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Source: *Lundellia*, 24(1) : 11-23

Published By: The Plant Resources Center, The University of Texas at Austin

URL: <https://doi.org/10.25224/1097-993X-24.1.11>

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YUCCA CARRII (ASPARAGACEAE), A NEW SPECIES FROM THE NORTHERN GULF COAST PRAIRIE OF TEXAS

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Abstract: *Yucca carrii*, a new species from the northern Gulf Coastal Prairie of Texas is described. Morphology indicates that *Yucca carrii* belongs to Section *Chaenocarpa* (the dry-fruited yuccas). A lack of observed fruit production to date and reduced pollen viability indicates that this species may be of hybrid origin. The sole pollinator, the yucca moth, has not been documented within the species' range; it may be extirpated. Although known from few populations, the wide-ranging distribution of this species along the north Texas coast, ecological niche specificity and morphological distinctiveness indicates this taxon may have a long history in the area and persists by way of vegetative reproduction. It may be vulnerable to extinction and conservation measures are warranted. DNA analyses are needed to determine the origin and relationship of *Y. carrii* within *Yucca* L. Examination of the type of *Y. tenuistyla* Trel. revealed *Y. tenuistyla* is a *nomen confusum* that was described from a mixed collection of three yucca species, none of which is *Y. carrii*.

Resumen: Se describe *Yucca carrii*, una nueva especie de la pradera costera del norte del golfo de Texas. La morfología indica que *Yucca carrii* pertenece a la Sección *Chaenocarpa* (las yucas de frutos secos). La falta de producción de fruta observada hasta la fecha y la viabilidad reducida del polen indican que esta especie puede ser de origen híbrido. El único polinizador, la polilla de la yuca, no ha sido documentado dentro del rango de la especie; puede estar extinto. Aunque se conoce a partir de pocas poblaciones, la amplia distribución de esta especie a lo largo de la costa norte de Texas, la especificidad del nicho ecológico y la distinción morfológica indican que este taxón puede tener una larga historia en el área y persiste mediante la reproducción vegetativa. Puede ser vulnerable a la extinción y se justifican las medidas de conservación. Se necesitan análisis de ADN para determinar el origen y la relación de *Y. carrii* dentro de *Yucca* L. Examen del tipo de *Y. tenuistyla* Trel. reveló que *Y. tenuistyla* se describió a partir de una colección mixta de tres especies de yuca, ninguna de las cuales es *Y. carrii*.

Key words: ASPARAGACEAE, *Yucca* L., *Yucca carrii*, *Yucca tenuistyla*, yucca moth, sea level rise, global warming, plant conservation, species vulnerability, Gulf Coast Prairies and Marshes Ecoregion, Texas, U.S.A.

INTRODUCTION

While examining herbarium specimens from Galveston County, Texas in the late 1990s, the first author came across collections made by George L. Fisher in 1915 and Floyd R. Waller and J.A. Bauml in 1971 of an unknown species of *Yucca* L. from a coastal prairie remnant in San Leon. In May 2003, she found a few small, scattered populations growing along the fringes of Dickinson Bayou and Galveston Bay. Soon after, Bill Carr provided the locations of a

yucca he had seen while botanizing on the Texas Nature Conservancy's Texas City Prairie Preserve, which turned out to be the same species that Fisher, Waller and Bauml had collected. In 2006, we confirmed the presence of *Y. carrii* at Brazoria National Wildlife Refuge. Since then, other small populations of this plant in similar habitat have been found in Brazoria, Chambers, Galveston, Harris and Matagorda counties. The new species is here described, illustrated, distinguished by key from other yucca species that grow in the area. The systematic

relationship within *Yucca* L. and conservation status are discussed.

Yucca carrii Clary & T.P. Adams, sp. nov. (Fig. 1).

TYPE: UNITED STATES. TEXAS. Galveston Co.: Texas City, Texas Nature Conservancy (TNC) Texas City Prairie Preserve, plants growing between TNC Visitor's Center and banks of Moses Lake, 29.424306, -94.958303, 2.76 m, 20 May 2005, *Clary 427* (HOLOTYPE: TEX!; ISOTYPES: MO!, US!).

This species is distinguished by an acaulescent rosette, flaccid grass-like leaves with reddish-brown, smooth margins, 1 to several tall (3+ m) inflorescences per clump, often falling over when mature, a broadly obovate-elliptic panicle topped by a single panicle branch, puberulous-tomentose panicle branches that spread upward and outward, wavy in shape when dry, a long, slender pistil (to 3.7+ cm), ovary (green) and style (white) of near equal length, style lobes deeply divided (2-4+ mm) and stigma lobes notched (0.5-1.0+ mm deep), often unequal in length.

