

# Microhistological References of Plants Available for Ungulates in Sonora, Mexico

Peralta-Pardo, Raúl<sup>1</sup><sup>(10)</sup>, Palacio-Núñez, Jorge<sup>1</sup><sup>(10)</sup>, Tarango-Arámbula, Luis A.<sup>1</sup><sup>(10)</sup>, Olmos-Oropeza, Genaro<sup>1\*</sup><sup>(10)</sup>, Martínez-Montoya, Juan F.<sup>1</sup><sup>(10)</sup>; Saucedo-Uuh, Krisly<sup>1</sup><sup>(10)</sup>

<sup>1</sup> Colegio de Postgraduados, Campus San Luis Potosí, Posgrado de Innovación en Manejo de Recursos Naturales. Iturbide 73, Salinas de Hidalgo, San Luis Potosí, México, C.P. 78620.

\* Correspondence: olmosg@colpos.mx

#### ABSTRACT

The microhistological technique is the most popular methodology used to determine the wild and domestic ungulates diet; its success depends on the development of a reliable reference catalog.

**Objective**: To describe and analyze the epidermal structures of the plants available for wild and domestic ungulates in Sonora, Mexico, using the microhistological technique.

**Methodology**: A comprehensive collection of the plants available for wild and domestic ungulates was carried out at UMA Rancho Noche Buena, in order to subsequently identify their genus and species. Plant structures were analyzed with the modified microhistological technique, scraping the beam and underside cuticle of leaf plants. In addition, at least one photograph was taken to characterize them.

**Results and Discussion**: The epidermal structures of 95.95% of the analyzed plants were observed with the microhistological technique and its modification. The distinctive structures of 74 plant species were identified, in order to describe the main characteristics of each species. A catalog of microhistological references was developed from the data collected; it included information about the morphology and arrangement of structures such as: epidermal cells, stomata and trichrome. A difference was found between the beam and underside of the leaves in 23% of the species.

**Conclusion**: A reliable microhistological reference catalog should consider possible differences between the beam and underside of the leaves of plants.

Keywords: Scraping, beam, underside, leaves.

### **INTRODUCTION**

Hunting in northern Mexico is one of the most successful wildlife management activities in the country (Valdez *et al.*, 2006). A large number of ranches in Sonora (categorized as Wildlife Conservation Management Units [UMAs]) have benefited economically from the sale of hunting licenses. Various herbivores —especially mammals, such as bighorn sheep, mule deer, white-tailed deer, and collared peccary— are hunted each year, providing an important economic benefit for the state (Guajardo-Quiroga and Martínez-Muñoz, 2004). In many cases, the sustainable use of these animals involves knowledge of their ecology and the implementation of management practices, favoring their conservation,

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as well as knowledge of their food resources. Different techniques were used to determine the herbivores diet, including behavioral observation, NIRS, stable isotopes, and DNA. However, one of the most popular techniques was microhistology, proposed by Baumgartner *et al.* (1939) and modified by Sparks and Malechek (1968) (Garnick *et al.*, 2018). It is based on the identification and quantification of digestion-resistant epidermal tissues found in the stomach, rumen, or excreta contents (Holechek *et al.*, 1982). On the one hand, its main advantages are its low cost and non-invasiveness; on the other hand, its main disadvantage is the time investment required to train the personnel that will develop the reference catalogue (Holechek, 1982; Garnick *et al.*, 2018).

The catalogue or references is a set of images (photographs or drawings) and descriptive notes of leaf epidermal structures; they are representative of each plant found in the habitat of the herbivore under study (González and Améndola, 2010). The possible differences between the beam and underside of the same leaf should be taken into consideration, since they are sometimes recorded as if they belonged to different species. A comprehensive and representative catalogue of the plants available in the habitat is one of the essential components of the technique and it helps to determine diets with a high level of accuracy (Alipayo *et al.*, 1992; Cuartas and García-González, 1996; Garnick *et al.*, 2018). Epidermal scraping (González and Améndola, 2010) is one of several procedures used to obtain the epidermal tissues to be analyzed (Catán *et al.*, 2003; Castellaro *et al.*, 2007; González and Améndola, 2010; Arellano *et al.*, 2019). It keeps intact the structures used to characterize and describe the epidermal conformation of the leaves of each plant species. Moreover, two additional advantages are that it requires few inputs and that its application is relatively simple.

