ΦYTON
REVISTA INTERNACIONAL DE BOTÁNICA EXPERIMENTAL
INTERNATIONAL JOURNAL OF EXPERIMENTAL BOTANY
FUNDACION ROMULO RAGGIO
Gaspar Campos 861, 1638 Vicente López (BA), Argentina
www.revistaphyton.fund-romuloraggio.org.ar

Capsaicinoids concentration in *Capsicum annuum* var. *glabriusculum* collected in Tamaulipas, Mexico

Concentración de capsaicinoides en *Capsicum annuum* var. *glabriusculum* colectado en Tamaulipas, México

Aguirre-Hernández E[1], R San Miguel-Chávez[2], M Palma Tenango[2], ME González-Trujano[3], E de la Rosa-Manzano[4], G Sánchez-Ramos[4], A Mora-Olivo[4], A Martínez- Palacios[5], JG Martinez-Ávalos[4]

Abstract. HPLC analysis of capsaicin and dihydrocapsaicine concentrations obtained from fruits of *Capsicum annuum* var. *glabrisculum*, collected from four localities in the state of Tamaulipas, Mexico, was investigated. Selection of localities for this study was carried out considering that 65% of the people living in them gather wild chilli pepper for either autoconsumption or commercial purposes. An ANOVA analysis was carried out for determining capsaicinoids composition variation. Also, a multivariate analysis was carried out using Pearson correlation and principal components analysis to elucidate the influence that environmental factors might have on capsaicinoids concentration. Fruits from the locality El Gavilán had the higher concentration of capsaicine among the populations examined. Also, fruits from this location had the higher concentration for both alcaloids (994 µg/g), and the locality Comas Altas showed the lower concentration with 498 µg/g. A positive correlation between capsaicin and dihydrocapsaicin concentrations was found. PCA analysis showed that altitude, temperature and vegetation were the main factors responsible for the production of both alcaloids in *C. annuum* var. *glabriusculum*, and rainfall was the less important variable. Regarding the effects of altitude on alcaloids production it was found that locality El Gavilán, placed 578 m a.s.l., had the higher concentration for both alcaloids whereas the locality Comas Altas, placed at 65 m a.s.l., showed the lower concentrations. Locality La San Juana also had high concentrations of capsaicinoids. Apparently, this was due to the microclimate conditions present on the vegetation types on the above mentioned localities. Results showed that pungency degree of *C. annuum* var. *glabriusculum* fruits was influenced by environmental factors such as altitude, temperature and vegetation.

Keywords: Capsaicinoids; Capsaicin; Dihydrocapsaicin; *Capsicum annuum* var. *glabriusculum*.

Resumen. Se describen los análisis por HPLC del contenido de capsaicina y dihidrocapsaicina en frutos de *Capsicum annuum* var. *glabriusculum* recolectados en Tamaulipas, México. Las localidades fueron seleccionadas con base en que 65% de sus habitantes cosecha los frutos de esta especie para autoconsumo y su comercialización. Se realizó un análisis de varianza (ANOVA) para determinar la variación en la composición de capsaicinoides, y análisis multivariado mediante correlación de Pearson y componentes principales (PCA) para conocer la influencia de los factores ambientales en la concentración de éstos. Los resultados mostraron diferencias significativas en la concentración de capsaicina entre las poblaciones. Los frutos de El Gavilán tuvieron la mayor concentración de ambos alcaloides (994 µg/g) y los de Comas Altas la menor (498 µg/g). Se encontró una correlación positiva entre la concentración de capsaicina y dihidrocapsaicina, ya que conforme se incrementó la concentración de capsaicina, también aumentó la de dihidrocapsaicina. El PCA demostró que la altitud, temperatura y vegetación fueron los principales factores que influyeron en la producción de ambos capsaicinoides en *C. annuum* var. *glabriusculum*. La precipitación fue la variable que afectó en menor medida. De las cuatro poblaciones evaluadas con vegetación de matorral espinoso tamaulipeco, los frutos colectados en El Gavilán, zona ubicada a 578 msnm, presentaron la mayor concentración de ambos alcaloides; la menor en Comas Altas (65 msnm). Por otro lado, los frutos colectados en el bosque de mezquite, donde se ubica La San Juana, sintetiza también altas concentraciones de capsaicinoides debido al microclima especial generado por la vegetación, haciendo un hábitat propicio para una mayor producción de estos compuestos. En este sentido, el grado de pungencia de los frutos de *C. annuum* var. *glabriusculum* fue influenciado por factores ambientales, siendo los de mayor impacto la altitud, temperatura y vegetación.

Palabras clave: Capsaicinoides; Capsaicina; Dihidrocapsaicina; *Capsicum annuum* var. *glabriusculum*.

