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# Diversity and traditional knowledge of pulque agave landraces in the community of San Pedro Tlalcuapan, Tlaxcala, Mexico

# Diversidad y conocimiento tradicional de agaves pulqueros en la comunidad de San Pedro Tlalcuapan, Tlaxcala, México

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#### Abstract

There is scarce information available about how peasants recognize *Agave* landraces. This study aims to identify the agave species and landraces used for "pulque" production and analyze their morphological variability in a Nahua community, San Pedro Tlacuapan (SPT), Tlaxcala, Mexico. To this end, we used traditional knowledge and conventional statistical methods. Semi-structured interviews were administered; from these, the frequency of mentions, lexical marking, and perception about each landrace were evaluated. A total of 36 morphological vegetative characters were measured in the main landraces, and ordination analyses (PCA) were performed. SPT is home to 8 landraces of *Agave americana* and *Agave salmiana* subsp. *salmiana*. "Manso" was the most frequently mentioned landrace, and 5 qualitative characters used by local experts to identify landraces were observed. One morphological cluster was associated with each landrace analyzed, and the variables characterizing them are rosette size, leaf shape, number of leaves, and tooth shape. The information about agave diversity from local experts can be integrated into measurements of morphological diversity through conventional statistical methods.

Keywords: Aguamiel; Morphological diversity; Native people; Cultural significance

### Resumen

Poco se ha descrito sobre la manera en que los campesinos reconocen las variedades tradicionales de *Agave*. El objetivo de este estudio fue identificar las especies y sus variedades tradicionales utilizadas para la producción de pulque y analizar su variabilidad morfológica, a través del conocimiento tradicional y de métodos estadísticos convencionales en una comunidad Nahua, San Pedro Tlacuapan (SPT), Tlaxcala, México. Se realizaron entrevistas

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semiestructuradas y se evaluó la frecuencia de mención, el marcaje léxico y la percepción. Se midieron 36 caracteres morfológicos vegetativos en las variedades tradicionales más significativas y se realizaron análisis de ordenación (PCA). SPT presenta 8 variedades tradicionales de *Agave americana* y *Agave salmiana* subsp. *salmiana*. "Manso" fue la variedad tradicional con mayor mención y se observaron 5 caracteres cualitativos que los expertos tradicionales utilizan para identificar las variedades tradicionales. Se asocia un grupo morfológico por cada variedad tradicional analizada y las variables que los caracterizan son: el tamaño de la roseta, la forma y el número de hojas y la forma de los dientes. Tanto el reconocimiento de la diversidad por parte de los campesinos, como la medición de la diversidad morfológica por métodos estadísticos tradicionales, es complementario y se puede integrar.

Palabras clave: Aguamiel; Diversidad morfológica; Pueblos originarios; Importancia cultural

#### Introduction

Biodiversity results from multiple historical and abiotic factors, ecological interactions, and evolutionary processes (Futuyma, 2013). Some living organisms, such as plants, have been of interest to humans; one result of this interaction is that humans have constantly used and selected them. These actions have driven diversity in cultivated or exploited plants through manipulation and artificial selection as part of the evolutionary process of domestication (Pickersgill, 2007; Zeder et al., 2006). One consequence of domestication is morphological diversity, the raw material on which peasants rely for selecting plant varieties because morphological features are conspicuous. When artificial selection fixes some characters, making morphological differences noticeable, peasants usually assign different names to denote groups of plants with characteristics (Berlin, 1992; Pickersgill, 2007; Zeder et al., 2006). These local names may correspond to landraces. Theoretically, landraces are cultivated plants that differ from their wild relatives by having adaptations that allow them to grow outside of their natural habitat; landraces can also show morphological changes known as "domestication syndrome", which reflects artificial selection (Casañas et al., 2017; Pickersgill, 2007).

Mexico is a diversity center of the genus *Agave* L. since it is home to 79% of the species, most being endemic to the country (García-Mendoza, 2011). In addition, the diversity of the genus *Agave* is also due to the use, manipulation, and human selection of agave plants (Eguiarte et al., 2021; Gentry, 1982). Humans have been using them for at least 10,000 years; virtually the whole plant is harnessed, and more than 92 uses have been reported (Colunga-GarcíaMarín et al., 2017; Trejo et al., 2022). The uses of agave include the elaboration of distilled spirits such as tequila or fermented beverages such as "pulque". The latter is a traditional beverage obtained from the fermentation of *Agave* sap ("aguamiel") that has been prepared since pre-Hispanic times that has recently regained relevance. The state of Tlaxcala ranks second in

Mexico in "pulque" agave and "aguamiel" production, after the state of Hidalgo (SIAP, 2018).