PLANTS acaulescent solitary rosettes or forming small clumps (2-10 plants). ROSETTE asymmetrical, untidy, with erect younger leaves and flaccid older leaves. MATURE LEAF BASE white to pale yellow, the base slightly wider than the widest part of blade, proximal half of blade narrow, stiff and thickened along raised midrib. BLADE OF MATURE LEAF flat, narrowly rhombic-oblan-ceolate, 55-75+ cm in length, narrowest at union with base, gradually broadened to middle and slightly above, tapering to a long acuminate apex with a sharp, thin, narrow brown to reddish-brown spine, greatest width 1.7-2.5+ cm, distal half often in-rolled, dried out and dying, young leaves blue green with yellowish-brown, smooth margins, mature leaves green with brown to reddish-brown, smooth margins, lacking exfoliating fibers. INFLORESCENCE 2.5-3.0+ m tall, often falling to ground under its weight, scape of equal length or longer than panicle, green to maroon, with bracts, scape smooth

below panicle. PANICLE raised high above leaves, 1.25-2.0+ m in length, branched throughout, obovate, axis with both shorter branches (13-17+ cm) and longer branches (22+ cm) numerous but uncrowded by spreading outward, panicle branches puberulous-tomentose, green to maroon, flowers pendant. PERIANTH 3.9-5.3+ cm in length, white to creamy white, base cup-shaped, with thin, (midline of segment not thickened) acute, ovate inner segments and ellipsoid outer segments with a darkened tip. FILAMENTS slender, reaching to above narrowed base of style, 1.9-2.3+ cm in length, strap-shaped, slightly swollen at tip, hairs hirsute and strigose. ANTERS 1.0-2.0+ mm in length. PISTIL 3.1-3.7+ cm in length with ovary and style of almost equal length. OVARY pale green, 1.5-2.6+ cm in length, oblong-ovate, neck short, constricted, shoulders well-defined, prominent, slightly wider than base. STYLE white, long, slender, tapering, 1.3-1.8+ cm in length, papillae scabrate to verrucate, extending the length of the style, lobes deeply divided (2-4+ mm). STIGMA LOBES white, notched (0.5-1.0+ mm deep), often unequal in length, upright, slightly spreading outward. STIGMAS white. FRUIT unknown.

FLOWERING: May and June.

HABITAT AND FREQUENCY: Plants infrequent, in small, scattered clumps of rosettes where they occur. Of note is the ability of plants to grow on banks immediately adjacent to salt and brackish waters and in seasonally inundated coastal flatlands. On Late-Pleistocene Beaumont Formation, Brazoria, Chambers, Galveston Harris and Matagorda counties. Growing on flat, poorly drained clay and clay loam soils (Francitas Clay, Verland Silty Clay Loam, Morey Silt Loam and Ijam Clay series) (NRCS Web Soil Survey, 2013) of coastal prairie, dredged spoil bank on or adjacent to Galveston Bay, banks of bayous, Laguna Madre Intracoastal Waterway (ICWW), in swales of mima mounds and on edge of eroding coastal chenier plain woodlands on Veston soil (NRCS Web Soil Survey, 2013).

DISTRIBUTION: *Yucca carrii* plants have been observed in five counties to date:

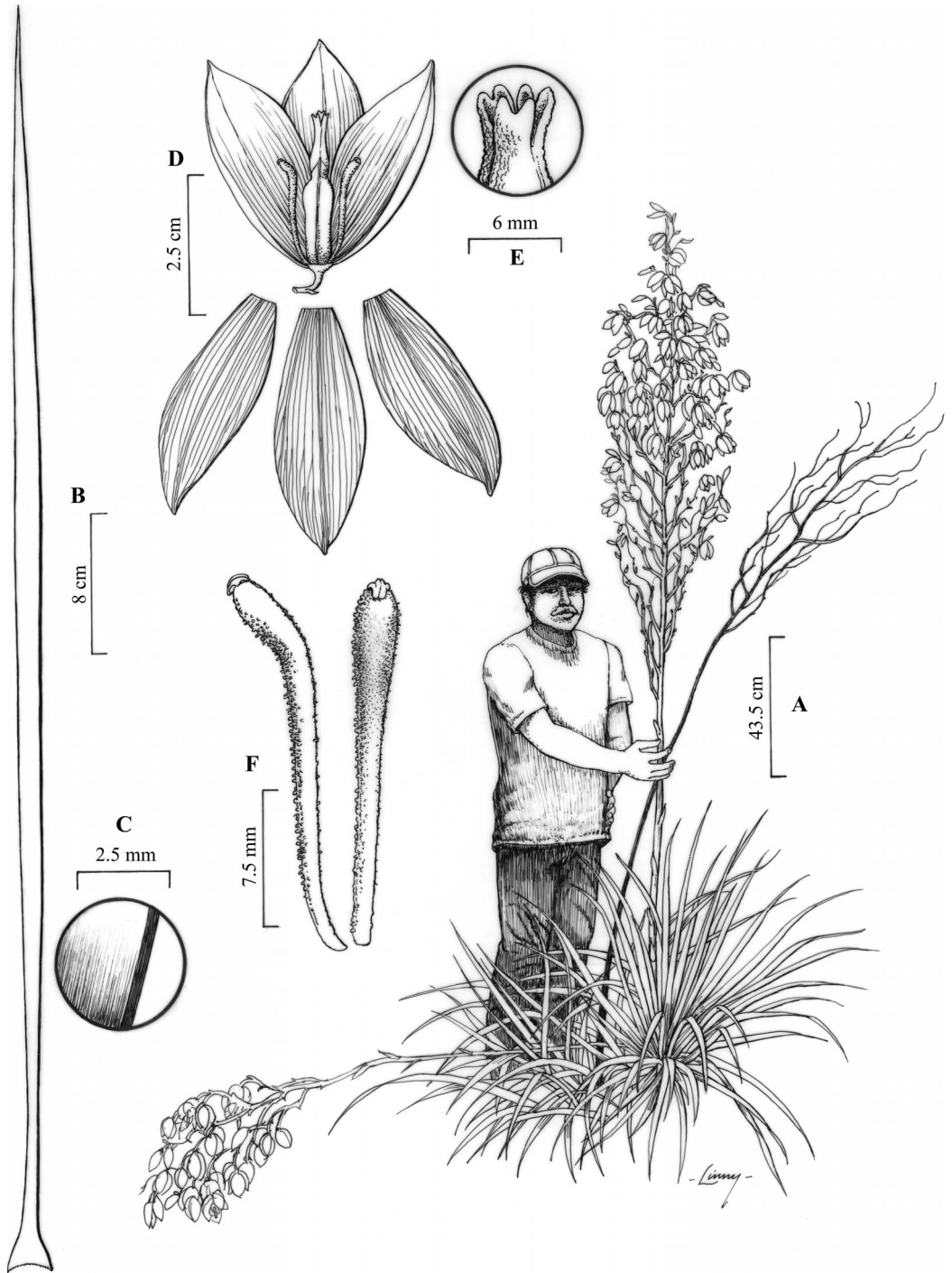


FIG. 1. *Yucca carrii*. A. Habit. B. Representative leaf, dorsal surface. C. Leaf margin, detail. D. Flower. E. Stigma lobes, detail. F. Stamens, enlarged, front view and side view.



FIG. 2. *Yucca carrii* type locality. (Clary 427 [TEX-LL]). Texas Nature Conservancy (TNC) Texas City Prairie Preserve. Photo by J.H. Clary.

Galveston, scattered around San Leon on the north side of Dickinson Bayou (about 30 plants total), along the Texas City sea wall and the TNC Texas City Prairie Preserve (about 40 plants total; Fig. 2); Chambers, on the banks of Cotton Lake (about 10 plants); Brazoria, on the south side of Chocolate Bayou (about 50-100 plants), Brazoria National Wildlife Refuge (BNWR) (Fig. 3). Plants have been identified by photograph in Harris Co., the San Jacinto Battleground State Historic Site (Eric Keith), and Brazoria, Galveston and Matagorda counties (iNaturalist.org, <https://www.inaturalist.org>). Limited surveys on existing roads have been undertaken to find more populations in similar habitat along the coast in Galveston (Galveston Bay, Galveston Island) and surrounding Chambers (Anahuac National Wildlife Refuge), Harris and Matagorda (San Bernard National Wildlife Refuge, Big Boggy National Wildlife Refuge) counties, but none have been found.

ASSOCIATED SPECIES: Coastal Prairie - *Baccharis halimifolia* L., *Baptisia* sp., *Eryngium yuccifolium* Michx., *Euthamia* sp., *Liatris acidota* Engelm. & A. Gray, *Mimosa strigillosa* Torr. & A. Gray, *Paspalum plicatulum* Michx., *Pennisetum glaucum* (L.) R. Br., *Polytaenia texana* (J.M. Coult. & Rose) Mathias & Constance, *Rubus* sp., *Sida spinosa* L., *Spartina spartinae* (Trin.) Merr. ex Hitchc., *Yucca louisianensis* Trel., *Schizachryium scoparium* (Michx.) Nash.

Edge of Chenier Plain Woodland - *Ulmus crassifolia* Nutt., *Celtis laevigata* Willd., *Xanthoxylum* sp., *Maclura pomifera* (Raf.) C.K. Schneid., *Prunus mexicana* S. Watson, *Baccharis halimifolia* L., *Cornus drummondii* C.A. Mey., *Sabal minor* (Jacq.) Pers., *Tamarix* sp., *Opuntia* sp., *Ilex vomitoria* Aiton, *Malvaviscus arboreus* Dill. ex Cav. var. *drummondii* (Torr. & A. Gray) Schery, *Onosmodium bejariense* DC. ex A. DC. var. *hispidissimum* (Mack.) B.L. Turner, *Centaurea americana* Nutt., *Allium canadense* L., *Stachys floridana* Shuttlw. ex



FIG. 3. Habit. *Yucca carrii* population, Brazoria National Wildlife Refuge. Photo by T.P. Adams.