[1] Departamento de Ecología y Recursos Naturales, Laboratorio de Fitoquímica, Facultad de Ciencias, Universidad Nacional Autónoma de México, Ciudad Universitaria, Coyoacan, 04510 México, D.F.
[2] Posgrado en Botánica, Laboratorio de Fitoquímica, Colegio de Postgraduados, Campus Montecillo, Texcoco Estado de México 56230, México.
[3] Dirección de Investigaciones en Neurociencias del Instituto Nacional de Psiquiatría Ramón de la Fuente Muñiz, Calz. México-Xochimilco 101, Col. San Lorenzo Huipulco, 14370 México, D.F., México.
[4] Programa de Desarrollo Regional Sustentable. Instituto de Ecología Aplicada, Universidad Autónoma de Tamaulipas, Calle División del Golfo 356. Col. Libertad, 87019 Cd. Victoria Tamaulipas, México.
[5] Instituto de Investigaciones Agropecuarias y Forestales, Universidad Michoacana de San Nicolás de Hidalgo, Tarímbaro, Michoacán, México.
Address correspondence to: José Guadalupe Martínez Ávalos, Programa Forestal. Instituto de Ecología Aplicada, Universidad Autónoma de Tamaulipas, Calle División del Golfo 356. Col. Libertad, 87019 Cd. Victoria Tamaulipas, México. Phone: +52(834)3162721, *e-mail:* jmartin@uat.edu.mx ; sucoland@hotmail.com
Received 17.VIII.2015. Accepted 5.VI.2016.

INTRODUCTION

The genus *Capsicum* (Solanaceae) comprises about 30 species, *Capsicum annuum* L. is one of the most important from the economical point of view due to its long process of domestication and genetic improvement (Hernández-Verdugo et al., 1999). The variety *Capsicum annuum* var. *glabriusculum* (Dunal) Heiser & Pickersgillis is considered the ancestor of the cultivated chilli and bell pepper, *C. annuum* var. *annuum* (Pickersgill, 1971). This is the most economically important domesticated of five *Capsicum* species. *Capsicum annuum* var. *glabriusculum* is a perennial bush, its distribution range goes from Colombia to the South Western United States (Hernández-Verdugo et al., 1999). In Mexico, it can be found from the Yucatan Peninsula and the Gulf of Mexico, growing in deep soils covered with dense evergreen vegetation, to xeric regions like those found at Sonora desert and the central plateau, where it is commonly associated with nursing trees (Tewksbury et al., 1999). This variety has a high phenotypic plasticity, evidenced by the variation of traits such as leaf morphology, fruit shape, pattern of seed germination and resistance to pathogens (Hernández-Verdugo et al., 2001). In the northern states of Mexico particularly Sonora and Tamaulipas, *C. annuum* var. *glabriusculum* fruits are locally known as "chiltepín" or "chile piquín". The fruits are gathered from wild populations to use them as a spice owing their pungency (González-Jara et al., 2011; González-Zamora et al., 2015). The collection and production of "chile piquín" involves 13 municipalities of Tamaulipas: Abasolo, Aldama, Burgos, Casas, Mainero, Méndez, San Carlos, San Nicolas, Llera de Canales, Soto la Marina, Hidalgo, Victoria and Villagrán. An estimated 65% of the population from these municipalities collects both green and red fruits for consumption and trade. During the year, the cost per kilogram may fluctuate between 60.00 to 1200.00 MXN (Mexican pesos) (differential ambit= 1140) in a majority of the Mexican market (65%), although a proportion (35%) is exported to the U.S.A. (Kraft et al., 2013). Throughout the period from August to December the collection of "chile piquin" is the main activity of the people from the rural communities of Tamaulipas. The local economic structure drives whole families to depend on this resource for the improvement of their living conditions. Wild populations of "chile piquin" in Tamaulipas are under heavy anthropogenic pressure due to immoderate collection and over exploitation of the resource.

Capsaicinoids are a group of alkaloidal compounds that cause the spicy sensation (pungency) of chili pepper fruits. The main capsaicinoid is capsaicin, followed by dihydrocapsaicin, nordihydrocapsaicin, homodihydrocapsaicin, and homocapsaicin. Capsaicin and dihydrocapsaicin are predominant, accounting for almost 90% of all capsaicinoids (Singh et al., 2009; Aza-González et al., 2011; Giuffrida et al., 2013; González-Zamora, 2015). The alkaloids are abundant in *Capsicum* fruit, specifically in the external layer of the placenta, that is the tissue that holds the seeds, and under the thick and irregular epidermis. Capsaicinoids can be produced at varying amounts, depending on the genotype, developmental stage and growth conditions (Aza-González et al., 2011).

The objective of the present work was to compare the concentration of capsaicin and dihydrocapsaicin in the pericarp and placenta of fruits from *C. annuum* var. *glabriusculum* collected in different locations in Tamaulipas, Mexico.

MATERIALS AND METHODS

Plant material. Fruits of *C. annuum* var. *glabriusculum* were collected during October and November 2010 in six municipalities of Tamaulipas, Mexico: "El Gavilán", "La San Juana", "La Pasadita", "San Lázaro", "Lázaro Cárdenas" and "Comas Altas". The sites were selected according to diverse environmental factors: altitude, precipitation, ambient temperature and vegetation type of these locations (Table 1). Identification of the species was carried out by botanists from the Instituto de Ecologia Aplicada. Samples of the collected material were deposited at the herbarium of this Institute in the Universidad Autónoma de Tamaulipas (UAT).