The aim of this study was to describe and analyze the plants available to wild and domestic ungulates in Sonora, Mexico, using the microhistological technique.

# METHODOLOGY

#### Description of the study area

The Management Unit for Wildlife Conservation (UMA) Rancho Noche Buena is located in the southern Sonoran Desert, 120 km northwest of Hermosillo and 14.2 km away from the Sea of Cortez (29° 12' 19.45" N, 112° 0' 22.80" W). Its 16,800 ha are the natural habitat of bighorn sheep, mule deer, white-tailed deer, and collared peccary, although cattle has been introduced to the area. Climate is classified as BWh(x'), the average temperature is 22.3 °C (coldest, -1.5 °C; hottest, 47 °C), and the average rainfall varies from 172.9 to 193.9 mm (López *et al.*, 1999; García, 2004; SMN, 2019). The predominant vegetation is arbosufrutescent scrub, sarcocaulescent scrub, and crassicaule scrub. The most relevant species are *Carnegie gigantea*, *Pachycereus pringlei*, *Cylindropuntia* spp., *Olneya tesota*, *Parkinsonia microphylla*, and *Bursera microphylla* (López *et al.*, 1999; León *et al.*, 2018).

# Sample collection and preparation of the reference catalogue

From August to October 2018, an extensive plant collection was carried out at UMA Rancho Noche Buena. Taxonomic identification of the collected material was then carried out with the support of Mr. Diego Valdez Zamudio (ScD), a specialist in the vegetation of Sonora and in the literature regarding the flora of northwestern Mexico (Shreve, 1951; León *et al.*, 2018; SEINet, 2019).

The reference catalogue was developed following the methodology described by González and Améndola (2010). Leaves were hydrated for one day and the epidermis —both on the beam and the underside— was scraped with a razor blade and then rinsed with 12% sodium hypochlorite. The epidermal tissues obtained were then mounted on a slide with glycerin jelly and placed on a  $24 \times 40$  mm slide. The samples were labelled, stored, and left to settle for two weeks. Three preparations were made for each species. Once the mounting medium had solidified, representative areas of each of the epidermal tissues were located using a Leica<sup>®</sup> microscope with a 10x objective; based on those tissues, the epidermal structures of each species was described. Photographs were taken of the representative areas using a Nikon D5600 camera (Annex 1).

## Description of epidermal structures and data analysis

Characterization took into consideration the easily identifiable structures (cells, stomata, trichomes) and special structures, as well as the differences between the beam and underside of the same species (Peña and Habib, 1980; Johnson *et al.*, 1983; González and Améndola, 2010). Epidermal cells were catalogued according to the combination of factors specific to both the cells that make up the regular epidermis and other cells or structures, such as stomata, trichomes and glands. In the case of epidermal cells, their arrangement (irregular or regular), shape (puzzle piece, polygonal, irregular, or rectangular), and size (small, medium, or large) were taken into account. Cell wall specificities such as thickness (thick, medium, or thin), levels of undulation (faint, short, or deep), and texture (smooth, striated, or granular) were also considered.

Stomata were classified according to their shape (oval, elongated oval, round, or rhombic), size (small, medium, and large), orientation (random and unidirectional), and arrangement (actinocytic, anisocytic, anomocytic, cyclocytic, diacytic, exposed, hexacytic, implanted, unexposed, paracytic, pentacytic and tetracytic). Trichomes were classified according to their type (simple unicellular, unicellular with two branches, unicellular with bulbous base, glandular unicellular, simple bicellular, simple multicellular, multicellular stellate, multicellular vesicular and glandular). The following special structures were recorded: asperidia, cork cells, silica cells, crystals, druses, glands, papillae, raphidia and tannins.