Studies on the morphological diversity of agave plants have been based on measuring all possible morphological —mainly vegetative—characteristics. Reproductive traits are hard to observe because the floral scape is removed by peasants for the consumption of flowers and the floral scape but mainly to stimulate the accumulation of sugars in the stem in order to produce sap (Figueredo-Urbina, Álvarez-Ríos, & Zárraga-Cortés, 2021). Little research has been conducted on how peasants identify their landraces (Colunga-GarcíaMarín & May-Pat, 1993; Ramírez-Manzano et al., 2020). Traditional knowledge can be essential in understanding agave diversity and its management and selection processes.

The present work explored the diversity of agave plants in the Nahuatl locality of San Pedro Tlalcuapan, Tlaxcala, focusing on the following objectives: *i)* identify species, subspecies, and landraces; *ii)* characterize the morphological attributes used by peasants to identify landraces; *iii)* integrate conventional statistical and traditional-knowledge approaches to supplement the description of agave diversity in the study area.

### Materials and methods

The study was conducted in San Pedro Tlalcuapan (SPT), a Nahuatl indigenous community located northwest of La Malinche National Park (PNLM, for its acronym in Spanish) (19°16′50.02" N, 98°09′06.30" W), in the municipality of Chiautempan, state of Tlaxcala, Mexico (Fig. 1). SPT is located at an elevation of 2,411 m, with annual mean precipitation and temperature values of 901 mm and 15.7 °C that characterize a subhumid temperate climate C(w<sub>1</sub>) (García, 1988). The vegetation is made up of coniferous forest (*Abies* and *Pinus*) and mixed (*Pinus-Quercus* and *Quercus-Pinus*) temperate forests under different levels of conservation, as well as induced pastures and seasonal crops alternating with rows of agave plants, locally known as "metepantles" (INEGI, 2010).



Figure 1. Geographic representation of the Nahuatl indigenous community of San Pedro Tlalcuapan, municipality of Chiautempan, Tlaxcala, Mexico.

The name 'Tlalcuapan' derives from the Nahuatl ("tlali" - land, "cuapan" - header), which means "at the head of the earth" or "on the slopes of the Malintzi". San Pedro Tlalcuapan has a population of 3,613 inhabitants living in 837 households: of these, 1,313 (36.34%) inhabitants are indigenous persons with a medium marginalization level (Secretaría de Bienestar, 2010), 45% are between 20 and 59 years old and 10% are over 60 years old (INEGI 2010).

Previously, the local economy was based almost entirely on forest natural resources, including mushrooms, plants, and animals. This relationship led to the acquisition of knowledge, practices, and other cultural elements. One of the occupations is "tlachiquero" (subfix-ero; "tlachique" - the one who scrapes), the person who knows how to extract sap from agave plants; "pulque" has been produced since at least 500 years ago. Currently, there is a decreasing trend in the abundance of forest natural resources, along with disinterest in the ancestral culture and changes in traditional knowledge, likely associated, partially with the degradation of the local ecosystems (Bello-Pérez, 1994; Bello & Pérez, 2019).

To address this situation, community members and local authorities have focused on revaluing, reaffirming, and supporting their biocultural heritage conservation and fostering sustainable development. Tlalcuapan is a community governed under a 'uses-and-customs' regime, i.e., it is ruled by its own laws and regulations.

The highest authority is the community assembly, which has maintained autonomous government and resource management (Bello-Pérez, 1994; Bello & Pérez, 2019).

Identification of landraces through traditional knowledge

Interviews were administered to agave producers, and field trips were conducted with them from July to December 2016 in SPT, following the guidelines of the Code of Ethics for Ethnobiological Research in Latin America (Argueta et al., 2016). The support of some members and community representatives allowed us to approach the local authorities and persons familiar with the management of agave plants. These persons were informed about the objectives and activities to be performed, requesting their support and participation in the research.

A total of 10 semi-structured interviews were administered, and 3 field trips were conducted. Five interviews were applied to "pulque" producers ("tlachiqueros") and 5 to non-"tlachiqueros". "Pulque" producers were selected through a non-probabilistic sampling known as the snowball method (Bernard, 2006), which consists of locating 1 or 2 persons who lead to others, and so on. All the local experts were interviewed since few persons currently work as "tlachiqueros". Three visits were conducted based on the availability of these experts; each guided us through their land and showed us the different agave landraces.

During the interviews, 3 variables were included following Turner (1988): frequency of mentions (number of times a group of people mentions a plant), lexical marking (naming a plant taxon; this name does not vary across groups of people when the plant is of great cultural importance), and perception (including factors such as plant size or attributes that confer notoriety to agave landraces in a locality, including leaves, flowers, and fragrance, among the characters of importance for identification).