Samples preparation. Pericarp and placenta were excised from the fruits, air dried and then milled. The samples (500 mg) were placed each in a test tube with 5 mL of high performance liquid chromatography (HPLC) grade acetonitrile, immersed in a water bath at 60 °C shaken every 30 min for 5 h. The supernatant was then sieved through a syringe filter and transferred to 2.0 mL autosampler vials.

HPLC analysis. Samples were analyzed with an HP 1100 series system (Hewlett Packard) with a Hypersil ODS C18 column (150 x 4.7 mm i.d., 5 μm particle size) maintained at 25 °C. Diode array detector in absorbance mode and 202 nm wavelength. Mobile phase consisted in 65:35, acetonitrile-buffer solution (35 mM KH_2PO_4), isocratic elution, flow rate 1.7 mL/ min. The injection volume was 20 μL.

Calibration curves of capsaicin and dihydrocapsaicin (Sigma St. Louis, Mo, U.S.A.) were obtained from at least 6 points in a range from 0.024 to 2.0 mg. The interpolations were calculated with Chem Station software (Agilent Technologies).

Statistical analysis. The capsaicinoids amounts of each location were compared by a one-way analysis of variance (ANOVA) followed by Tukey test. The multivariate analysis using Pearson correlation coefficient between different variables ($P<0.05$) and a principal component analysis (PCA) were performed. This was to discriminate between environmental factors on the basis of their locations and altitude on the concentration of capsaicin and dihydrocapsaicin in the fruit of *C. annuum* var. *glabriusculum* collected in different locations using SAS software (SAS Institute, 2002).

RESULTS AND DISCUSSION

Based on the vegetation type (INEGI, 2005; Challenger & Soberon, 2008), populations of *C. annuum* var. *glabriusculum* thrive primarily in the understory of three plant communities: The Tamaulipan thornscrub (TTS), the mesquite forest (M) and the tropical deciduous forest (TDF). The TTS is characterized by two strata: (1) one composed of shrubs and trees of a height equal or lower than four meters, and (2) another formed by herbaceous forbs and grasses shorter than one meter. The TTS covers approximately 185000 km^2 in the states of Texas in the U.S.A., and Coahuila, Nuevo Leon and Tamaulipas in Mexico (Fig. 1). In the latter state the area encloses 858520 ha (11.07%) with hilly plains and mountainous elevations reaching over 1500 m. This habitat is dominated by shrubs such as: *Celtis pallida*, *Leucophyllum frutescens, Vachelia rigidula, Vachelia berlandieri, Porlieria angustifolia, Phaulothamnus spinescens, Ziziphus obtusifolia*, and *Karwinskia humboldtiana*.

The second plant community, M, covers 179724 ha (2.31%) of Tamaulipas. Two species of thorny trees characteristically predominate: *Prosopis glandulosa* and *P. tamaulipana*. During the drought season, this vegetation type remains green because of its deep root system. It can be found alongside drains or runoffs (known as ramaderos) that remain dry most of the year.

Finally, TDF is the most widespread tropical vegetation type in Mexico (Trejo & Dirzo, 2002), and considered one of the most diverse environments accounting for a high number of endemic vascular flora. It covers 556229 ha (7.17%) of Tamaulipas, encompassing a vast part of the Sierra de Tamaulipas and various mountain areas in the Sierra Madre Oriental. A recent study estimated that more than 100 species of woody plants are present in the tropical deciduous forest of southern Tamaulipas, forming the tree and shrub layer in turn composed by a few plant families. Amongst the commonly found species are: *Drypetes lateriflora*, *Ebenopsis ebano*, *Randia latevirens* and *Ocotea tampicensis* (Ascencio-Garcia & Martinez-Avalos, 2010).

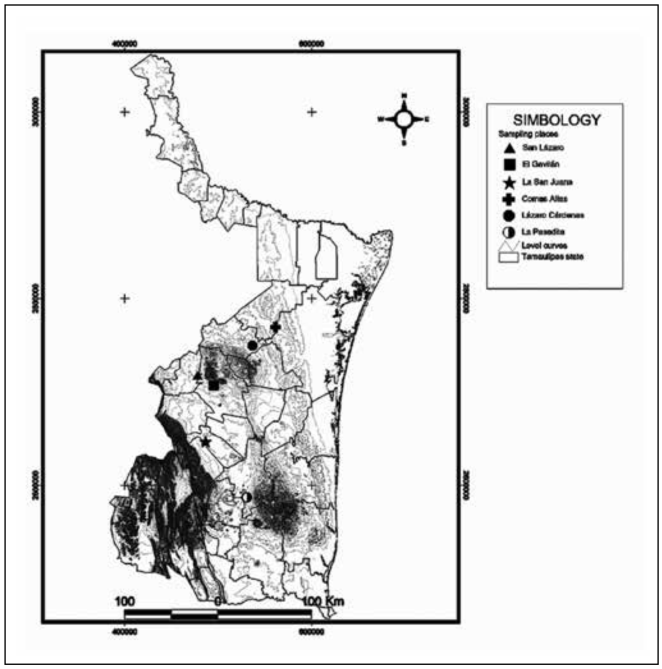

Fig. 1. Location of the six populations of *Capsicum annuum* var. *glabriusculum* collected in Tamaulipas, Mexico.
Fig. 1. Ubicación de las seis poblaciones de *Capsium Annuum* var. *glabriusculum* recolectadas en Tamaulipas, México.

