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Morphological range changes in Mexican *Astragalus radicans* Hornem. (Fabaceae: Galegeae): review of its taxonomy and nomenclature

EDUARDO ESTRADA CASTILLÓN^{1,4*}, JOSÉ ÁNGEL VILLARREAL QUINTANILLA^{2,5} & ALFONSO-DELGADO SALINAS^{3,6}

¹ Facultad de Ciencias Forestales, Universidad Autónoma de Nuevo León, C.P. 67700, Linares, Nuevo León, México.

² Departamento de Botánica, Universidad Autónoma Agraria Antonio Narro, C.P. 25315, Buenavista, Saltillo, Coahuila, México.

³ Instituto de Biología, Universidad Nacional Autónoma de México, Apartado Postal 70-367, Alcaldía Coyoacán 04510 Ciudad de México, México.

⁴ ✉ aeduardoestrada@prodigy.net.mx; <https://orcid.org/0000-0003-1061-9862>

⁵ ✉ javillareal00@hotmail.com; <https://orcid.org/0000-0001-9672-8693>

⁶ ✉ adelado@ib.unam.mx; <https://orcid.org/0000-0002-9322-9968>

*Author for correspondence: ✉ aeduardoestrada@prodigy.net.mx

Abstract

Continuing with the study of the *Astragalus* of Mexico and based on the measurement of 30 morphological characters and applying multivariate statistics, the two varieties of *Astragalus radicans*: *A. radicans* var. *radicans* and *A. radicans* var. *harshbergeri* were ranked at the species level and have their nomenclature updated. The results of the analysis allow the two infraspecific constitute isolated groups without overlapping between their individuals; therefore, they are raised from varieties to species, *Astragalus radicans* and *Astragalus harshbergeri*. A map with the updated distribution was included, and a dichotomous key is provided to identify the species. Both species are placed under the category of Threatened.

Keywords: *Astragalus*; morphometry; multivariate analysis; new status; Mexico

Resumen

Continuando con el estudio de los *Astragalus* de México y con base en la medición de 30 caracteres morfológicos y aplicando estadísticas multivariadas, las dos variedades de *Astragalus radicans*, *A. radicans* var. *radicans* y *A. radicans* var. *harshbergeri* fueron jerarquizadas a nivel de especie y su nomenclatura actualizada. Los resultados del análisis permiten que las dos categorías infraespecíficas constituyen grupos aislados sin traslape entre sus individuos, por lo tanto, se proponen nuevas jerarquías taxonómicas a nivel de especie como *Astragalus radicans* y *Astragalus harshbergeri*. Se incluye un mapa de distribución y se provee una clave dicotómica para identificar las especies. Ambas especies se ubican en la categoría de Amenazada.

Palabras clave: *Astragalus*; morfometría; análisis multivariado; estatus nuevos, México

Introduction

Astragalus L. (1753: 755) is the most diverse genus within Fabaceae (Barneby, 1989), comprising approximately 2,900 species (Zarre & Azani 2013), and subdivided in almost 245 sections around the world (Lewis et al, 2005). Its largest diversity occurs in temperate regions of the Northern Hemisphere, mainly in central-western Asia, including Turkey, Iran, and Afganistan (around 1500 species), Mongolia, central Asia, Siberia, Europe, northeastern China and Japan (around 620 species); Canada and western USA (376 species), South America (100 species) (Barneby 1989; Lewis & Shrire, 2003), and Mexico (almost 100 species). By including the recently new section *Sagitticarpus* A. E. Estrada, Villarreal & Encina (2020: 164), and the new species *Astragalus sagitticarpus* A. E. Estrada, Villarreal & Encina (2020: 166) (Estrada et al. 2020), the diversity *Astragalus* L. (1753: 755) in Mexico includes almost 30 sections, 99 species, and 47 infraspecific categories, almost all of these reported by Barneby (1964). Those sections,

