	19.083 $\pm$ 0.843	8.100 $\pm$ 0.714	10.671 $\pm$ 0.412	0.694 $\pm$ 0.096	0.619 $\pm$ 0.023
01	2012	0.760 $\pm$ 0.064	18.551 $\pm$ 0.791	10.129 $\pm$ 0.430	9.579 $\pm$ 0.456	0.756 $\pm$ 0.081	0.656 $\pm$ 0.018
05	2012	0.741 $\pm$ 0.057	18.828 $\pm$ 0.915	10.182 $\pm$ 0.460	9.087 $\pm$ 0.565	0.731 $\pm$ 0.083	0.639 $\pm$ 0.023
42	2012	0.675 $\pm$ 0.043	17.232 $\pm$ 0.615	9.582 $\pm$ 0.428	8.654 $\pm$ 0.478	0.599 $\pm$ 0.058	0.654 $\pm$ 0.025
47	2012	0.734 $\pm$ 0.059	17.976 $\pm$ 0.895	9.864 $\pm$ 0.448	8.903 $\pm$ 0.493	0.663 $\pm$ 0.072	0.648 $\pm$ 0.024
50	2012	0.787 $\pm$ 0.097	19.420 $\pm$ 1.103	10.713 $\pm$ 0.532	9.418 $\pm$ 0.837	0.827 $\pm$ 0.137	0.644 $\pm$ 0.026



<b>Provenance</b>	<b>Year</b>	<b>Weight ± SD (g)</b>	<b>Length ± SD (mm)</b>	<b>Width ± SD (mm)</b>	<b>Thickness ± SD (mm)</b>	<b>Volume ± SD (mm<sup>3</sup>)</b>	<b>Sphericity ± SD</b>
55	2012	0.734 ±0.059	18.448 ±0.754	9.813 ±0.463	8.990 ±0.529	0.684 ±0.084	0.638 ±0.019
57	2012	0.680 ±0.068	18.022 ±0.799	10.032 ±0.704	8.665 ±0.798	0.661 ±0.120	0.644 ±0.030
71	2012	0.764 ±0.072	18.276 ±0.693	10.690 ±0.488	9.858 ±0.523	0.809 ±0.088	0.681 ±0.022
72	2012	0.772 ±0.116	18.277 ±1.052	10.536 ±0.645	9.219 ±0.647	0.750 ±0.128	0.662 ±0.017
73	2012	0.867 ±0.093	19.215 ±0.707	10.635 ±0.454	10.119 ±0.506	0.868 ±0.089	0.663 ±0.020
77	2012	0.699 ±0.063	18.129 ±0.684	9.896 ±0.409	9.166 ±0.429	0.691 ±0.073	0.651 ±0.016
87	2012	0.732 ±0.077	18.310 ±0.829	9.773 ±0.667	8.976 ±0.615	0.677 ±0.104	0.639 ±0.024
89B	2012	0.775 ±0.068	18.505 ±0.833	10.199 ±0.517	9.237 ±0.565	0.732 ±0.088	0.651 ±0.025
92	2012	0.720 ±0.059	18.529 ±0.716	10.187 ±0.404	8.982 ±0.460	0.711 ±0.065	0.644 ±0.019
103	2012	0.757 ±0.060	18.618 ±0.719	9.856 ±0.394	8.814 ±0.494	0.678 ±0.063	0.631 ±0.025
104	2012	0.727 ±0.043	18.172 ±0.619	10.050 ±0.353	9.337 ±0.427	0.715 ±0.059	0.658 ±0.020
105	2012	0.809 ±0.093	18.809 ±1.048	10.370 ±0.525	9.393 ±0.616	0.768 ±0.086	0.651 ±0.035