The collection sites for *C. annuum* var. *glabriusculum* has the previously described vegetation types (TTS, M and TDF), its latitude range from 23° 23' 37.03" to 25° 02' 25.85" N, longitude from 98° 23' 32.19" to 99° 13' 07.75" W, altitude from 65 to 578 m a.s.l. The precipitation regime is 698 ± 69 mm, and the mean annual temperatures go from 22.5 to 23.5 °C (Table 1). The lowest altitude belongs to "Comas Altas" with 65 m a.s.l. and the highest altitude to "El Gavilán" with 578 m a.s.l. Regarding precipitation, the lowest annual meanis recorded

Table 1. Climatic conditions and vegetation type at the colecting sites of *C. annuum* var. *glabriusculum* in Tamaulipas, Mexico.
Tabla 1. Condiciones climáticas y tipo de vegetación en los sitios de recolección de *C. annuum* var. *glabriusculum* en Tamaulipas, México.

Population	Municipalities	Latitude (N)	Longitude (W)	Altitude (m)	Precipitation (mm)	Annual Mean Temperature (°C)	Vegetation type
El Gavilán	San Carlos	24° 28' 20.53"	99° 02' 49.25"	578	545	22.5	TTS
San Lázaro	Villagrán	24° 34' 17.92"	99° 13' 07.75"	336	500	22.5	TTS
La Pasadita	Casas	23° 23' 37.03"	98° 41' 57.30"	297	896	22.5	TDF
La San Juana	Güémez	23° 56' 04.56"	99° 07' 57.86"	199	675	23.5	M
Lázaro Cárdenas	Burgos	24° 51' 38.55"	98° 38' 13.77"	172	896	22.5	TTS
Comas Altas	Méndez	25° 02' 25.85"	98° 23' 32.19"	65	675	22.5	TTS

TTS = Tamaulipan thornscrub
M = Mezquite forrest
TDF = Tropical deciduous forest (INEGI, 2005).

in "San Lázaro" and the highest mean in "La Pasadita" and "Lázaro Cárdenas" (Table 1).

In the present study, capsaicinoid concentrations in fruits were significantly different between all collection sites, from high to low: "El Gavilán", "La San Juana", "La Pasadita", "San Lázaro", "Lázaro Cárdenas" and "Comas Altas" (Fig. 2). The capsaicin and dihydrocapsaicin amounts (57% and 217%, respectively) in samples were higher in "El Gavilán" than in "Comas Altas". The last one had the lowest values, and therefore it was considered as the basal reference (Fig. 2). According to the Pearson coefficient, there was a close positive correlation between the concentration of capsaicin and dihydrocapsaicin ($r = 0.93$, $P<0.0001$) (Table 2, Fig. 2), indicating that high amounts of capsaicin were associated with high amounts of dihydrocapsaicin, and viceversa. In our study, capsaicin had the highest concentration, agreeing with the results of González-Zamora et al. (2015). Further, other authors refer to a 1:1 capsaicin:dihydrocapsaicin ratio as common, or 2:1 in very hot fruits (Higashiguchi et al., 2006; Cisneros-Pineda et al., 2007; Garces-Claver et al., 2007; Singh et al., 2009).

It can be noted that the sampling sites have geographic and environmental attributes in concordance for the optimal development of this wild variety of C. *annuum* as well as a characteristic vegetation type. On the topic of capsaicinoids,

Table 2. Pearson correlation matrix between capsaicinoids concentrations and climate for collections sites of *C. annuum* var. *glabriusculum.*
Tabla 2. Matriz de correlación de Pearson entre concentraciones de capsaicinoides y clima para los sitios de recolección de *C. annuum* var. *glabriusculum*.

Variable 1	Variable 2	Pearson	P-value
Capsaicin	Dihydrocapsaicin	0.93	< 0.0001
Capsaicin	Altitude	0.68	0.0019
Capsaicin	Precipitation	-0.39	0.1061
Capsaicin	Temperature	0.56	0.0156
Dihydrocapsaicin	Altitude	0.83	< 0.0001
Dihydrocapsaicin	Precipitation	-0.50	0.0329
Dihydrocapsaicin	Temperature	0.27	0.2727
Altitude	Precipitation	-0.46	0.0576
Altitude	Temperature	-0.21	0.4051
Precipitation	Temperature	-0.07	0.7936

n = 18.