distributed in Mexico and North America has been studied by molecular (Sanderson & Doyle 1993) and phylogenetic (Sanderson 1991) techniques, and have tested the monophyly of a large group of species belonging to them and their subsections. One of these sections, *sect. Strigulosi* Jones (1923: 184) group 28 species, four of these discovered after Barneby's monumental work (1964), those are: *A. cenorrhynchus* Barneby (1982: 78) (Barneby 1982), *A. mariosousae* A. E. Estrada, Villarreal & C. Yen (2005: 314) (Estrada *et al.* 2005), *A. spellenbergii* A. E. Estrada, S. González & Villarreal (2016: 91) (Estrada *et al.* 2016), *A. guanajuatensis* Rzed. & Calderón (2015: 2) (Rzedowski & Calderón, 2015). The highest diversity of this section distributes in Mexico, in fact, we can consider it as a Mexican section, since 24 (82%) of its species are native and occur only in Mexico and many of them are endemic and present narrow distribution; the other four species distributes in south of the USA. This section also contains, *Astragalus radicans* Hornem. (1815: 708), which includes *A. radicans* Hornem. var. *radicans* (1964: 164) and *A. radicans* Hornem. var. *harshbergeri* Barneby (1964: 165), with distinctive morphological characters, with creeping growth with the presence of adventitious roots at the leaf nodes, connate stipules, much wider than stems, and evidently petiolate leaves, unlike almost all species in Mexico, whose leaves are subsessile. To examine how far nomenclatural issues, apply to *Astragalus radicans* Hornem., it was first necessary to determine the current understanding of the classification of this species from published revisions and online resources. *Astragalus radicans* Hornem. var. *harshbergeri* Barneby (1964: 165), was originally published under the name *Atelophragma harshbergeri* Rydb. (1928: 160) from samples collected by J. W. Harshberger (13 Aug 1896) at Sierra Las Cruces at Salazar (Delegación Cuajimalpa). Both infraspecific taxa are endemic in central Mexico, distributed in Mexico City and two adjacent states (México and Michoacán). *Astragalus radicans* var. *radicans* has a wider distribution, since it is found in Michoacán state and surroundings of Mexico City, while *A. radicans* var. *harshbergeri* distributes only in the Valley of Mexico. Even though both taxa are sympatric in some areas, and that no phylogenetic studies have been conducted in these species, as far as we know, there are no reports that both taxa hybridize, but also, we have not found specimens that could represent hybrids between the two taxa, neither on national (ANSM, CFNL, ENCB, IEB, IBUG, MEXU) nor foreign herbaria (CAS, NYBG, TEX, US) (Thiers 2020). Two concepts can be applied in general terms for the taxonomic categories currently recognized in *Astragalus radicans*, one concerning a species consisting of two varieties, which is used when there are patterns of evident morphological variation and allopatric geographic distribution within the populations of a species but they overlap (Stuessy 2009), and the other one, that the two varieties of one species are actually two different species, this second statement can be applied to a species under the biological concept that defines it as a group of individuals with related morphological characteristics, defined geographic distribution as well as reproductive isolation (Crisci 1994). There are several methods to solve part of the infraspecific natural complexity and provide an aid to clarify dilemmas at the hierarchical levels, among the most common is the phenetic way or through multivariate statistical analyzes (Stuessy 2009). In this study we analyze the two varieties of *Astragalus radicans* to assess whether there is multivariate statistical support to recognize each of these as different species.

Material and methods

To analyze and compare the morphological variation between the two varieties of *Astragalus radicans*, specimens of different herbaria were studied, ANSM, CAS, CIIDIR, ENCB, IEB, MEXU, NYBG, US (acronyms of herbaria according to Thiers (2020)). Also, the digital images of these taxa stored in herbaria of University of Copenhagen (C), Botanisches Museum Berlin–Dahlem (B), Gray Herbarium (GH), Muséum National d'Histoire Naturelle (P), Academy of Natural Sciences (PH), New York Botanical Garden (NY), and Smithsonian Institution (US) were examined. To analyze the information to differentiate the taxa selected, a data matrix was prepared by selecting 30 morphological characters (Table 1) as well as several ecological variables. Twenty herbarium samples which represent the whole morphological variability along their distribution were evaluated: 14 samples of *A. radicans* var. *harshbergeri*, and 6 samples of *A. radicans* var. *radicans*. The information was classified (cluster analysis) by means of TWINSPAN (Hill & Šmilauer 2005) and by means of DCA (Detrended Canonical Analysis) by using CANOCO software ver. 4.5 (ter Braak & Šmilauer 2002). The natural distributions of both species are presented in a map.

Results

According to the measurements obtained from the studied samples, there are striking morphological differences between *A. radicans* var. *radicans* and *A. radicans* var. *harshbergeri*, standing out for the heterogeneity between both taxa, the diameter of stems, diameter of peduncles, distance between nodes, size of leaves, number of leaflets per leaf, size of leaflets, size of racemes and number of flowers per raceme. The figure 1 show the classification of the samples according TWINSpan, and two homogeneous groups emerge. The first dichotomy (letter D) constitutes two different groups of samples, Group I associate the samples 1–14 representing *A. radicans* var. *harshbergeri*, and Group II, agglomerate the samples 15–20, representing *A. radicans* var. *radicans*. In this latter group, the second dichotomy (letter D) clearly indicates that, the sample 20 is the most morphologically heterogenous among samples. The figure 2 shows the grouping of the samples along Axes I and II in the DCA, both axes accumulate 78.5% of total variance; all samples are positively correlated to the Axis 1, it shows the formation of two groups of samples, Group I include the samples 1–14 corresponding to *A. radicans* var. *harshbergeri*, while Group II includes the samples 15–20 corresponding to *A. radicans* var. *radicans*. The Group I is homogeneously compact, while Group II it is less compact, showing greater heterogeneity in the measurements of its structures and this is observed again in the location of sample 20, the most heterogeneous or morphologically dissimilar from the rest of the group, which brings it closer to Group I, however, sample 20 has the general morphology of plant of Group II, keeping it as a member of this group. It has very compact growth, but still with at least double in length size, longer stipules, leaves, leaflets, peduncles, number of flowers per raceme, and longer flower size than the ones of *A. radicans* var. *harshbergeri*. Since both groups represent entities lacking continuity between them, we propose the change of the *radicans* and *harshbergeri* varieties to a species category. Their distribution is showed in the figure 3. Barneby (1964) and Rzedowski & Rzedowski (2001) provide dichotomous keys to identify both taxa, however, they do not include the size of the stipules or the size of the petals, especially the banner and the wings. These three strongly contrasting morphological characters between both taxa and easily discernible are included in this study together with their updated distribution.