The qualitative data collected were sorted into 2 categories: common name mentioned and plant morphology. This information was analyzed using descriptive statistics, percentage of each Agave landrace recorded, and morphological characters used to identify landraces. Data matrices were built for the frequency of mentions using a binary encoding (1 = mentioned; 0 =not mentioned). In the case of the morphology category, descriptive statistics were applied only to the 3 landraces with the highest frequency of mentions, which coincided with the most abundant ones ("Manso", "Prieto", and "Amarillo"). The perception analysis considered all terms used by peasants to describe the morphological traits of landraces; the result was simple adjectives not based on any parameter established in official metrics or tables. Three categories were assigned for plant size and rosette length, 3 categories were assigned: large = 3, medium = 2, small = 1; 2 for rosette width: thick = 2, thin = 1; 2 for spine stiffness (terminal spine): flexible = 2, stiff = 1; and 3 for plant color: dark green = 3, yellow = 2, ash green = 1.

The common names (landraces) used by local experts to identify the agave varieties were recorded; subsequently, the scientific name of each variety was determined using taxonomic keys (Gentry, 1982; García-Mendoza, 2011). In addition, leaves were collected to prepare herbarium specimens deposited at the National Herbarium of Mexico, MEXU, UNAM, and the Herbarium of Universidad Autónoma de Tlaxcala TLXM, UATx.

## Morphological diversity between landraces

To explore the morphological diversity of the agave landraces used by the community of study, adult plants at least 8 years old were selected from the most abundant landraces located in "metepantles" (rows of agave plants alternating with rows of other cultivated species) grown in corn, broad bean, and common bean plots. Since less than 3 individuals were found for most landraces, we measured 25 individuals of "Manso" and "Prieto" and 11 of "Amarillo". Thirty-two morphological characters (https://zenodo.org/record/3976297#.Xy35gChKjIU) selected according to Gentry (1982), Colunga-GarcíaMarín and May-Pat (1996), and Vargas-Ponce et al. (2007) were recorded for each plant; 25 were quantitative and 7, qualitative.

Measurements were made with a vernier for small attributes (mm), a tape measure for larger attributes (cm, m), and a micrometer (to measure the cuticle). Color was recorded using the Munsell color chart (1991), and samples of whole leaves were collected to obtain morphometric data. Only vegetative characters were considered due to the absence of plants with reproductive structures.

Qualitative multistate characters were transformed by assigning an ordinal function; a basic data matrix consisting of 61 rows (individuals) and 32 columns (morphological characters) was built using the collected information. Morphological character data were standardized; then, a correlation analysis was run to identify highly correlated variables and exclude them from the matrix. After the correspondence analysis, only 11 characters remained. A similarity matrix was generated using the Euclidean distance and a dendrogram of morphological distances was built with the farthest neighbor method to assess the similarity between specimens of the 3 local landraces. Subsequently, a principal component analysis (PCA) was carried out to arrange the specimens according to their morphological characteristics and identify redundant variables and those with the highest variation that dominated the arrangement.

Finally, a multivariate analysis of variance was performed using the values of the 11 morphological variables, followed by a Tukey's test to identify those variables with statistically significant differences between landraces. All statistical analyses were performed in the R platform (R Core Team, 2016).

## Results

In the locality of SPT, 2 species and 8 landraces of agave are recognized. Among these, *Agave americana* L. landraces "Palma" and "Cenizo" are identified by interviewees as introduced, whereas *Agave salmiana* Otto ex Salm-Dyck subsp. *salmiana* landraces "Amarillo", "Chino", "Mahuahuac", "Manso", "Matecon", and "Prieto" are considered native to the locality (Fig. 2).

Recognition of landraces through traditional knowledge

The most frequently mentioned landraces are those with the highest abundance in the area. "Manso" ranks first in frequency of mentions, being the most abundant landrace in the area. "Amarillo" ranks second despite being less abundant than "Prieto", which is in third place (Fig. 3).