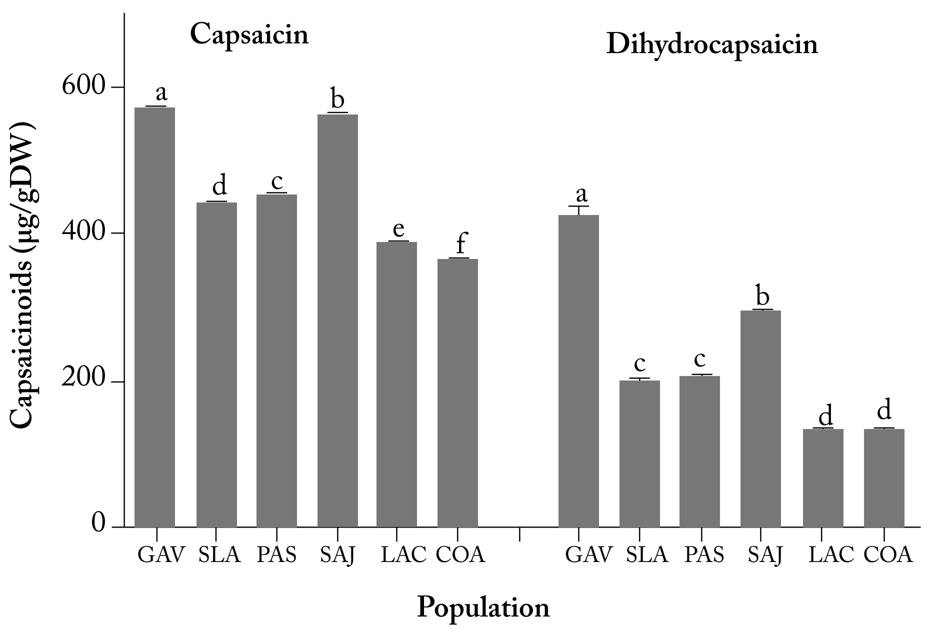

Fig. 2. Concentration of capsaicin and dihydrocapsaicin in fruits of *C. annuum* var. *glabriusculum* from samples collected in different regions of Tamaulipas, Mexico (GAV = El Gavilán; SLA = San Lázaro; PAS = La Pasadita; SAJ = La San Juana; LAC = Lázaro Cárdenas, and COA = Comas Altas). The values expressed represent means ± standard error (S.E.); means followed by different letters within either Capsaicin or Dihydrocapsaicin differed significantly ($P<0.05$) from each other.
Fig. 2. Concentración de capsaicina y dihidrocapsaicina en frutos de *C. annuum* var. *glabriusculum* de muestras recolectadas en diferentes regiones de Tamaulipas, México (GAV = El Gavilán; SLA = San Lázaro; PAS = La Pasadita; SAJ = La San Juana; LAC = Lázaro Cárdenas, and COA = Comas Altas). Los histogramas representan la media ± 1 E.E.; los promedios seguidos por letras diferentes dentro de capsaicin o dihydrocapsaicin difieren significativamente (P<0,05).

our results are based on the PCA analysis which showed a relation between altitude, temperature and vegetation in the populations studied regarding of the concentration of capsaicine and dihidrocapsaicine (Fig. 3 A y B). This analysis allowed the formation of five groups arising from the populations where the recollections of fruits of *C. annum* were made. Both principal components (PC1 and PC2) explain the total variation of 86.4 found in populations from Lázaro Cárdenas and Comas Altas. They are a very close group as they showed the lower capsaicine and dihidrocapsaicine concentration as well as temperature and vegetation type. Both of these populations are temperature related with San Lázaro and La Pasadita. However, the later localities form different groups for vegetation and altitude, respectively. Finally, locations El Gavilán and La San Juana which showed the higher concentrations for both alkaloids form separated groups, the first due to altitude differences and the second due to temperature and vegetation variables (Cu 1, Fig. 3 A y B).

Regarding altitude, the highest amount of capsaicinoids in fruits was found in samples from "El Gavilán" at an altitude of 578 m.a.s.l. and the lowest at "Comas Altas" at 65 m.a.s.l. (Table 1, Fig. 2). Concerning this factor, it is only mentioned that wild populations of *C. annuum* grow well in low altitude sites, seldom more than 1000 m (Hernández-Verdugo et al., 2001). It is noteworthy that this is the first report of the influence of altitude in the amounts of capsaicinoids in this hot pepper variety. Previous works on the variation of alkaloid amounts of other plant species mentioned that it is very important to consider altitude, temperature, light availability and precipitation (Ralphs & Gardner, 2001; Varma et al., 2011).

In addition to the mentioned physical environment components, extreme conditions exist (stress) that also affect the pungency of pepper: these are high mean temperatures, and excessive or insufficient watering (Saha et al., 2010; Ruiz-Lau et al., 2011). In this study, capsaicin amounts were significantly correlated with ambient temperature, while dihydrocapsaicin was not correlated to that factor (Table 2). Such is the case for the population of La San Juana, the only locality with the higher mean annual temperature value (23.5 °C) and the higher concentration of capsaicine after the El Gavilán (Table 1, Fig. 2). These results are in concordance with the information mentioned by Cisneros-Pineda et al. (2007) referring that as a result of the temperature rise, capsaicinoids amounts increase more in hot than in sweet pepper varieties. Moreover, González-Zamora et al. (2013) mention that high temperatures in other pepper varieties (e.g. arbol and jalapeño) might be a negative factor in the accumulation of capsaicinoids. Mean annual temperature was 22.5 *°C* in most collection sites of this study. Research showed that chili plants grow well in tropical regions and require high temperatures for its development, with an optimal between 25 to 30 °C. Therefore wild chillies are more tolerant to high temperatures (González-Zamora et al., 2013).