Benth., *Sambucus* sp., *Verbena stricta* Vent., *Hibiscus laevis* All., *Rubus* sp., *Ibervillea* sp., *Passiflora incarnata* L., *Vitis mustangensis* Buckley, *Campsis radicans* (L.) Seem. ex Bureau, *Borrhichia* sp., *Cuscuta* sp., *Cissus* sp., *Spartina spartinae* (Trin.) Merr. ex Hitchc., *Schoenoplectus americanus* (Pers.) Volkart ex Schinz & R. Keller, *Phragmites* sp., *Sorghum halepense* (L.) Pers., *Elymus canadensis* L., *Chloris* sp.

ADDITIONAL SPECIMENS EXAMINED: **UNITED STATES. TEXAS. Galveston Co.:** San Leon, 5 June 1915, *Geo L. Fisher 1542* (US 504959); San Leon, Dickinson Bayou at end of Avenue W, small population in prairie site on bank of bayou, 15 May 1971, *Floyd R. Waller and J. A. Bauml 3618* (TAES 554409); San Leon, at SH 146 and NE side of Dickinson Bayou, in disturbed area on banks of bayou, just east of SH 146 approach to bridge over bayou, part of small scattered population between Avenue T (E of SH

146), and Avenue W (W of SH 146), 2.76 m, 23 May 2003, *Clary 417* (TEX-LL); San Leon, at SH 146 and NE side of Dickinson Bayou, in disturbed area on banks of bayou, just east of SH 146 approach to bridge over bayou, part of small scattered population between Avenue T (E of SH 146), and Avenue W (W of SH 146), growing on edge of bank above water line, 4.0 m, 23 May 2003, *Clary 418* (TEX-LL); Texas City, Texas Nature Conservancy Texas City Prairie Preserve, N end of TNC preserve on N side of floodgate that protects Moses Bay from Galveston Bay, single clump found growing on east side of levee, 70 m W of pipeline, 28 m from W of water's edge of Dickinson Bayou, on grassy edge of bayou, 20 May 2005, *Clary 428* (TEX-LL); **BRAZORIA CO.:** Brazoria National Wildlife Refuge. On flat, poorly drained clay and clay loam soils of coastal prairie, south of FM 2004 and on south side of Chocolate Bayou, plants found

in swales of *mima* mounds populated by *Y. louisianensis*, 20 May 2006, Clary 432 (TEX-LL); CHAMBERS Co.: On the eroding banks of Cotton Lake, 7 June 2013, Clary 442 (TEX-LL).

ETYMOLOGY: This species is named in honor of William R. Carr, Texas botanist, conservationist, teacher, most prolific plant collector, writer and friend who generously shares his knowledge of the Texas flora for the enrichment of all who study plants.

DISCUSSION

EARLY COLLECTIONS: Although the history of plant collecting in the area dates back to Ferdinand Lindheimer's 1843 collection of yuccas on nearby Galveston Island (see MO 148758), *Y. carrii* is poorly represented in the herbarium record. One specimen from Fisher in 1915 (US 504959) and two specimens from Waller and Bauml in 1971 (TAES 554409 & GH s.n.) are the only ones known from 2,700 yucca specimens examined prior to 1997 (Clary 1997) from representative US herbaria (A, ARIZ, BRY, CAS, DS, FLAS, GA, GH, MO, NCU, NLU, NMC, NMCR, NMSU, NY, RM, SBSC, SMU, SRSC, TAES, TEX, UC, UNCC, US, USCH).

SYSTEMATICS AND TAXONOMY: In *Yucca* L., the existing classification is based primarily on fruit type: Section *Chaenocarpa* Engelm., dry fruits, Sect. *Sarcocarpa* Engelm., fleshy fruits and Sect. *Clistocarpa* Engelm., spongy fruits (Flora North America, 2002: 413-440). Phylogenies based on DNA analyses are consistent with the taxonomic circumscription based on fruit type (Clary, 1997; Pellmyr et al., 2007). In addition to fruit type, characteristics of habit, leaf base color, leaf rigidity, scape length, panicle insertion, stigma and style morphology are associated with the sectional divisions.

Morphological characteristics place *Yucca carrii* in sect. *Chaenocarpa*, the dry-fruited yuccas. The plant habit is a basal, trunkless rosette (Fig. 4), the leaf base is cream colored (not red/brown/maroon), the leaves are grass-like and flexible, the inflorescence is tall, exerted 3x or more above the basal rosette (not short and within or barely

exceeding). The tepals are thin, (not succulent) and the pistil is long and narrow (Fig. 5).