# **RESULTS AND DISCUSSION**

The catalog of microhistological references of vegetation at UMA Rancho Noche Buena was developed using the epidermal tissues of 74 species from 25 taxonomic families (Table 1 and Annex 1). The highest number of species were recorded for Fabaceae, Euphorbiaceae, and Cactaceae, with 11, 8, and 7 species, respectively. The microhistological structures of 71 species (95.95%) could be observed without any difficulty; on the contrary, certains structure were difficult to observe in the following three species (4.05%): *Abutilon incanum* 

and *Solanum hindsianum* (due to the high density of trichomes) and *Bebbia juncea* (because the size and structure of leaves prevented the scraping).

As a result of the characterization, 86.5% of the species were determined to have an irregular cell arrangement, 12.2% a regular arrangement and 1.4% could not be observed. The shape proportion of the cell walls were classified as follows: 42.5% irregular (17.2% with short undulations, 14.9% with faint undulations, and 10.3% with deep undulations), 41.4% polygonal (32.2% with straight cell walls and 9.2% with faint undulations), 11.5% rectangular (6.9% with short undulations, 2.3% with faint undulations, 1.1% with straight cell walls, and 1.1% with deep undulations), 2.3% oval, and 2.3% not observed. Cell texture was 54.1% smooth, 29.7% granular, 12.2% striated, and 4.1% not observed. Regarding this same collection, epidermal cell size was considered as medium (43.4%), small (34.2%), large (19.7%), and not observed (2.6%). Finally, cell walls were determined to be thin (56.8%), thick (39.2%) and not observed (4.1%).

Regarding their shape, stomas were oval (61.0%), elongated oval (16.9%), round (15.6%), rhombic (2.6%), and not observed (3.9%). In terms of their size, stomas were medium (37.7%), small (32.5%), large (26.0%) and not observed (3.9%). Regarding their orientation, 71.6% of the stomas were random, 23% were unidirectional and 5.4% were not observed. The stomatal arrangement was 29.6% tetracytic, 20.4% anisocytic, 11.2% paracytic, 9.2% pentacytic, 7.1% exposed, 6.1% hexacytic, 5.1% cyclocytic, 4.1% unexposed, 2.0% actinocytic, 2.0% diacytic, 2.0% implanted and 1% anomocytic.

Trichomes were recorded in 68.9% of the species. In proportion to their type, trichomes were unicellular simple (45.6%), simple bicellular (12.3%), simple multicellular (10.5%), multicellular stellate (8.8%), unicellular with two branches (7.0%), unicellular glandular (5.3%), multicellular glandular (5.3%), vesicular (3.5%), and unicellular with bulbous base (1.8%).

Special structures were observed in 71.6% of the species, divided as follows: 25.7% druses, 21.4% tannins, 12.9% raphidia, 11.4% papillae, 10.0% cork cells, 8.6% silica cells, 4.3% glands, 2.9% crystals and 2.9% asperidia.

In 23% of the species, differences between the beam and the underside were observed. These differences accounted for 55.6% of the cell shape, 11.1% greater abundance of trichomes on the underside, 11.1% more abundant trichomes on the upper side, 11.1% stomata only on the underside, 5.6% trichomes only on the underside and 5.6% in cell size.

Characterization allowed to identify patterns for different plant families and species. The most representative characteristics of family Poaceae were regular cell arrangement, rectangular cell shape, unidirectional stomata, and the presence of asperidia, silica cells, and cork. Family Cactaceae was characterized by polygonal cell shape, unidirectional stomata, and absence of trichomes. For their part, most of the species of family Fabaceae had simple unicellular trichomes and tannins. Finally, Zygophyllaceae had unicellular simple trichomes and papillae, while Malvaceae had a high density of stellate trichomes.