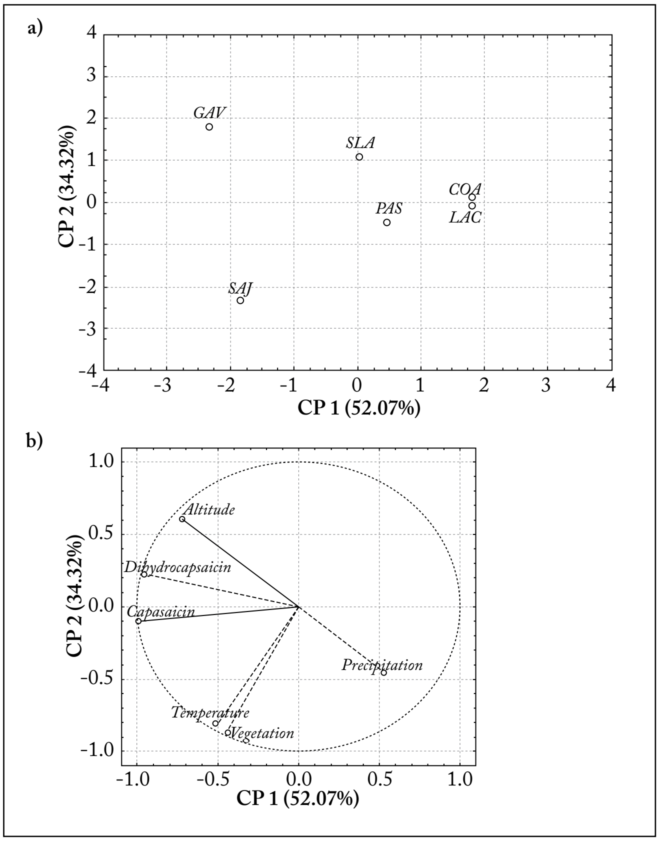

Fig. 3. Principal component analysis, based on the concentration of capsaicin and dihydrocapsaicin, to discriminate between (a) population and (b) environmental factors. The variation explained by the axes was 86.39%. Codes of the populations from Tamaulipas, Mexico were GAV = El Gavilán; SLA = San Lázaro; PAS = La Pasadita; SAJ = La San Juana; LAC = Lázaro Cárdenas, and COA = Comas Altas.

Fig. 3. Análisis de componentes principales basado en la concentración de capsaicina y dihyrocapsaicina para discriminar entre los factores (a) población y (b) ambientales. La variación explicada por los ejes fue 86,29%. Los códigos de las poblaciones de Tamaulipas, México fueron: GAV = El Gavilán; SLA = San Lázaro; PAS = La Pasadita; SAJ = La San Juana; LAC = Lázaro Cárdenas, and COA = Comas Altas.

Regarding the effect of soil humidity, previous studies have demonstrated that capsaicinoids amount rise under drought conditions (Ruiz-Lau et al. 2011; Phimchan et al., 2012; Giuffrida et al., 2013; Barbero et al., 2014). Principal component analysis from this investigation showed that altitude, temperature and vegetation type are factors that have a positive influence on the concentrations of capsaicine and dihidrocapsaicine and that rainfall was the less important variable. Altitude was the most important variable for the locality El Gavilán, and temperature and vegetation for the locality La San Juana (Fig. 3 A y B).

Variation of concentration of capsaicinoids in the collecting sites of *C. annuum* could be explained by the climatic differences from habitat, that is altitude, and microclimatic conditions since climate form each locality was different. Localities El Gavilán and La San Juana were the most contrasting for alkaloids concentration, and are clearly different from the other localities (Fig. 3 A). The locality El Gavilán has a TTS vegetation, and the mean annual temperature was 22.5 °C, rainfall remained at 545 mm, and altitude was 578 m.a.s.l. The locality La San Juana had an M vegetation, and a mean annual temperature of 23.5 °C, a mean rainfall of 675 mm and an altitude of 199 m.a.s.l. (Table 1). The TTS vegetation community had trees and shrubs of approximately 4 m tall and 1 m tall herbs, whereas the M vegetation had a habitat with *Prosopis* sp. trees. They have a great ecological importance due to the fact that they fostered a microhabitat with shadow and food for other species. These differences in vegetation might be responsible for the higher concentration of alkaloids in fruits from the locality La San Juana. Regarding these literature reports the incidence of solar radiation on the plant canopy is critical for the photosynthetic activity, specifically the production of carbon skeletons that serve as construction blocks for the molecules synthesized by the secondary metabolism. In nature, plants thrive where its solar radiation demands are fulfilled. In some cases light is a contributing factor that determines the amount of alkaloids produced. In the case of wild peppers, or chiltepines, the plants are not generally found under direct sunlight, but distributed under the canopy of diverse nurse plants that generate micro environmental differences. Therefore, the distribution of these peppers was closely related to the biotic and abiotic factors of the growing site (Tewksbury et al., 1999; González-Zamora et al., 2015).