SPECIES ORIGIN: The lack of fruit in the species and relatively low pollen viability counts (see below) beg the question of its origin, possibly hybrid. *Yucca carrii* does not share morphological characteristics that would easily signal potential parentage from other species which grow along the Texas coast. These include *Yucca aloifolia* L., *Y. treculeana* Carr. (sect. *Sarcocarpa*), *Y. louisianensis* Trel. (sect. *Chaenocarpa*) and *Y. gloriosa* L. (a hybrid between *Y. aloifolia* and *Y. flaccida* Haw. (sect. *Chaenocarpa*). A rare Texas endemic, *Yucca cernua* Keith (sect. *Chaenocarpa*), grows in the adjacent Piney Woods (Jasper and Newton Co. - approximately 160-240 km north-east of Galveston Bay) but has not been found on the northern Gulf Coastal Prairie.

SEXUAL REPRODUCTION AND YUCCA MOTH ABSENCE: No fruits have been found to date in any population. Dry-fruited yucca capsules tend to persist from year to year on the panicle if fruits are produced. In the Brazoria NWR population, *Y. louisianensis*, also dry-fruited, grows in sympatry with *Y. carrii* but no capsules have been observed in either species. The lack of fruits for both species area-wide suggests that the sole pollinator, the yucca moths (*Tegeticula* and *Parategeticula*, Lepidoptera, Prodoxidae) (Trelease, 1902; Baker, 1986: 556, Pellmyr, 2003: 41) may be extirpated in these areas. To date, no yucca moth records for the north Texas Gulf Coast have been found within entomological databases (Global Biodiversity Information Facility (GBIF)-North American Database (<https://www.gbif.org/species/3256140>); Symbiota Collections of Arthropods Network (SCAN) (<https://scan-bugs.org/portal/collections/list.php>); Mississippi Entomological Museum's Moth photographers Group (<http://mothphotographersgroup.msstate.edu/>).

Review of existing yucca specimen records in SEINet (<https://swbiodiversity.org/seinet>) and iNaturalist.org (www.inaturalist.org) show the nearest location of yucca moth presence, evidenced by fruiting



FIG. 4. Leaf rosette (Clary 432) [TEX-LL]. Chocolate Bayou, Brazoria National Wildlife Refuge. Photo by K.H. Clary.

Y. treculeana (iNaturalist obs. 24312828 and 221194239), near Mad Island WMA, Matagorda Co., the southern end of the range of *Y. carrii*.

Over the past 150 years, most coastal prairie soils in the region have been converted to pasture and to rice and soybean production (Hatch 2020: 31). Pesticide use including aerial spraying, red fire ant predation and ongoing human-induced disturbance of soils in which the yucca moth larvae would undergo diapause (Davis, 1967; Fuller, 1990; Hurlburt, 2001 *in* Environment and Climate Change Canada, 2017) may be likely causes of yucca moth extinction.

POLLEN STAINABILITY ANALYSIS: Pollen stainability counts using aniline blue dye (see Appendix, Pollen Stainability Analysis) were conducted on *Yucca carrii* (unknown fertility), *Yucca louisianensis* (fertile) and *Yucca gloriosa* (fertile) (Trelease, 1902) and of known hybrid origin (Rentsch and Leebens-Mack, 2012). Pollen stainability counts for *Yucca glauca* Nutt. (fertile) were

obtained from Dodd and Linhart (1994: 821).

Although our sample size is very small, results (Appendix, Pollen Stainability Analysis, Table 1) show variation between species. *Yucca carrii* has lower pollen stainability ($\bar{X} = 44.5\%$) than *Y. louisianensis* ($\bar{X} = 99\%$) and *Y. glauca* ($\bar{X} = 94\%$) (Dodd and Linhart 1994: 821) and similar stainability to *Y. gloriosa* ($\bar{X} = 36\%$). It should be noted that although pollen stainability counts are used to gauge pollen viability, the count is not a direct measure of fertility. Species with lower pollen stainability counts can have higher pollen tube germination rates and fertilization success (Rico Reyes-Estanzislao, 2019). A species with relatively low pollen stainability counts, *Y. gloriosa* for example, bears fruit (Trelease 1902) and is not sterile.

POLLINATION STUDIES: *In situ* and *ex situ* field and yucca moth translocation pollination experiments were undertaken during the flowering season (2017-2020) to test fertility of *Y. carrii*. To date, none of the



FIG. 5. Flower (Clary 418 [TEX-LL]). Dickinson Bayou and SH 146, Galveston Co. Photo by J.H. Clary.

pollination experiments has yielded fruit set (see Appendix, Pollination Experiments). *Yucca* moth larvae were translocated from Burnet and Lampasas counties to the BNWR population (2018) and *in situ* hand pollination efforts were made. Plants were translocated from BNWR to an area with live moth populations in Travis County, TX. No fruits have been produced to date.