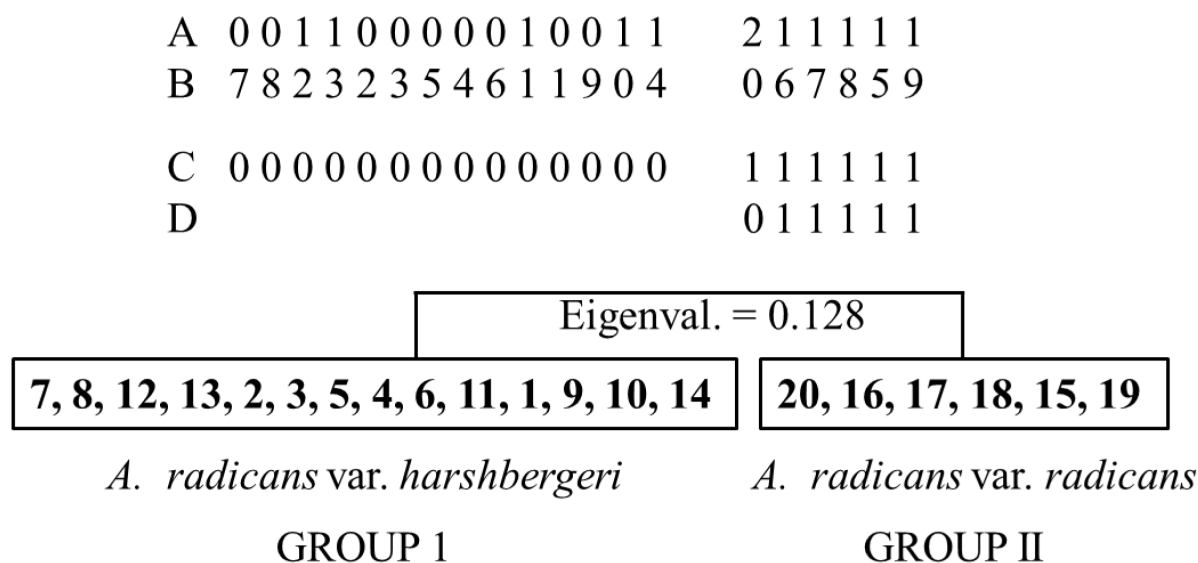


FIGURE 1. Classification of 20 samples by means of TWINSpan, rows A and B represent the number of the sample 07, 08, 12, etc. The row C represent the first dichotomy 0's and 1's. The Group I associate 14 samples, 7 to 14 and represent *A. radicans* var. *harshbergeri*; the Group II agglutinates 6 samples, 20 to 19 and represent *A. radicans* var. *radicans*. The row D represents the second dichotomy (showed only for Group II), the only 0 means that the sample 20 is the most heterogeneous one of the Group II.

Dichotomous key to identify the studied taxa

1. Stipules 3–5.5 mm long; leaves up to 10 cm long; leaflets 17–26, 4–12 mm long; mature racemes with 5–12 flowers and peduncles measuring 2–7 cm long; banner 11.5–13 (rarely up to 15) mm long; wings petals 11–12 mm long; most commonly 2900–3330 m *A. harshbergeri*
- Stipules 7–12 mm long; leaves 10.5–30 cm long; leaflets 27–38, 18–28 mm long; mature racemes with 20–34 flowers and peduncles (<10)20–25.5 cm long, rarely longer, if shorter than 10 cm, then the racemes with 20 or more flowers; banner 14–15 mm long; wings 12.5–15 mm long; most commonly 2200–2400 m alt..... *A. radicans*