The modification proposed by González and Améndola (2010) for the development of the reference catalog proved to be a reliable tool for the identification of epidermal structures (cells, stomata, trichomes, special structures, and beam/underside differences) and for the characterization of plant species at UMA Rancho Noche Buena.

				Cells				Stor	nas		Trichomes		əp
Texture Texture	Агталgеment Shape Теxture	Shape	Texture		əziZ	Cell wall	Shape	əzi2	Orientation	Arrangement	əqvT	Special Structures	Beam/undersi difference
Justicia californica I Po L	I Po L	Po L	Г		Р	Gr	0	Μ	Α	Di y Ani	Bs	Dr	
Ruellia californica I Ip L	I Ip L	Ip L	Г		М	D	Oa	М	Α	Di	Us		
Trianthema portulacastrum I It Es	I It Es	It Es	$\mathbf{E}_{\mathbf{S}}$		G	D	OyRed	IJ	Υ	Tet		$\mathrm{Dr}$	
Amaranthus palmeri I It <sup>1</sup> Ip <sup>2</sup> L	I It <sup>1</sup> Ip <sup>2</sup> L	$It^1 Ip^2 I$	Г		Р	D	0	Ь	Α	Ani y Tet			Fc
Tidestromia lanuginosa I I Ip L	I Ip L	Ip L	L		G	D	0	Μ	Υ	Ano	Us	Ra	
Ambrosia dumosa I I Ic <sup>1</sup> It <sup>2</sup> L	I Ic <sup>1</sup> It <sup>2</sup> L	$Ic^{1} It^{2}$ L	Γ		Р	D	0	Р	Α	Ani	$\mathbf{P}_{\mathbf{S}}$	GI	
Bebbia juncea I Po N	I Po N	Po N	Ν		М	Ν	Ν	N	Ν	Ν	$\mathbf{P}_{\mathbf{S}}$		
Encelia farinosa I I Ic L	I Ic L	Ic L	Γ		Р	D	0	Μ	Α	Tet	Us y V	GI	Те
Pectis rushyi I Ip L	I Ip L	Ip L	Γ		G	D	Oa	G	n	Tet		Ra	
Trixis californica I I Gra	I It Gra	It Gra	$\mathbf{Gra}$		Μ	Gr	Oa	უ	Α	He	$\mathbf{P}_{\mathbf{S}}$		
Cordia parvifolia I Po <sup>1</sup> Ic <sup>2</sup> L	$I Po^1 Ic^2 L$	$Po^{1} Ic^{2}$ L	Γ		Ρ	D	0	$M \mathrel{y} G$	V	Tet	Ωd		$\mathbf{Fc}$
Bursera laxiflora I Po Es	I Po Es	Po Es	Es		Μ	D	0	Μ	Α	He		Dr	Ee
Bursera microphylla I Po L	I Po L	Po L	Γ		Ρ	D	0	Μ	Υ	Pen			