THREATS AND SPECIES CONSERVATION STATUS: The conversion of coastal prairie from Trinity/Galveston Bay, Houston, Texas City and other outlying coastal towns to suburbs, new highways, oil and gas facilities, farms and ranches is accelerating with human population expansion. These ground disturbing activities threaten the survival of existing and unknown *Y. carrii* populations on unprotected lands. Ongoing coastal erosion, subsidence and increasing sea level rise due to climate change are destroying *Y. carrii* habitat (Fig. 6). To date, we have located six populations total and approximately 200 plants. Populations on protected

lands occur at San Jacinto Monument, BNWR and TNC Texas City Prairie Preserve. However, based on current understanding of the species range, distribution and threats, *Y. carrii* should be considered vulnerable to extinction both within and outside of protected lands.

TAXONOMIC EVALUATION OF *YUCCA TENUISTYLA* TREL.: Early in the taxonomic study of the *Y. carrii*, the question arose as to whether *Y. carrii* was actually *Y. tenuistyla* Trel., because its range, “Southeastern Texas, from about Galveston to Sealy and New Braunfels” (Trelease 1902) overlaps and because the name “*tenuistyla*” refers to a slender style, which *Y. carrii* also possesses. The *Y. tenuistyla* syntype (MO 148761) was examined by the first author and determined to represent a *nomen confusum*, described from a mixed collection of three *yucca* species. The long-styled flowers belong to *Y. rupicola* Scheele, the two large leaves, racemose inflorescence stalk and thick-styled flowers belong to *Y. arkansana* Trel. and the



FIG. 6. *Y. carrii* on eroding bank of Cotton Lake, Chambers County. In foreground, 3 plants, one in flower, showing panicle and rosette, with Jason Singhurst, Texas Parks and Wildlife Department. Photo by K.H. Clary.

small leaves belong to *Yucca* cf. *glauca* Nutt. (Det. K. H. Clary (TEX-LL), 2007). (see Tropicos.org., <https://www.tropicos.org/name/18401450>). Additional specimens (MO 148758, 148765, 148766, 148781, 5421263) identified by Trelease as *Y. tenuistyla* are now classified as *Y. louisianensis* Trel. (see Tropicos.org., <https://www.tropicos.org/name/18401450>).

CONCLUSIONS

Reviewers of an earlier manuscript questioned the uniqueness of *Y. carrii* as a species because no fruits had been found, and, at the time, it was only known from three locations, BNWR (Brazoria Co.), San Leon and Texas City (Galveston Co.). They noted that this could be a clonal population of an extant species or a localized escapee

from cultivation. Our research indicates that it is of uncertain origin but not a localized escapee. *Yucca carrii* is distinct based on morphology, habitat and ecological niche specialization. It is not a synonym of *Y. tenuistyla* Trel. The known range of distribution has expanded considerably. More populations have been located around the original sites and new ones in Brazoria, Chambers, Harris, and Matagorda counties. However, the inability to find fruits or achieve successful results from pollination experiments is confounding. Our experiments have been simple and preliminary to date. Given the highly structured process of the yucca moth life cycle and pollination constraints, substantially more refined research, most importantly, of DNA, is warranted to understand the phylogenetic relationship of *Y. carrii* in *Yucca* L. We have

established that *Y. carrii* is uncommon and that its full geographic range is not known. We encourage researchers to further research on this species, assess its global (G) and state (S) conservation status ranks (<https://explorer.natureserve.org/>) and promote its conservation.

KEY TO THE YUCCA SPECIES FROM THE NORTHERN GULF COAST OF TEXAS

Six species of yucca including *Y. carrii* occur in north coastal Texas. Each species is distinguishable by flower, fruit morphology and habit (Diggs et al. 2006). However, the spring flowering period is relatively short (approximately 6 weeks), not all flowers persist and characteristics of habit can be confusing. A key that distinguishes these species based on persistent characteristics of habit, leaf and panicle is provided below.

1. Mature plants with tall trunks (> 1m) and thick, rigid leaves.
 2. Leaves evergreen and persistent for entire length of trunk, leaf surface waxy, leaf margin denticulate.....*Y. aloifolia* L.
 2. Leaves only persistent for upper part of trunk, leaf surface dull, leaf margin smooth, with no fibers typically.....*Y. treculeana* Carr.
1. Mature plants with short trunk (to 1 m, more or less) or no trunk (acaulescent) and thin, flexible or flaccid leaves.
 3. Mature plants with short trunk (to 1 m, more or less), base of panicle branches occurring on a short scape within or just above crown to (0.5 m) of leaf rosette, leaf margin smooth, panicle branches smooth to sparsely puberulous *Y. gloriosa* L.
 3. Mature plants with little or no trunk, base of panicle branches occurring on a long scape, well above crown (1.2 m or more) of leaf rosette, leaf margin fibrous, smooth, or denticulate, panicle branches sparsely to densely puberulous-tomentose or floccose.
 4. Mature leaves flaccid, green, leaf margin smooth or fibrous, panicle branches sparsely to densely puberulous-tomentose.
 5. Leaf margin white and fibrous, panicle and branches forming a symmetrical, broadly elliptic to obovate candelabrum-shaped inflorescence, with a well-defined apex, panicle branches sparsely puberulous..... *Y. louisianensis* Trel.
 5. Leaf margin brown to reddish brown and smooth, panicle and branches asymmetrical, with open and outwardly spreading inflorescence branches with no well-defined apex, panicle branches puberulous-tomentose..... *Y. carrii* Clary & T. P. Adams
 4. Mature leaves stiff, green with a blue-green cast, leaf margin yellow, denticulate, branch-