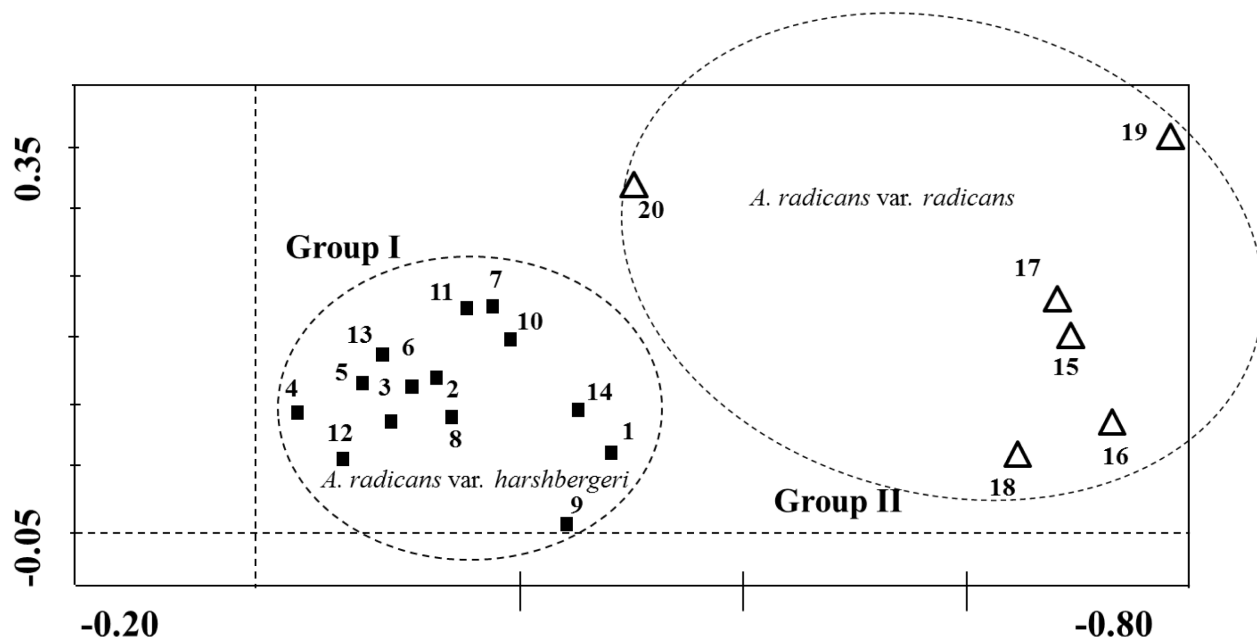


Fig.2.

FIGURE 2. Detrended Correspondence Analysis showing the two groups of samples, Group I agglutinates 14 samples 1–14, representing *A. radicans* var. *harshbergeri*, Group II agglutinates 6 samples, 15–20, representing *A. radicans* var. *radicans*. The sample 20 is the most heterogeneous of the group, showing all the morphological structures shorter than the rest.

TABLE 1. Morphological characters measured and used to differentiate both varieties of *Astragalus radicans*.

Stem diameter (mm)	Banner length (mm)
Distance between nodes (cm)	Banner wide (mm)
Leaf size (cm)	Wing length (mm)
Petiole size (cm)	Wing wide (mm)
Rachis leaf size (cm)	Wing claw length (mm)
Leaflets length (mm)	Wing blade length (mm)
Leaflets wide (mm)	Keel length (mm)
Peduncle length in flowers stage (cm)	Keel wide (mm)
Peduncle length in fruit stage (cm)	Keel claw length (mm)
Raceme length in flowers stage (cm)	Keel blade length (mm)
Raceme length in fruit stage (cm)	Stamen column length (mm)
Number of flowers per raceme	Fruit length (mm)
Calyx size (mm)	Fruit wide (mm)
Calyx tube size (mm)	Number of ovules
Calyx teeth size (mm)	Seed length (mm)

Astragalus radicans and *A. harshbergeri* show sufficiently stable and homogeneous morphological characteristics between them that they can be recognized as two different species. To date, no specimens have been collected that suggest, at least morphologically that there is hybridization between them, since the reviewed and measured specimens, according to their variation, belong to one or the other species. The classification and grouping of samples using multivariate techniques with TWINSpan and CANOCO, although both techniques derive from different mathematical and statistical methods, similarly group the two groups of samples. The morphological variability of both species concerning to stipule size, leaf size, leaflets size and number per leaf, peduncle size, number of flowers per raceme, and petal size, are the main characteristics that allow differentiating both species. In fact, it is easier to confuse *Astragalus harshbergeri* with *A. lyonnetii* Barneby (1964: 162) than with *A. radicans*. Both species are sympatric and are physiognomically almost identical, however *A. lyonnetii* can be differentiated due to the absence of roots in the nodes of the flowering nodes, its stipules are not twice as wide as the stems, and by its almost 3 mm long stipe (1 mm long in *A. harshbergeri*). Ecologically, *A. radicans* distributes in lower altitudes, 2200–2400 m, associated to low grasslands, secondary vegetation, and abandoned crop areas, while *A. harshbergeri* inhabits higher altitudes, 2900–3330 m alt., rarely found at lower altitudes (2250 m), almost always associated to conifer forest.