Carnegiea gigantea I Po Gra	I Po Gra	Po Gra	Gra		М	$\mathrm{Gr}$	Red	G	U	Ci			
Cylindropuntia fulgida I Po L	I Po L	Po L	Γ		Ρ	$\mathrm{Gr}$	0	М	N	Ci		$\mathrm{Dr}$	
Cylindropuntia thurberi I Po L	I Po L	Po L	Γ		Р	D	Red	Μ	Ŋ	Ci		$\mathrm{Dr}$	
Ferocactus wislizenii I Po Es	I Po Es	Po Es	$\mathbf{E}_{\mathbf{S}}$		G	Gr	0	G	N	$\operatorname{Par}$			
<i>Lophocereus schottii</i> I Ic Es	I Ic Es	Ic Es	$\mathbf{E}_{\mathbf{S}}$		G	Gr	0	IJ	Α	Par			
Pachycereus pringlei I Po Es	I Po Es	Po Es	$\mathbf{E}_{\mathbf{S}}$		Ρ	Gr	0	G	Α	Ci		$\mathrm{Dr}$	
Stenocereus thurberi I Po Gra	I Po Gra	Po Gra	$\mathbf{Gra}$		М	$\mathrm{Gr}$	Oa	М	U	$\operatorname{Par}$			
Croton sonorae I I L	I Ic L	Ic L	Γ		Ρ	D	0	Р	Α	Tet	Pe y Us	$\mathrm{Dr}$	
Ditaxis lanceolata I Po L	I Po L	Po L	Γ		М	Gr	Oa	Μ	Α	Par y Tet	Ud	$\mathrm{Dr}$	$\operatorname{Th}$
Euphorbia eriantha I Po Gra	I Po Gra	Po Gra	$\mathbf{Gra}$		М	D	0	Р	Α	Ani y Tet	$\mathbf{P}_{\mathbf{S}}$	Ra	$\mathrm{T}_{\mathrm{s}}$
Euphorbia hyssopifolia I I Ic <sup>1</sup> Ip <sup>2</sup> Gra	$I$ $Ic^{1} Ip^{2}$ $Gra$	$Ic^1 Ip^2$ Gra	Gra		М	Gr	0	Р	Υ	Ani y Tet	Bs	$\mathrm{Dr},\mathrm{Pa}~\mathrm{y}~\mathrm{Ra}$	
Euphorbia prostrata I Pt <sup>1</sup> It <sup>2</sup> Gra	I $Pt^{1} It^{2}$ Gra	$Pt^{1} It^{2} Gra$	Gra		G	Gr	0	Р	Α	Ani		$\mathrm{Dr}$	Fc
Jatropha cinerea I Po Es	I Po Es	Po Es	$\mathbf{E}_{\mathbf{S}}$		М	Gr	0	Μ	Υ	Tet	Us	$\mathrm{Dr}$	
Jatropha cuneata I Po Gra	I Po Gra	Po Gra	$\mathbf{Gra}$		М	D	0	Μ	Α	Tet			
Sebastiania bilocularis I Po L	I Po L	Po L	Γ		Ρ	Gr	Red	Μ	Α	Par y Tet		$\mathrm{Dr}$	
Caesalpinia palmeri I Ic Es	I Ic Es	Ic Es	$\mathbf{E}_{\mathbf{S}}$		IJ	Gr	0	Ь	Υ	Tet	Us		