lets of panicle drooping, panicle branches moderately to densely floccose.... *Y. cernua* Keith

ACKNOWLEDGMENTS

We thank Bill Carr, Acme Botanical Services, Brandon Crawford, Tiffany Lyon, Tim O'Connell, Aaron Tjelmeland, Texas Nature Conservancy (TNC), for assistance in locating the new populations at the TNC Texas City Prairie Preserve. The staff and volunteers at Brazoria National Wildlife Refuge (BNWR), Chris Best (United States Fish and Wildlife Services (USFWS) and volunteers Walter and Mary Stewart (Bee Cave, TX) supported field research and ongoing pollination studies and Mike Quinn and Valerie Bugh for shared information on yucca moth records in Texas. Jason Singhurst, David Riskind and Andy Sipocz, Texas Parks and Wildlife Department (TPWD) and Eric Keith, Raven Environmental, helped find new populations. We thank James Clary for assistance in making field collections, Linny Heagy for the Figure 1 illustration, Kay McMurry for assistance with the pollen viability study, Beryl Simpson, for use of UT lab facilities, George Yatskievych and Tom Wendt for herbarium support at TEX-LL and the curators at MO, US and TAES for specimen loans. We thank the earlier manuscript reviewers for their insightful and helpful comments.

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APPENDIX. POLLINATION EXPERIMENTS

The following experiments were conducted to determine if the *Y. carrii* population at BNWR could be successfully pollinated and produce fruit. To date, our efforts have not been successful.

HAND POLLINATION: In May 2017, the second author and volunteers at BNWR engaged in hand pollination of blooming yucca plants by mimicking yucca moth pollination on two concurrent nights. Two groups of 10-15 *Y. carrii*, separated by approximately two hundred m were selected as pollen donors and recipients. Pollen grains were collected from open donor flowers and placed at the apex of each recipient's stigma using a dissection needle. A blunt toothpick was used to push the pollen down into the stigma to a depth of approximately 0.25 cm. After each insertion, the needle was wiped with an alcohol swab. Recipient yuccas were identified with plastic ribbon wrapped around their stalks. After one month, there was no observed fruit set in any of the hand-pollinated yuccas. In early June, 2020, three blooming yuccas were hand-pollinated using a donor four-hundred m from the recipients. This effort was not successful.

YUCCA MOTH LARVAE TRANSLOCATION: In July 2018, larvae bearing *Y. pallida* McKelvey fruits collected in Burnet and Lampasas counties, TX, were transported BNWR. *Y. pallida* and *Y. carrii* bloom concurrently. The larvae-bearing fruits (470 fruits collected from 118 individuals, approximately 1,880 moth larvae) were deposited among *Y. carrii* and sympatric *Y. louisianensis* populations at BNWR. This effort was based on reports indicating that successful translocation of extant yucca moth larvae from a yucca population with resident yucca moths to one without may be possible (Lee Lentz, James Henrickson, pers. comm.; Alberta Environment and Sustainable Resource Development, 2013; Environment and Climate Change Canada, 2017). In-ground yucca moth diapause ranges from 1-4 years. After diapause, larvae pupate and emerge from the soil as adults, typically coinciding with flowering by the yucca plant (Davis 1967; Fuller 1990; Hurlburt 2001 in Environment and Climate Change Canada 2017).

Yucca carrii and *Y. louisianensis* were monitored for fruit in late summer 2019 and 2020 but none was produced. The lack of fruit may be due ongoing diapause that has not reached the moth emergence stage.

YUCCA PLANT TRANSLOCATION: In May 2020, two potted BNWR *Y. carrii* with stalks of immature panicles were transported to a property in Travis Co., TX. In this area, yucca moths pollinate *Y. rupicola* Scheele, which blooms concurrently with *Y. carrii*. The intent was to attract yucca moths to *Y. carrii* to determine if fruit set could be achieved through

hybridization. (If fruits were to be produced, they would be destroyed to eliminate possibility of hybridization with *Y. rupicola*). This effort was not successful. Yucca moth pollinations for the 2020 season were extremely low in the area overall and few *Y. rupicola* set fruit.