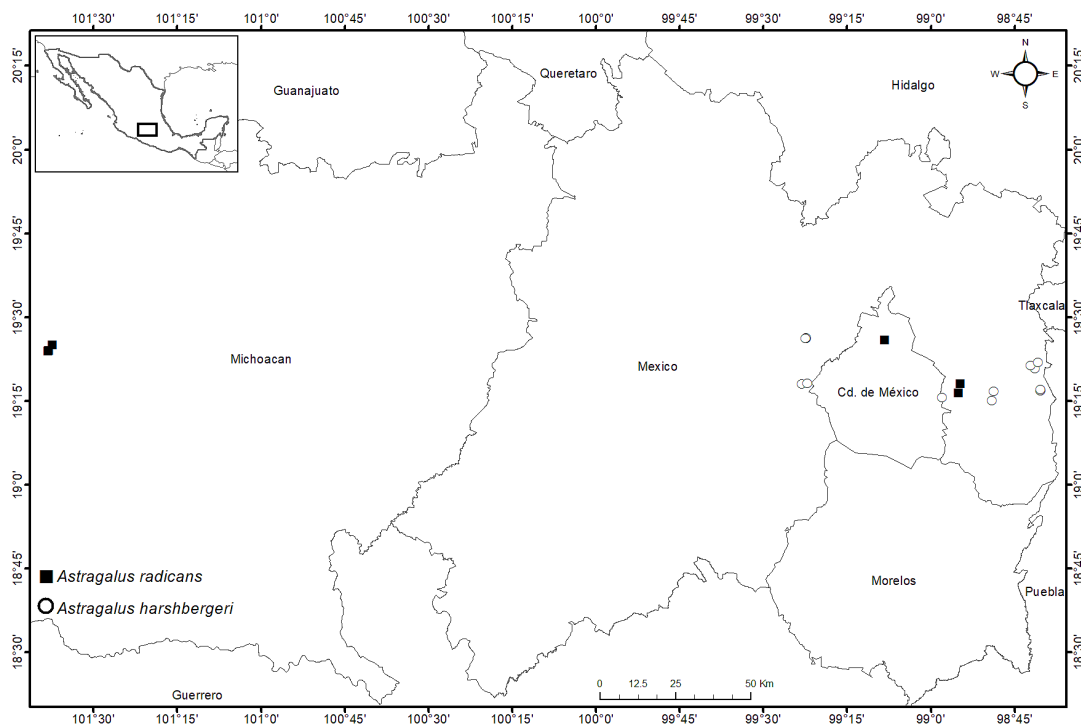


FIGURE 3. Map showing the distribution of *Astragalus radicans* and *A. harshbergeri* in Mexico.

Taxonomy

Astragalus radicans Hornem. var. *harshbergeri* Barneby originally published as *Atelophragma harshbergeri* Rydb. (1928: 160) is recognized at the species rank, and accordingly, a new combination *Astragalus harshbergeri* (Rydb.) A.E. Estrada, Villarreal & A. Delgado is proposed. Species rank for this taxon is advocated in this study on basis of morphological distinctions between this species and the closely related *Astragalus radicans* Hornem. In addition, to facilitate further taxonomic research, we consider it necessary to typify all the names of the taxa to stabilize their usage and synonymy.

Astragalus harshbergeri (Rydb.) A.E. Estrada, Villarreal & A. Delgado, *comb. nov. & stat. nov.* (Fig. 4 A–J).

Astragalus radicans Hornem. var. *harshbergeri* Barneby (1964: 165).

Basionym: *Atelophragma harshbergeri* Rydb. (1928: 160).

TYPE:—MEXICO. Mexico City, Salazar, Sierra de Las Cruces, alt. 11000 ft. 13 Aug 1896, *J. W. Harshberger 20* (holotype: GH!; isotypes: PH!, PH00005764; PH00005765; US!).