On the lexical marking, it was found that some landraces are named in Nahuatl because San Pedro Tlalcuapan is a community that still preserves indigenous traditions. This is the case of the following landraces: "Manso" - Teometl (teotl = deity, metl = agave), "Amarillo" - Cosmetl (costic

# Agave salmiana subsp. salmiana

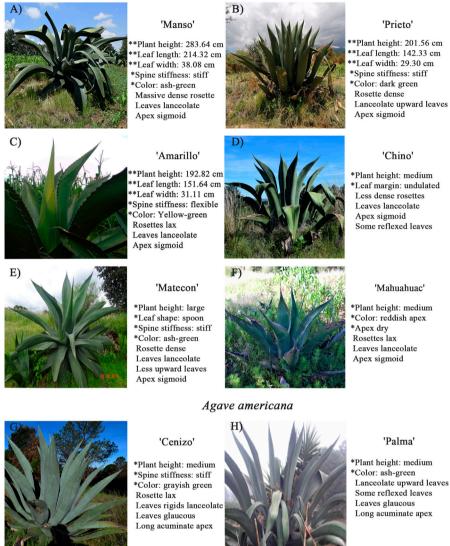


Figure 2. "Local varieties" (landraces) of *Agave* in San Pedro Tlalcuapan. *Agave salmiana* subsp. *salmiana*: A) "Manso", B) "Prieto", C) "Amarillo", D) "Mahuahuac", E) "Chino", G) "Matecon". *Agave americana*: F) "Palma" y H) "Cenizo". The characters marked with (\*) are used by peasants to recognize landraces; the characters marked with (\*\*) are used by peasants and supported with average morphological data from statistical analyses. Data with no asterisks correspond to taxonomic observations.

= yellow, metl = agave), "Prieto" - Xilumetl (xilutl = tender, metl = maguey), "Cenizo" - Nexmetl (nextic = ash-colored, metl = maguey), and "Chino" - metl chinte (metl = agave, chinte = Chinese).

Finally, regarding perception, we observed 7 qualitative characters used by experts to identify traditional varieties (landraces). According to the analyses, 5 qualitative characters have the highest number of mentions to identify

the 3 most abundant landraces of *Agave salmiana* subsp. *salmiana* (Fig. 2). However, specific characters are also used for identifying each landrace. "Manso" is determined by peasants as a large agave, more prominent than "Prieto" and "Amarillo". It has long, thick leaves and stiff spines, and the color of the plant is ash-green. "Amarillo" is identified as being smaller than "Manso" but larger than "Prieto", with medium-sized thin leaves. Its distinctive

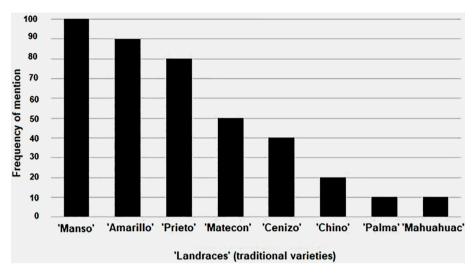


Figure 3. Frequency of mentions of landraces or local varieties in San Pedro Tlalcuapan.

traits are its flexible spines and yellowish-green leaves. "Prieto" is described as a small-sized agave with thin, small, dark-green leaves and stiff spines.

Today, all the landraces reported in SPT are used mainly for the manufacture of "pulque", but in the past these plants were also used for the extraction of a range of materials, including fibers, "mixiote", and soap, among others; in addition, agave played a role in the protection of cropland and the delimitation of parcels. These uses and the discontinuation of some of them may explain the presence of the distinctive characteristics of the different varieties and the decline of some populations. For example, the "Prieto" landrace is mainly used to delimit parcels, in addition to serving to protecting the soil and storing moisture; this smaller landrace optimizes space for the free movement of animals in the field, which would explain the selection of this characteristic.

## Morphological diversity across landraces

The correlation analysis allowed excluding those variables that provided little information in the distribution of the data; this yielded 11 attributes (8 quantitative and 3 categorical variables) to perform the final analyses (Table 1).

According to the PCA performed with the 11 variables, 73% of the total variance is explained by the first 3 components. The first component accounted for 46% of the variance, in which the attributes with the greatest contribution were height, leaf length, and width at the leaf base; the second component explained 19% of the

variance, and the variables with the greatest contribution were tooth shape, number of leaves, and spine length; for the third component, the attribute with the greatest contribution was tooth length (Table 1).

Figure 4 shows that "Manso" plants are grouped to the left of the graph, along with the variables that characterize this group (plant, leaf, tooth, and spine size); "Amarillo" plants are located at the top right side, characterized by the curved tooth shape and leaf margin shape, while most "Prieto" plants were grouped at the bottom right side, identified by leaf shape. In other words, axis 1 separates "Manso" from the other 2 landraces, while axis 2 segregates "Prieto" from "Amarillo".

The hierarchical cluster analysis yielded a cophenetic relationship of 0.82, indicating that the clustering was well-supported. The dendrogram produced 3 groups that were confirmed through the PCA. The first group is composed mainly of "Manso", with a classification certainty of 87%, which displayed a greater morphological variation. The second group clusters all "Amarillo" plants, characterized by curved teeth and a short spine; these agave plants are of intermediate size and, as shown in the dendrogram, are more similar to the "Prieto" agave. The latter is grouped in the cluster on the right, with a 58% certainty, characterized by ascending leaves, a smaller number of teeth, and a smaller plant size (Fig. 5).