Continues	
Ë.	
Table	

əp	Beam/undersie bifference			Ee											Fc							Fc	Fc		Th	$\mathbf{Fc}$			Fc y Tc	
	Special Structures		${ m Dr}$ y Ta	${ m Dr}~{ m y}~{ m Ta}$	Та		Ta	Та		Ta	Та	$\mathbf{Pa}$	Ta		Ra	$\mathrm{Dr}$	Та		GI		Ra	Ra y Cr	Ra	Та	$\mathrm{Dr}$		Cc	$C_{S}$	$\mathrm{Cs},\mathrm{Cc}\;\mathrm{y}\mathrm{As}$	$C_{S}$ y Pa
Trichomes	əqvī	Us	Us	Us			Us	Us	Us	$U_{\rm S}$	Us		Us	Ud	Ub	Ud		$\mathbf{Pe}$	$\mathbf{Pe}$	${ m Pe~y~Ug}$	$\mathrm{Pg}$	Ug	Ug	Us	Us	$P_{g}$	Bs	Bs y Us	$U_{S}$	$_{\mathrm{Bs}}$
	Arrangement	Tet	Ac	Ani y Tet	Par	Par y Ani	Ani y Tet	Tet	Par	Par	Par y Ani	Pen y He	Tet y Pen	Ani	Pen y He	Tet	Tet	Ν	z	Ani y Tet	Ani y Tet	Ani y Tet	Tet	Pen y He	Ani y Tet	Ani	Im	Im	Im	Im
nas	Orientation	Α	Α	Υ	Α	Α	Α	Α	Α	Y	Α	Α	n	N	Α	Α	Α	Ν	z	Υ	Α	Α	Α	Α	Υ	Y	n	n	U	D
Stor	əziZ	Ь	Р	Ρ	Ь	Ь	Ь	Ь	Μ	Ρ	U	IJ	IJ	Р	G	IJ	IJ	Ν	Μ	М	Μ	IJ	IJ	Ь	М	Р	Μ	Μ	Ρ	Μ
	эdвdZ	0	0	Oa	Oa	0	0	0	0	0	0	0	0	$\operatorname{Red}$	Re y O	Oa	Oa	Ν	Oa	0	0	0	Red	Red	0	0	Oa	Red	0	0
	Cell wall	D	D	Gr	D	D	G	D	D	Gr	D	Gr	D	D	D	D	D	Ν	D	D	Gr	Gr	Gr	D	D	D	Gr	Gr	D	D
	əziZ	Ь	Μ	Μ	Μ	М	Ь	Ь	IJ	Μ	Μ	Р	Μ	Μ	Ь	Μ	IJ	Ν	Μ	Р	Μ	IJ	Μ	Р	М	Р	Р	Μ	$\mathbf{M}^1\mathbf{P}^2$	Р
Cells	Texture	Gra	Gra	Gra	Gra	Г	Gra	$\mathbf{Gra}$	Γ	$\mathbf{Gra}$	Gra	Γ	L	Γ	L	$\mathbf{Es}$	Gra	Ν	L	L	Gra	Gra	Gra	Γ	L	Γ	L	Γ	$\mathbf{Gra}$	Γ
	ədend	Ic	$\mathbf{P}_{\mathbf{O}}$	$\mathrm{It}^{1}\mathrm{Ic}^{2}$	Pt	It	Ро	Po	Ip	$\mathbf{P}_{\mathbf{O}}$	Ро	$\mathbf{P}_{\mathbf{O}}$	Pt	Ic	$Ic^1 Po^2$	It	Ic	Ν	Ро	$\mathbf{P}_{\mathbf{O}}$	It	$Pt^1 It^2$	${\rm Pt}^1{\rm Ic}^2$	Ic	$_{\rm Ip}$	$\mathrm{It}^{1}  \mathrm{Ip}^{2}$	m Rc	Rc	$Rc^{1} Rp^{2}$	$\mathbf{Rc}$
	Arrangement	I	I	I	I	I	Г	I	I	I	Г	I	I	I	I	I	I	Ν	I	Ι	I	I	I	I	I	I	R	К	R	Ч
	Species	Dalea mollissima	Ebenopsis confinis	Eysenhardtia orthocarþa	Mariosousa willardiana	Mimosa laxiflora	Olneya tesota	Parkinsonia microphylla	Phaseolus grayanus	Prosopis juliflora	Senna covesii	Fouquieria splendens	Krameria erecta	Linum lewisii	Mentzelia pumila	Callaeum macropterum	Galphimia angustifolia	Abutilon incanum	Hibiscus denudatus	Melochia tomentosa	Allionia incarnata	Boerhavia coccinea	Boerhavia coulteri	Passiflora arida	Passiflora quercetorum	Pseudorontium cyathiferum	Aristida ternipes	Bouteloua aristidoides	Bouteloua barbata	Cathestecum erectum
	Family					-	rabaceae					Fouquieriaceae	Krameriaceae	Linaceae	Loasaceae	Malaite	Maipigniaceae		Malvaceae			Nyctaginaceae		D	rassiliotaceae	Plantaginaceae		D	I UAUGAG	