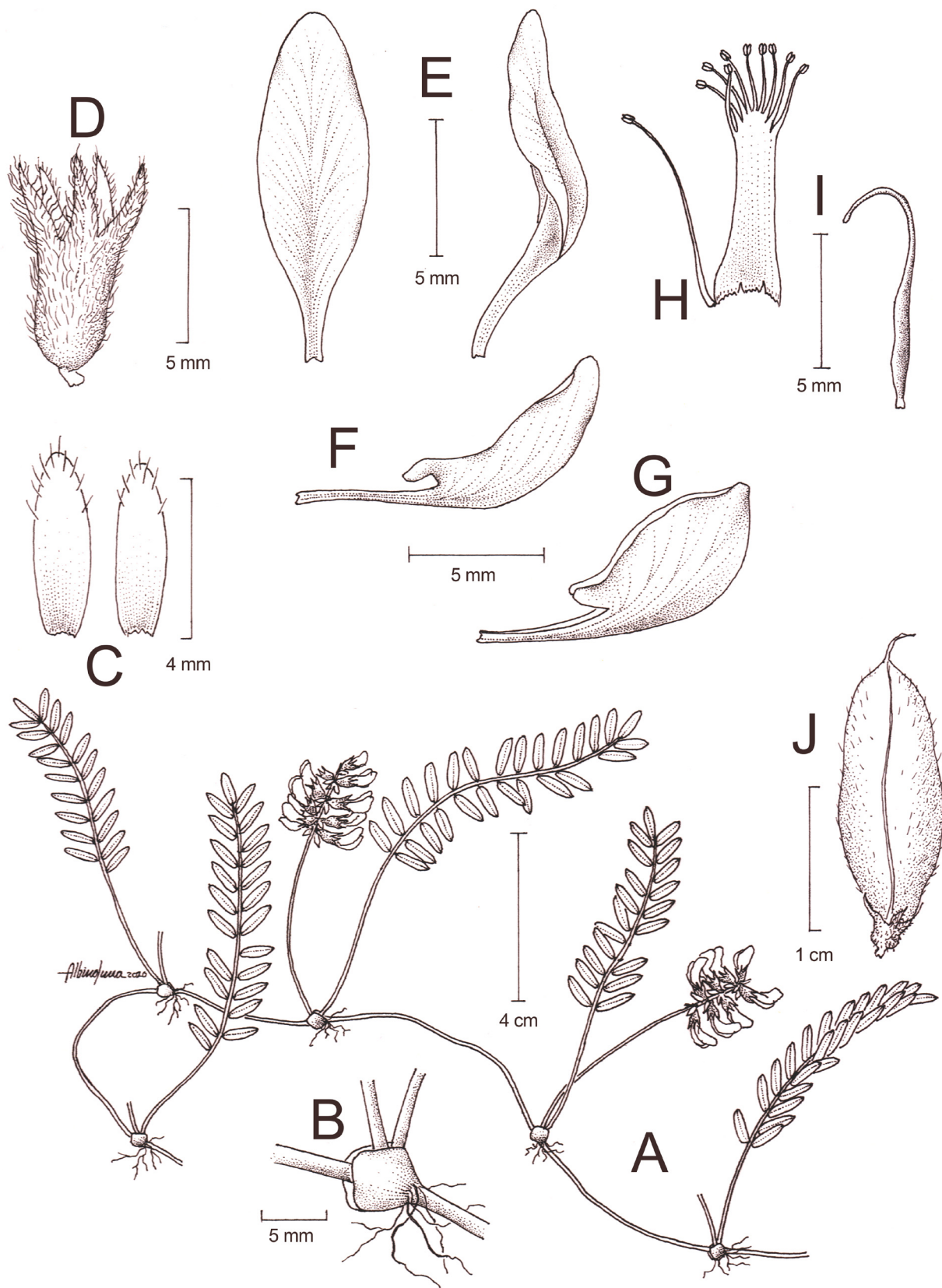


FIGURE 4. *Astragalus harshbergeri*. A. Whole plant showing leaves, inflorescences, and flowers. B) Stipules, much wider than stem. C) Bracts. D) Calyx. E) Banner front view (left) and profile view (right). F) Wing. G) Keel. H) Stamens. I) Ovary. J) Fruit.

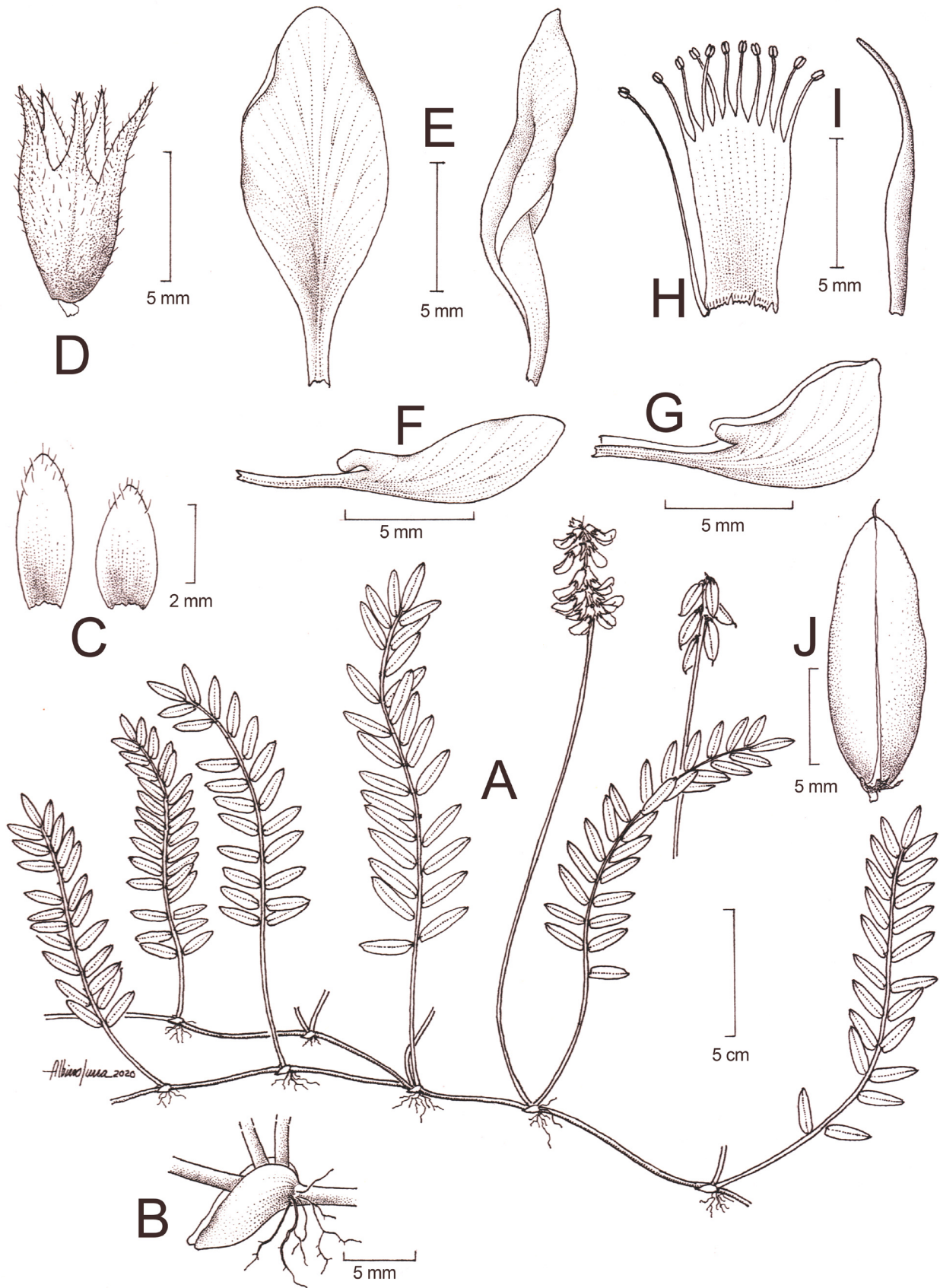


FIGURE 5. *Astragalus radicans*. A. Whole plant showing leaves, inflorescences, flowers, and fruits. B) Stipules, much wider than stem. C) Bracts. D) Calyx. E) Banner front view (left) and profile view (right). F) Wing. G) Keel. H) Stamens. I) Ovary. J) Fruit.