Tukey's test showed that 10 of the 11 variables displayed significant differences between landraces; 2 of these attributes were categorical and 8 were continuous (Table 2). "Manso" is the largest variety, with individuals

Table 1 Characteristics evaluated in the analysis of morphological variation and eigenvalues of the first 3 principal components (PC1, PC2 and PC3) (n = 61).

Variable	Code	Unit of measure	PC1	PC2	PC3
Plant total length	PL	cm	-1.3978	0.1447	-0.02095
Number of leaves	NL	number	-0.4704	-0.8792	-0.37154
Leaf length	LL	cm	-1.375	0.3703	0.12703
Leaf width at the base	LWB	cm	-1.3351	0.4926	0.07127
Leafh shape	SLL	Categorical	0.6608	-0.7184	0.57993
Leaf margin shape	LMS	Categorical	1.3258	0.479	-0.02065
Spine length	SL	cm	-0.7542	-0.8808	0.35466
Spine width at the base	SWB	mm	-0.9366	-0.4552	-0.52659
Tooth length	TL	mm	-0.9881	-0.153	0.9468
Number of teeth	NT	Number	-1.1446	0.7745	-0.23716
Tooth shape	ST	Categorical	0.3931	1.1188	0.37717
Percentage of variance explained by the component			46%	19%	8%

Table 2

Tukey's statistical test of 10 morphological attributes that showed significant differences between the 3 landraces of "pulque" agave in Tlalcuapan.

Landrace	PL	NL	LL	LWB	SLL	LMS	SL	SWB	NT	TL
"Manso" (n = 25)	282.64 (23.72)a	214.32 (25.28)a	34 (6.93)a	50.09 (4.15)a	1(0)a	1 (0)a	7.22 (0.85)a	9.70 (1.5)a	60 (8.53)a	21.41 (2.84)a
"Prieto" (n = 25)	201.56 (30.63)b	142.33 (20.25)b	37 (12.72)a	34.01 (4.52)b	2 (0.5)b	2(0.10)b	7.00 (0.81)a	8.59 (1.59)b	35.84 (5.75)b	18.51 (2.69)b
"Amarillo" (n = 11)	192.82 (26.08)b	151.64 (23.80)b	18 (5.66)b	37.94 (6.47)b	1 (0.3)a	3(0)c	5.47 (0.41)b	7.38 (1.33)b	47.91 (47.91)c	16.88 (2.46)b

The standard deviation is shown in parenthesis. Means with the same letter in each column indicate that they are statistically equal (Tukey,  $\alpha = 0.05$ ). PL = Plant total length, NL = number of leaves, LL = leaf length, LWB = leaf width at the base, SLL = leaf shape, LMS = leaf margin, SL = spine length shape, SWB = spine width at the base, NT = number of teeth, TL = tooth length.

measuring 2.82 m in height on average. This variety shows leaves and spines broader at the base and the longest teeth; these attributes are significantly different from those observed in "Prieto" and "Amarillo", with average heights of 2.01 m and 1.92 m, respectively. "Amarillo" shows significant differences in the number of leaves (F = 15.861; p < 0.001) and spine length (F = 20.719; p < 0.001); this landrace has fewer leaves per rosette (18 on average), unlike the other varieties that have 34 or 37 leaves on average. "Amarillo" displays the shortest spines of the varieties studied, measuring an average of 5.47 cm. On the other hand, one of the characteristic attributes of the "Prieto" landrace is the rosette shape

with densely packed lanceolate and ascending leaves, whereas the other varieties have loosely arranged lanceolate leaves.

Only 2 of the 9 variables were significantly different across the 3 varieties: one was leaf margin (F = 77.28; p < 0.001); in "Manso", the leaf margin is straight (attribute coded with number 1), while "Amarillo" shows an undulated leaf margin and "Prieto", semi-undulated leaf margin (coded with numbers 3 and 2, respectively). The number of teeth also differed significantly (F = 62.265; p < 0.001) across landraces, with "Manso" showing the largest number of teeth (60 on average), followed by "Amarillo" (48), and then "Prieto", with the lowest (36).