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	Structures Structures	ç	Cc, Cs y A	Cs y Cc	Cr, Ta y C	Cs y Cc	Ra	Та	Dr y Ta	Dr y Ta	Pa			$\mathbf{Pa}$	$\mathbf{Pa}$		Pa
Trichomes	əqYpe	Bs	Bs		Us	Us		Us y Gl	$\mathbf{P}_{\mathbf{S}}$	$\mathbf{P}_{\mathbf{S}}$		$\mathbf{Pe}$	$U_{s y} V$		Us		Us
	Arrangement	Ex	Im	Im	Im	Ex	Ani y Tet	Tet	Ci	Ani y Tet	Ani y Pen	Ν	Par y Ani	Pen y He	Tet y Pen	Zygophyllaccae     Larrea tridentata     I     Po     L     P     Gr     O     P     A     Ac     Us     Pa       Tribulus terrestris     I     Pt     Es     G     Gr     OyRed     P     A     Pen     Us     Pa	
mas	Orientation	D	D	D	D	D	A	A	Α	Α	Α	Z	A	Α	Α		Z
Sto	əzi8	U	Μ	М	М	IJ	М	Ь	Р	$\rm P \ y \ M$	$\rm P \ y \ M$	Z	Μ	М	Μ	ç	$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$
	Shape	$R_{O}$	0	$R_0$	Red	0	0	0	$\operatorname{Red}$	0	0	Ν	0	Oa	Oa	Logophyliaceae     Larrea tridentata     I     Po     L     P     Gr     O     P     A     Ac     Us     Pa       Tribulus terrestris     I     Pt     Es     G     Gr     OyRed     P     A     Pen     Us     Pa	
	Cell wall	Gr	D	D	D	D	D	D	Gr	D	Gr	Z	Gr	D	D	C	5
	əziZ	Μ	IJ	Μ	Μ	ს	IJ	Μ	Р	Μ	$G^1  y  P^2$	N	М	М	Р	F	ч
Cells	Texture	г	Г	Г	Г	Г	Gra	Г	$\mathbf{Gra}$	Г	Г	Z	Г	Γ	Γ	F	ntata         I         Po         L         P         Gr         O         P         A           estris         I         Pt         Es         G         Gr         OyRed         P         A
	ədeyg	Rt	Rc	$Rc^{1} Re^{2}$	Rt	0	Pt	Ip	$\mathbf{P}_{\mathbf{O}}$	${\rm Pt}^1  {\rm Ic}^2$	0	Z	$\mathbf{P}_{\mathbf{O}}$	Ic	It	Ceale     Larrea tridentata     I     Po     L     P     Gr     O     P     A     Ac       Tribulus terrestris     I     Pt     Es     G     Gr     OyRed     P     A     Pen	
	Arrangement	Я	R	К	К	Я	I	I	Ι	I	I	I	I	I	I	ŀ	-
	Species	Cenchrus ciliaris	Echinochloa colona	Eragrostis pectinacea	Eriochloa acuminata	Panicum hirticaule	Colubrina viridis	Cardiospermum corindum	Simmondsia chinensis	Datura discolor	Lycium berlandieri	Solanum hindsianum	Lippia palmeri	Fagonia laevis	Kallstroemia parviflora		Larrea tradentata
	Family			Poaceae			Rhamnaceae	Sapindaceae	Simmondsiaceae		Solanaceae		Verbenaceae		T	Cygophyllaccae         Larrea tridentata         I         Po         L         P         Gr         O         P         A         Ac         Us           Tribulus terrestris         I         Pt         Es         G         Gr         O         P         A         Ac         Us	

Epidermal cells

Arrangement: I, irregular; R, regular; and N, not observed.

- Shape: Po, polygonal; Ip, irregular with deep undulations; It, irregular with faint undulations; Ic, irregular with short undulations; Pt, polygonal with faint undulations; Rc, rectangular with short undulations; Rp, rectangular with deep undulations; Rt, rectangular with faint undulations; Re, rectangular; O, oval; N, not observed; 1, beam; 2, underside. Cell texture: L, smooth; Es, striated; Gra, granular; N, not observed.

· Size: P, small; M, medium; G, large; N, not observed.