Plant perennial. Stems up to 80 cm long, 10–30 cm tall, prostrate, creeping, commonly rooting in nodes, the trichomes up to 0.6 mm long, appressed. **Stipules** 3–5.5 mm long, elliptic, obovate to almost orbicular, connate, attached to the half of its length or little more, much wider than stems. **Leaves** up to 10 cm long, leaflets 17–26, 4–12 mm long, lanceolate, oblong, elliptic, obtuse, truncate and apiculate, adaxially glabrate or sparsely so, abaxially strigose. **Peduncles** 2–7 cm long, erect, perpendicular to foliage growth, trichomes black; the racemes short, 1–2.5 cm long, flowers 5–12. **Flower bracts** 2–5 × 1–1.5 mm, ovate to wide ovate. **Flowers** yellow, cream, yellowish to ochroleucous; the calyx 5.5–8.9 × 2.7–3.6 mm, strigose, trichomes mainly black, the tube 3–6 mm long, campanulate, the teeth 2–3.5 mm long, lanceolate, ventral pair wider; the banner 11.5–13 (very rarely up to 15 mm) × 4–6 mm, elliptic, oblong to oblanceolate, shallowly notched, the wings 11–12 × 2–2.5 mm, the claw 5–6.5 mm long, the blade 5.5–7.3 mm long, narrowly oblong oblique, elliptic; the keel 8–10 × 2.1–3 mm, the claw 4–4.5 mm long, the blade 4–6 mm long, distally oblique; androecium diadelphous, stamens 10, 8.5–9 mm long, anthers yellow, 0.5 mm long; gynoecium 9 mm long, ovary 4.5 mm long, oblong, glabrate, style linear, 4.5–5 mm long, stigma minute. **Pod** deflexed, subsessile or very shortly stipitate (stipe 0.4–1 mm long), oblong to elliptic, 15–21 × 5–8.2 mm, somewhat dorsoventrally compressed, basally rounded or narrowed, distally abruptly apiculate, ventrally carinate, dorsally open and shallowly sulcate, the valves rigid, papery, strigose, trichomes black or rarely also with white trichomes, mixed, septum incomplete; ovules 14–24; seeds 1.8–2.6 mm long, mitten shape, pale–brown to brown, opaque.

Conservation status:—According to the herbarium samples reviewed for this species, its actual distribution is restricted to the mountains surrounding the Valley of Mexico. The species is proposed to be placed under the category of Threatened under the Official Mexican Standard NOM-059-SEMARNAT-2010 (SEMARNAT 2010), since it could be in danger of disappearance in the short to medium term, if factors that adversely affect their viability continue to operate (e.g., damage or modification of habitat or directly reducing the size of their populations).

Additional specimens examined: **Estado de México:** not locality, 3200 m snm, *L. W. Boege 290* (MEXU); near Río Frío at Llano Grande, 3200 msnm, *A. J. Sharp 4485* (MEXU); Ixtapaluca, Estación Experimental Forestal de Zoquiapan, 3300 msnm, *M. C. Obieta 40* (MEXU); Municipio Ixtapaluca, Estación Experimental de Investigación y Enseñanza de Zoquiapan, 8 km al sur de Río Frío, casi 200 m al N de Aculco, sobre camino seis, 3220 m snm, *R. Vega A. 196* (ENCB); Municipio Ixtapaluca, Llano Pinahua, 10 km al sur de Llano Grande, *Rzedowski 36796* (IEB, MEXU); Llanos de Salazar, 2900 m snm, *F. Medellin, G.C. Rzedowski s.n.* (ENCB); Municipio Lerma, 3000 m snm, Parque Nacional Miguel Hidalgo y Costilla (La Marquesa), llanos, camino a Chalma, *Sánchez-León 66* (ENCB); Municipio Ixtapaluca, Estación Experimental y de Investigación y Enseñanza de Zoquiapan, 8 km al sur de Río Frío, Llano Xaxalpa, orilla de camino 5, pie del Cerro El Papayo, 3330 m snm, *R. Vega A. 381* (ENCB); Municipio Tlalmanalco, Llano Atlhuiyán, 10 km al E de San Rafael, 3200 msnm, *Rzedowski 37875* (IEB); Municipio Chalco, 1 km al N. de Llano Grande, en las faldas del el Cerro El Telapón, 3200 m snm, *Rzedowski 18438* (ENCB); Municipio Zoquiapan, Llano Grande, cerca de Río Frío, 3200 m snm, *R. Cruz C. 1268* (ENCB); Municipio Ixtapaluca, Llano Tepochaico, 10 km al sur de Río Grande, 3250 m snm, *Rzedowski 36807* (ENCB); Municipio Naucalpan, Villa Alpina, 3150 m snm, *Rzedowski 36323* (ENCB). **Ciudad de México:** Delegación Ixtapalapa, 2 km al E de Ixtapalapa, cerca de la estación XEW, *Rzedowski 20088* (ENCB).