Finally, given that the synthesis of a secondary metabolite originates from genetic expression, it can be expected that its presence or absence is subordinated to the factors which induce or inhibit the genes expression. Some of the main inducers of the synthesis of secondary metabolites are factors like climate, soil, plant-plant and plant-herbivore interactions (Moore et al., 2014). Various studies have evaluated the importance of the environment in the production of plant secondary metabolites (Kroymann, 2011; Külheim et al., 2011; Mithöfer & Boland, 2012). For the particular case of capsaicin and dihydrocapsaicin in *Capsicum* species, it has been demonstrated that the concentration variation was related to the species and environmental conditions (Cisneros-Pineda et al., 2007; Aza-González et al., 2011; González-Zamora et al., 2015). In some *Capsicum* species, the capsaicinoids amounts are directly affected also by the harvest period, water availability, light and ambient temperature (Harvell & Bosland, 1997; Zewdie & Bosland, 2000; Garces-Claver et al., 2007; Giuffrida et al., 2013).

CONCLUSIONS

Our results showed that there was a variability in the concentration of capsaicine and dihidrocapsaicine in the wild chilli peppers studied from the different sampled localities. The statistical analysis confirmed that altitude (336 -578 m), temperature and vegetation were the main factors involved in the concentration of these alkaloids. Results also showed that it was possible to determine the best localities for the production of populations of *C. annuum* var. *glabriusculum* with more or less pungency content in the state of Tamaulipas. This might help to propitiate the legal local and national trade to foster the production and participate in the international market. For example, products from sites with conditions favoring high pungency could be selected to be locally and regionally sold, while others from sites associated with low pungency could meet the requirements of international markets such as the Asian, American or European ones.

ACKNOWLEDGEMENTS

The authors thank Roberto Enrique Llanos-Romero, Rodolfo Martínez Gallegos and Vilma Paullette Ascencio García for their technical assistance.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

REFERENCES

Ascencio-García, V. & J.G. Martínez-Ávalos (2010). Diversidad y riqueza de árboles de la selva baja caducifolia de Tamaulipas, México. Primer Encuentro Estudiantil de Investigación "Forjando talentos universitarios en el desarrollo de las ciencias" Universidad Autónoma de Tamaulipas, Cd. Victoria, Tamaulipas, México. pp. 111-114.

Aza-González, R., H.G. Núñez-Palenius & N. Ochoa-Alejo (2011). Molecular biology of capsaicinoids biosynthesis in chili pepper (*Capsicum* spp.). *Plant Cell Reports* 30: 695-706.

Barbero, G.F., A.G. Ruíz, A. Liazid, M. Palma, J.C. Vera & C.G. Barroso (2014). Evolution of total and individual capsaicinoids in peppers during ripening of the Cayenne pepper plant (*Capsicum annuum* L.). *Food Chemistry* 153: 200-206.

Challenger, A., & J. Soberón (2008). Los ecosistemas terrestres. In: Comisión Nacional para el Conocimiento y Uso de la Biodiversidad (eds.), pp. 87-108. Capital natural de México, vol. I: Conocimiento actual de la biodiversidad. CONABIO, México, D.F. 108 p.

Cisneros-Pineda, O., L.W. Torres-Tapia, L.C. Gutiérrez-Pacheco, F. Contreras-Martín, T. González-Estrada & S.R. Peraza-Sánchez (2007). Capsaicinoids quantification in chili peppers cultivated in the state of Yucatan, Mexico. *Food Chemistry* 104: 1755-1760.

Garcés-Claver, A., R. Gil-Ortega, A. Álvarez-Fernández & M.S. Arnedo-Andrés (2007). Inheritance of capsaicin and dihydrocapsaicin, determined by HPLC-ESI/MS, in an intraspecific cross of *Capsicum anuum* L. *Journal of Agricultural and Food Chemistry* 55: 6951-6957.

Giuffrida, D., P. Dugo, G. Torre, C. Bignardi, A. Cavazza, C. Corradini & G. Dugo (2013). Characterization of 12 *Capsicum* varieties by evaluation of their carotenoid profile and pungency determination. *Food Chemistry* 140: 794-802.

González-Jara, P., A. Moreno-Letelier, A. Fraile, D. Piñero & F. García-Arenal (2011). Impact of human management on the genetic variation of wild pepper, *Capsicum annuum* var. *glabriusculum*. *PLoS ONE* 6: 1-11.

González-Zamora, A., E.Sierra-Campos, J. G. Luna-Ortega, R. Pérez-Morales, J.C.R. Ortiz & J.L. García-Hernández (2013**).** Characterization of different *Capsicum* varieties by evaluation of their capsaicinoids content by High Performance Liquid Chromatography, determination of pungency and effect of high temperature. *Molecules* 18: 13471-13486.