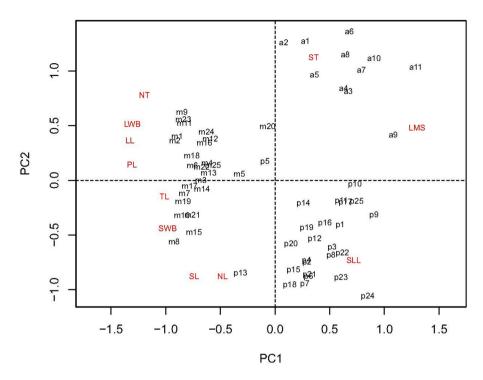


Figure 4. Ordination plot of the Principal Component Analysis (PCA) of the 61 agave plants belonging to the 3 traditional landraces (m: "Manso", a: "Amarillo", p: "Prieto"), with regard to the 11 morphometric variables showing the greatest variation: SWB = spine width at the base, LWB = leaf width at the base, SL= spine length, LL = leaf length, NL = number of leaves; NT = number of teeth; PL = plant total length; TL = tooth length; SLL = leaf shape; ST = tooth shape; LMS = leaf margin shape.

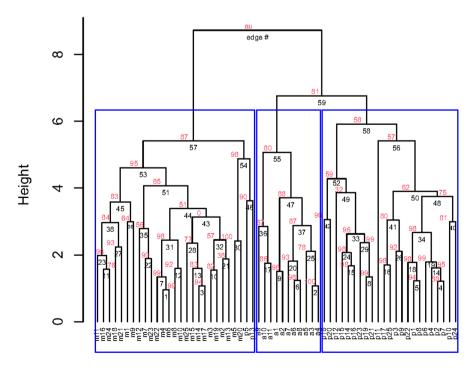


Figure 5 Tree of morphological distances of the plants analyzed (r = 0.82). The clustering method used was the farthest neighbor approach. Values under each node correspond to the group; figures in red indicate the value that supports the group. Abbreviations: m = ``Manso'', a = ``Amarillo'', p = ``Prieto''.

### **Discussion**

There are scarce records of *Agave* in Tlaxcala, Mexico. Gentry (1982), reported no records of agave in Tlaxcala. Some studies have reported agave species and landraces used to produce "pulque" in Tlaxcala. Mora-López et al. (2011) reported, for 4 localities in 4 municipalities of Tlaxcala, Agave mapisaga "Sabililla", A. mapisaga var. lisa and 4 landraces of A. salmiana var. salmiana. Álvarez-Duarte et al. (2018) recorded 9 landraces of A. salmiana subsp. salmiana for 9 localities of the municipalities of Nanacamilpa and Calpulalpan. Four landraces of Agave salmiana subsp. salmiana were reported in a locality of Nanacamilpa, Tlaxcala, (Álvarez-Ríos, Figueredo-Urbina et al., 2020; Ramírez-Manzano et al., 2020). Besides, Trejo et al. (2020) recorded A. mapisaga var. mapisaga "Palmilla" and 8 landraces of A. salmiana subsp. salmiana for 10 localities in 9 municipalities of Tlaxcala. Finally, Trejo et al. (2022) reported, for the central zone of Nanacamilpa, Tlaxcala, A. americana "Cenizo", A. mapisaga var. mapisaga "Palmilla", A. salmiana subsp. tehuacanensis "Cimarrón", 7 landraces of A. salmiana subsp. salmiana, and 2 unidentified landraces because they are no longer present in the area. It is worth noting that A. salmiana var. salmiana and A. salmiana subsp. salmiana can be considered synonyms, the former based on Gentry (1982) and the latter, on García-Mendoza (2011).

Although SPT is a small locality, it harbors a high diversity of landraces used to produce "pulque". *Agave americana* "Palma" and *A. salmiana* subsp. *salmiana* "Amarillo" and "Mahuahuac" have been reported only for SPT. All other landraces have been recorded at least in one other locality in Tlaxcala. This finding may reflect that indigenous populations growing agave under low cultivation intensity conserve a high landrace diversity, as suggested by some authors (Álvarez-Ríos, Pacheco-Torres et al., 2020; Reyes-Agüero et al., 2019).

The names of "pulque" agave landraces are assigned by persons who are well-acquainted with these plants. Traditional identification is based on characteristics such as plant color, plant size, spine length, and leaf length. However, the same variety can be given various names in different zones or localities, hence the importance of analyzing the lexical marking since it can provide information on the cultural significance of that plant if its name does not change within a zone or in adjacent areas (Berlin, 1992). A wide distribution of the "Manso" and "Colorado" landraces of *Agave salmiana* subsp. *salmiana* has been observed in the state of Tlaxcala (Álvarez-Duarte et al., 2018; Mora-López et al., 2011; Trejo et al., 2020). "Manso" has been reported as the landrace most extensively used in the Central Mexican Plateau (Álvarez-

Ríos, Figueredo-Urbina et al., 2020; Alfaro-Rojas, et al., 2007; Mora-López et al., 2011). Some landraces thrive in the central region of the state, such as "Prieto" and "Matecón" landraces of *Agave salmiana* subsp. *salmiana* (Trejo et al., 2020). *A. mapisaga* Trel. var. *mapisaga* "Penca larga" is grown in the southwest region of Tlaxcala (Trejo et al., 2020).