Astragalus radicans Hornem. (1815: 708). (Fig. 5 A–J)

TYPE:—Mexico: “Hab. F. intr. 1808 *sub. nom.* Astr. Radicans Humb.”. *Humboldt & Bonpland s.n.*

Lectotype (designated here):—MEXICO, at C, C10011808; islectotypes, at C, C10011807). Grown from seed at the Botanical Garden of Copenhagen. The specimen that best matches the description was chosen [digital image! Available at http://plants.jstor.org/stable/10.5555/al.ap.specimen.c10011807?searchUri=filter%3Dname%26so%3Dps_group_by_genus_species%26Base%26Query%3DAstragalus%2Bradicans].

Astragalus reptans Humb. & Bonpl. ex Willd. (1816: 88).

“*Astragalus reptans de Humb. and Bonpl.*... Habitat in Mexico.” *A.J.A. Bonpland s.n.* (lectotype at Herbarium Willdenow in Herbarium Berlin (B), B–W 13988–010, islectotypes (designated here):—MEXICO, at P, P00135206, P00135207; P00135208). Wrongly cited as *Astragalus reptans* Willd. (1816: 88).

Craccina reptans (Willd.) Steven (1832: 266).—*Tragacantha reptans* Kuntze (1891: 947);—*Atelophragma reptans* Rydb. (1928: 159).

Perennial. Stems up to 1 m long, prostrate, creeping, rooting in nodes, the trichomes up to 0.6 mm long, appressed. **Stipules** 7–12 mm long, elliptic, obovate to almost orbicular, connate, attached to the half of its length or little more, wider than stems. **Leaves** 10.5–30 cm long, leaflets 27–33, 18–28 mm long, oblong, elliptic, obtuse, truncate and apiculate, adaxially glabrate or sparsely so, abaxially strigose. **Peduncles** 7–27 cm long, erect, perpendicular to foliage growth,

trichomes black; the racemes 2.5–6 cm long, flowers 20–34. **Flower bracts** oblong to ovate 2–4.4 mm long, white, subglabrate or with very few black or white trichomes in margins. **Flowers** yellow, cream, yellowish to ochroleucous; the calyx 7–9 × 3–3.8 mm, strigose, trichomes mainly black, the tube 4–5 mm long, campanulate, the teeth 2.5–4 mm long, lanceolate, ventral pair wider; the banner 14–15 × 5–7 mm, elliptic, oblong to oblanceolate, shallowly notched, the wings 12.5–15 × 2.2–2.7 mm, the claw 5–6 mm long, the blade 7–9.5 mm long, narrowly oblong oblique, elliptic; the keel 9–12.6 × 2.3–3.4 mm, the claw 4.4–5.6 mm long, the blade 4.2–6.5 mm long, distally oblique; androecium diadelphous, stamens 10, 8.5–9.10.5 mm long, anthers yellow, 0.5 mm long; gynoecium 9–9.6 mm long, ovary 4.5–5.3 mm long, oblong, glabrate, style linear, 4.5–5.7 mm long, stigma minute. **Pod** deflexed, subsessile or very shortly stipitate (stipe 0.4–1 mm long), oblong to elliptic, 15–22 × 5–8.3 mm, dorsoventrally compressed, basally rounded or narrowed, distally abruptly apiculate, ventrally carinate, dorsally open and shallowly sulcate, the valves rigid, papery, strigose, trichomes black or rarely also with white trichomes, mixed, septum incomplete; ovules 19–30; seeds 1.8–2.6 mm long, mitten shape, pale–brown to brown, opaque.

Conservation status:—According to the herbarium samples reviewed for this species, its actual distribution is restricted to Valley of Mexico (Mexico City and State of Mexico) and Michoacán. As the other species, this one is also proposed to be placed under the category of Threatened under the Official Mexican Standard NOM-059-SEMARNAT-2010 (SEMARNAT 2010), since it could be in danger of disappearance in the short to medium term, if factors that adversely affect their viability continue to operate (e.g., damage or modification of habitat or directly reducing the size of their populations).

Additional specimens examined:—**Estado de México:** orilla de una milpa situada al E del sitio Arqueológico Tlapacoyan, cerca de la autopista, *J. Espinoza G. s.n.*, (NYBG); Municipio de Chalco, 2 km al S. de Tlapacoyan, 2250 m snm, *Rzedowski 29055* (ENCB); Municipio Chalco, 2 km al S. de Tlapacoyan, 2250 m snm, *Rzedowski 29055* (ENCB). **Mexico City:** Chapultepec, *Bilimek 107* (NYBG); knoll of low meadows, Valley of Mexico, 7300 ft, *C. G. Pringle 6315* (NYBG). **Michoacán:** Municipio Santa Clara del Cobre, Ejido Casas Blancas, 2400 m snm, *J. M. Escobedo 1044* (IEB); Municipio Santa Clara del Cobre, 2 km rumbo al Cerro San Miguel, *J. M. Escobedo 1470* (IEB).

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