- Cell wall: D, thin, Gr, thick; N, not observed.

- Shape: O, oval; Oa, elongated oval; Red, round; Ro, rhombic. Stomata

- Size: P, small; M: medium; G, large; N, not observed.

- Orientation: A: random; U, unidirectional; N, not observed.

- Arrangement: Di, diacytic; Ani, anisocytic; Ac, actinocytic; Ano, anomocytic; Tet, tetracytic; He, hexacytic; Pen, pentacytic; Ci, cyclocytic; Par, paracytic; Im, implanted; Ex, exposed; N, not observed. **Frichomes:** 

- Type: Bs, simple bicellular; Us, simple unicellular; Ps, simple multicellular; V, vesicular; Ud, unicellular with two branches; Pe, multicellular stellate; Ug, unicellular glandular; Gl, glands; Pg, multicellular glandular; Ub: unicellular with bulbous base.

Special structures: Dr. druses; Ra. raphidia; Gl. glands; Pa. papillae; Ta. tannins; Cr. crystals; Cc. cork cells; As. asperidia; Sc. silica cells.

Difference between beam and underside: Ee, stomata only on underside; Fc, cell shape; Tc, cell size; Te, trichomes more abundant on the underside; Th, trichomes more abundant on the beam; To, trichomes only on the underside. Although various modifications have been proposed for the development of the reference catalog (Catán *et al.*, 2003; Castellaro *et al.*, 2007; Arellano *et al.*, 2019), scraping has proven to be a simple and efficient method for the observation of epidermal structures. The application of this technique in the present study obtained a high percentage of observation of epidermal structures (95.95%) with a simple methodology, implying an economic saving in the purchase of inputs compared to the modifications proposed by other authors. In cases where some epidermal structures could not be clearly observed, especially because of the high density of stellate trichomes (*e.g.*, family Malvaceae), particular modifications should be made for certain plant groups (Catán *et al.*, 2007).

The combination of epidermal structures enabled the identification of the most common characteristics of certain families (Cactaceae, Fabaceae, Malvaceae, Poaceae and Zygophyllaceae), which can serve as a starting point for the development of identification guidelines for the plants present in the diet of herbivores (Desbiez and Santos, 2014). In fact, the collection of references and microhistological images from this catalog has already been used to characterize the diet of bighorn sheep, mule deer, white-tailed deer, and cattle at UMA Rancho Noche Buena (Peralta, 2020). Differences between beam and underside were found in 23% of the plant species analyzed, which is considered an important percentage. Consequently, given its absence from previous studies, future microhistological characterization works should consider both sides of the leaf.

### CONCLUSIONS

Scraping, which is a modification of the microhistological technique, provided a good description of the structures of the epidermis of the vegetation of UMA Rancho Noche Buena. Differences were found between the tissues of the beam and those of the underside of the same species; therefore, in some cases, they would appear to belong to different species. This reduces the estimation error regarding the composition of the diet of domestic and wild herbivores. In this way, the reference catalog developed for the vegetation of UMA Rancho Noche Buena can be used to determine with greater accuracy the diet of herbivores distributed in other parts of the Sonoran Desert.

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# ANNEX 1



Figure 1. A. Justicia californica; B. Ruellia californica; C. Trianthema portulacastrum; D. and E. Amaranthus palmeri beam and underside; F. Tidestromia lanuginosa; G. Ambrosia dumosa; H. Bebbia juncea.





Figure 2. A. Encelia farinosa; B. underside; C. Pectis rusbyi; D. Trixis californica; E. and F. Cordia parvifolia beam and underside; G. and H. Bursera laxiflora beam and underside.

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Figure 3. A. Bursera microphylla; B. Carnegiea gigantea; C. Cylindropuntia fulgida; D. Cylindropuntia thurberi; E. Ferocactus wislizenii; F. Lophocereus schottii; G. Pachycereus pringlei; and H. Stenocereus thurberi.