González-Zamora, A., E. Sierra-Campos, R. Pérez-Morales, C. Vázquez-Vázquez, M.A. Gallegos-Robles, J.D. López-Martínez & J.L. García-Hernández (2015). Measurement of capsaicinoids in chiltepín hot pepper: a comparison study between spectrophotometric method and high performance liquid chromatography analysis. *Journal of Chemistry* 2015: 1-10.

Harvell, K.P. & P.W. Bosland (1997). The environment produces a significant effect on pungency of chiles. *Hort Science* 32: 1292.

Hernández-Verdugo, S., P. Dávila & K. Oyama (1999). Síntesis del conocimiento taxonómico, origen y domesticación del género *Capsicum*. *Boletín de la Sociedad Botánica de* México 62: 171-181.

Hernández-Verdugo, S., R. Luna-Reyes & K. Oyama (2001). Genetic structure and differentiation of wild and domesticated populations of *Capsicum annuum* (Solanaceae) from Mexico. *Plant Systematics and Evolution* 226: 129-142.

Higashiguchi, F., H. Nakamura, H. Hayashi & T. Kometani (2006). Purification and structure determination of glucosides of capsaicin and dihydrocapsaicin from various *Capsicum* fruits. *Journal of Agricultural and Food Chemistry* 54: 5948-5953.

INEGI (Instituto Nacional de Estadística, Geografía e Informática) (2005). Carta de uso actual del suelo y vegetación. Serie III. México.

Kraft, K.H., J.J. Luna-Ruiz & P. Gepts (2013). A new collection of wild populations of *Capsicum* in Mexico and the southern United States. *Genetic Resources and Crop Evolution* 60: 225-232.

Kroymann, J. (2011). Natural diversity and adaptation in plant secondary metabolism. *Current Opinion in Plant Biology* 14: 246-251.

Külheim, C., S.H. Yeoh, I.R. Wallis, S. Laffan, G.F. Moran & W.J. Foley (2011). The molecular basis of quantitative variation in foliar secondary metabolites in *Eucalyptus globules. New Phytologist* 191: 1041-1053.

Mithöfer, A. & W. Boland (2012). Plant defense against herbivores: chemical aspects. *The Annual Review of Plant Biology* 63: 431-450.

Moore, B.D., R.L. Andrew, C. Külheim & W. Foley (2014). Tansley review: Explaining intraspecific diversity in plant secondary metabolites an ecological context. New *Phytologist* 201: 733-750.

Phimchan, P., S. Techawongstien, S. Chanthai & P.W. Bosland (2012). Impact of drought stress on the accumulation of capsaicinoids in *Capsicum* cultivars with different initial capsaicinoid levels. *Hort Science* 47: 1204-1209.

Pickersgill, B. (1971). Relationships between weedy and cultivated forms in some species of chili peppers (genus *Capsicum*). *Evolution* 25: 683-691.

Ralphs, M.H. & D.R. Gardner (2001). Aldaloid levels in Duncecap (*Delphinium occidentale*) and tall larkspur (*D. barbeyi*) grown in reciprocal gardens: separating genetic from environmental influences. *Biochemical Systematics and Ecology* 29: 117-124.

Ruiz-Lau, N., F. Medina-Lara, Y. Minero-García, E. Zamudio-Moreno, A. Guzmán-Antonio, I. Echevarría-Machado & M. Martínez-Estévez (2011). Water deficit affects the accumulation of capsaicinoids in fruits of *Capsicum chinense* Jacq. *Hort Science* 46: 487-492.

Saha, S.R., M.M. Hossain, M.M. Rahman, C.G. Kuo & S. Abdullah (2010). Effect of high temperature stress on the performance of twelve sweet pepper genotypes. *Bangladesh Journal of Agricultural Research* 35: 525-534.

Singh, S., R. Jarret, V. Russo, G. Majetich, J. Shimkus, R. Bushway & B. Perkins (2009). Determination of capsinoids by HPLC-DAD in *Capsicum* species. *Journal of Agricultural and Food Chemistry* 57: 3452-3457.

Tewksbury, J.J, P.G. Nabhan, D. Norman, H. Suzán, J. Tuxil & J. Donovan (1999). *In situ* conservation of wild chiles and their biotic associates. *Conservation Biology* 13: 98-107.

Trejo, I. & R. Dirzo (2002). Floristic diversity of Mexican seasonally dry tropical forest. *Biodiversity and Conservation* 11: 2063-2084

Varma, A., H. Padh & N. Shrivastava (2011). Ecogeographical phytochemistry of *Adhatoda vasica* Nees in relation to quantitative variation of alkaloids. *Journal of Planar Chromatography* 24: 406-411.

Zewdie, Y. & P. Bosland (2000). Evaluation of genotype, environment, and genotype-by environment interaction for capsaicinoids in *Capsicum annuum* L. *Euphytica* 111: 185-190.