The names used for the SPT landraces and those assigned in nearby regions are identified as the same species and are given the same common name; thus, the names indicate the cultural importance of these plants in the locality, in both the state of Tlaxcala and, actually in all central Mexico. It appears that landraces are developed from the selection of local resources, a phenomenon that has also been observed in other regions of Mexico (Álvarez-Duarte et al., 2018; Mora-López et al., 2011; Reyes-Agüero et al., 2019). This fact highlights the importance of the knowledge of the traditional characteristics used in the identification of *Agave* landraces; this is highly relevant for biocultural conservation and underlines the risk of local extinction facing agave plants and the loss of knowledge on them.

As in other ethnobotanic studies about Agave (Álvarez-Duarte et al., 2018; Ramírez, 2007; Vázguez-García, 2018), additional information was gathered from local experts (named "tlachiqueros"). The emigration of young generations has influenced the loss of traditional knowledge. The new generations are no longer interested in learning "tlachiquero" skills and prefer to emigrate to urban areas in search of better-paid job opportunities (Álvarez-Duarte et al., 2018; Vázquez-García, 2018). In SPT, all respondents were consistent in naming and identifying agave landraces. However, only 1 respondent provided the name of landraces in both Nahuatl and Spanish. The loss of knowledge of the Nahuatl names may be due to the decrease in the number of speakers of this language and the little interest in learning "tlachiquero" skills. To date, there are scarce studies addressing the names and identification of agave plants from the traditional knowledge perspective (Colunga-GarcíaMarín and May-Pat, 1993; Ramírez-Manzano et al., 2020; Reyes-Agüero et al., 2019).

In the state of Tlaxcala, the "Manso" landrace of Agave salmiana subsp. salmiana ranks first in the frequency of mentions, indicating the importance of this landrace for "pulque" production in the locality. The importance of this landrace has also been reported for a large portion of the "pulque"-producing zone in Mexico (Alfaro-Rojas et al., 2007; Álvarez-Duarte et al., 2018; Álvarez-Ríos, Figueredo-Urbina et al., 2020; Mora-López et al., 2011; Trejo et al., 2020). According to these reports, "Manso" is preferred because of its size, the flexibility of its leaves that facilitate handling, and, most of all,

the high quality of its sweeter sap of 9-15 °Brix (Mora-López et al., 2011; Ramírez-Manzano et al., 2020; Trejo et al., 2022).

In SPT, "Amarillo" ranked second in frequency of mentions; this landrace is very scarce in the area and is rarely used, but draws attention for the relevance it still has among the local inhabitants. The respondents mentioned that fifty years ago, "Amarillo" was extensively used to extract "ixtle" (fibers of agave leaves) to make "cuartas" (ropes) used in religious festivities and to produce "pulgue". However, due to the use of synthetic ropes, the current "ixtle" extraction is nil, which led to the decline of "Amarillo" cultivation. The landrace ranking third in frequency of mentions was "Prieto". Trejo et al. (2020) point out that this landrace is used to produce "pulque" mainly in the central region of Tlaxcala. It is likely that "Prieto" is synonymous with the "Negro" landrace also used for "pulque" production in the Estado de Mexico (Alfaro-Rojas et al., 2007).

The analysis of morphological variables in this work recognized morphological clusters that correspond to the 3 traditional landraces mentioned most frequently and are the most abundant landraces in the locality studied. *Agave salmiana* subsp. *salmiana*, fostered by the selection and management practices of peasants. Trejo et al. (2020) observed morphological diversity within the subspecies *salmiana*; however, no such differentiation was evident at the genetic level using nuclear and chloroplast markers. These findings suggest an incipient domestication scenario.

Three of the 5 characters used by peasants to differentiate landraces (plant length, leaf length, and leaf width) have been significant in most studies addressing the morphological diversity of agave plants (Table 3). In contrast, characteristics such as plant color have been poorly analyzed and spine stiffness has not been evaluated in these studies. The elements used by peasants to identify landraces and the statistical analysis of the variability of morphological characters are useful for identifying the differences between landraces. Therefore, the 2 approaches complement each other to recognize diversity.

Several studies have mentioned that plant size, number of teeth, tooth size, and distance between teeth are associated with *Agave* domestication and suggest that artificial selection has caused this morphological trend (Alfaro-Rojas et al., 2007; Figueredo et al., 2021; Mora-

López et al., 2011). On the one hand, larger plants produce a greater volume of sap and are, therefore, associated with a higher sap production; on the other, smaller or more separated teeth facilitate manipulation (Figueredo et al., 2021; Mora-López et al., 2011). This information partially coincides with the results of the present study since the preferred landrace by *tlaquicheros* for "pulque" production is "Manso", a landrace characterized by intermediate plant size. "Manso" is neither a large-sized landrace nor the one with the highest sap production; it displays mid-sized lateral spines and flexible leaves (Alfaro-Rojas et al., 2007; Mora-López et al., 2011; Ramírez-Manzano et al., 2020; Trejo et al., 2020).

The holistic or essentialist understanding of peasants to recognize their own landraces has not been addressed in detail. Understanding it may contribute to including additional characters to improve the description of the morphological diversity of landraces and the reasons for their selection. More detailed interviews with peasants would broaden our understanding of how they identify agave plants and better describe how agave cultivation is managed. Strategies should be developed aiming to gather more in-depth and detailed information from peasants, as the descriptions they have provided so far are just basic and general; the current approaches seem relatively ineffective as peasants do not provide detailed data and may even get impatient.

San Pedro Tlalcuapan is a small town that harbors a large number of landraces; however, most of these include just a few individuals, highlighting the urgent need to implement conservation strategies to prevent their disappearance. Such strategies shall promote the dissemination of these studies in the local communities and the use of all varieties, raise interest in the "pulque" production chain by the young generations, and promote the transfer of traditional knowledge and the learning of Nahuatl.

The qualitative morphological traits used by peasants to distinguish landraces are significant in most studies addressing the morphological diversity of agave plants. The two approaches complement each other and allow identifying biodiversity patterns. Furthermore, some traits used by peasants, such as the flexibility of the terminal spine, can be integrated into the traditional morphometric studies.

Table 3
Significant characters in the first 3 principal components or canonical functions in studies of morphological diversity in cultivated or managed agave.

Use*	SWB	TW	LWM	LWB	SL	LL	DBT	$\Gamma\Gamma$ S	PD	Ŋ	NT	PL	$C\Gamma$	DLBS	TC	IL	SLL	SBW	CW	ST	RL	SF	Reference
Fiber	X	X	X	X	X	X	X				X												Colunga-GarcíaMarín et al., 1996
Mezcal				X	X	X	X				X	X											Vargas-Ponce et al., 2007
Mezcal		X	X			X	X	X	X		X	X		X									Rodríguez-Garay et al., 2009
Mezcal					X	X		X															Zizumbo-Villarreal et al., 2012
Mezcal		X	X			X	X		X		X	X	X		X	X							Vázquez-Pérez, 2015
Mezcal			X		X		X		X	X	X	X						X					Avendaño-Arrazate et al., 2015
Mezcal					X	X		X		X	X												Rivera-Lugo et al., 2018
Mezcal			X						X		X	X											Cabrera-Toledo et al., 2020
Mezcal					X	X	X		X	X		X				X							Castañeda-Nava et al., 2019
Pulque					X						X												Alfaro-Rojas et al., 2007
Pulque		X		X		X			X	X	X	X											Mora-López et al., 2011
Pulque		X	X		X	X			X		X	X				X		X					Figueredo-Urbina et al., 2014
Pulque						X			X			X											Álvarez-Ríos, Pacheco- Torres et al., 2020
Pulque				X	X	X								X		X	X		X	X	X		Trejo et al., 2020
Pulque	X	X			X	X	X		X		X	X				X							Figueredo-Urbina, Álvarez-Ríos, García- Montes et al., 2021
Various Beverages			X			X	X		X			X	X			X							Figueredo-Urbina et al., 2017
Various Beverages			X			X			X	X		X											Torres-Morán et al., 2013
Various Beverages					X	X	X	X															Pórras-Ramírez et al., 2016
			X		X	X				X	X	X				X				X			This work
			X			X						X	X									X	Traditional knowledge
	2	6	10	4	12	17	9	4	11	6	12	14	2	2	1	7	1	2	1	2	1	1	Total

<sup>\*</sup>Main use. SWB = Spine width at the base; TW = tooth width; LWM = leaf width at mid-length; LWB = leaf width at the base; SL = spine length; LL = leaf length; DBT = distance between teeth; LLS = length of lateral spines; PD = plant diameter; NL = number of leaves; NT = number of teeth; PL = plant total length; CL = leaf color; DLBS = distance between last lateral and apical spines; TC = tooth color; TL = tooth length; SLL = leaf shape; SBW = spine width at the base; SS = spine shape; CW = cuticle width; ST = tooth shape; RL = percentage of rigid leaves; SF = spine flexibility.

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