POLLEN STAINABILITY ANALYSIS: A pollen stainability test was conducted on *Yucca carrii* and two other yucca species to determine the amount of stainable pollen in the species.

MATERIALS AND METHODS

Pollen of *Yucca louisianensis* and *Y. carrii* was collected from herbarium specimens from plants growing in natural populations. The pollen of *Y. gloriosa* was collected from local cultivars. Six anthers from 3-6 open flowers (anthesis) per individual herbarium specimen were collected. Anthers were placed in a 15 ml plastic test tube with ½ ml of 100% glycerin.

The anthers were mashed to release the pollen using the blunt end of a 2 mm diameter bamboo rod. A single droplet of aniline blue (cotton blue) dye (in a 2.5% concentration in 2% acetic mixed in a 1.5 ml of distilled water) was added using a glass pipette (Baba Nitsa et al., 2020; Zonneveld and Van Iren, 2001). This mixture was allowed to sit 24 hr before sampling. Just prior to microscope slide preparation, the mixture was mashed again to release more pollen from the anthers.

Microscope slides were prepared by adding one droplet of 100% glycerin on a microscope slide to which one droplet of the pollen mixture was added then covered with a cover slip. Pollen was examined at a 100X and 400X magnification. The pollen mixture under the slipcover was scanned using overlapping horizontal passes from top left to bottom right. Pollen grains found along each pass were counted. All pollen under the slipcover was counted to the extent possible. Pollen quantities varied. Some samples required two slides per individual specimen to count more than 100 pollen grains. Separate counts were made of pollen grains whose contents were completely stained within (full), partially stained within (3/4, 1/2, ¼ full) or completely empty. In our test, the percentage of fully stained pollen grains is considered the measure of potential pollen viability (Zonneveld and Van Iren, 2001). Pollen counts ranged between 91-558 grains per specimen.

RESULTS (TABLE 1)

Yucca louisianensis (N=1) contained 99% viable pollen, *Y. gloriosa* (N=3) exhibited 26%-41% viable pollen and *Y. carrii* (N=4) 33%-56% viable pollen. These findings indicate that *Y. carrii* pollen is not wholly viable but the mechanism that produces

TABLE 1. Pollen stainability count results for four yucca species.

Genus	Species	Source	No. slides	Pollen grain				Pollen grain empty n=	Total N=	Percent full pollen grains
				full n=	$\frac{3}{4}$ full n=	$\frac{1}{2}$ full n=	$\frac{1}{4}$ full n=			
<i>Yucca</i>	<i>louisianensis</i>	Clary 466	1	236	3	0	0	0	239	99%
<i>Yucca</i>	<i>carrii</i>	Clary 417	2	55	0	4	11	51	121	45%
<i>Yucca</i>	<i>carrii</i>	Clary 432	1	313	0	3	8	234	558	56%
<i>Yucca</i>	<i>carrii</i>	Clary 433	1	40	5	4	0	42	91	44%
<i>Yucca</i>	<i>carrii</i>	Clary 434	1	67	0	22	0	116	205	33%
<i>Yucca</i>	<i>gloriosa</i>	Clary 475	2	42	6	16	0	39	103	41%
<i>Yucca</i>	<i>gloriosa</i>	Clary 476	2	85	0	15	32	75	207	41%
<i>Yucca</i>	<i>gloriosa</i>	Clary 477	2	77	2	16	19	186	300	26%
<i>Yucca</i>	<i>glauca</i>	Dodd and (N=29 indiv.) Linhart 1994	–	–	–	–	–	–	–	94%

Locations:

Clary 417: SH 146 at Dickinson Bayou, Galveston Co.

Clary 432: Chocolate Bayou, Brazoria National Wildlife Refuge, Brazoria Co.

Clary 433: SH 146 at Dickinson Bayou, Galveston Co.

Clary 434: Texas City, Texas Nature Conservancy Attwater Prairie Chicken Preserve, Galveston Co.

Clary 446: CR 264, Sugar Loaf Mountain, Milam Co.

Clary 475: Vega Ave. and Eiger Rd., Austin, Travis Co. Cultivar.

Clary 476: Southwest Pkwy. and Boston Ln., Austin, Travis Co. Cultivar.

Clary 477: US 71/290W and Westgate Blvd., Austin, Travis Co. Cultivar.

Dodd and Linhart: northern Colorado.

reduced numbers of stainable pollen grains is undetermined.

DISCUSSION

Pollen stainability counts may provide comparative indices of pollen fertility potential (Dafni and Firmage, 2000) within a taxonomic group. A fertile outcrossing species can be expected to contain mostly viable pollen containing germplasm which takes up stain when dyed. A viable pollen grain has potential to germinate a pollen tube and fertilize an ovule. Sterile pollen grains have no germplasm and are empty of contents when viewed through the microscope. Species of hybrid origin may show decreasing amounts of stainable pollen from meiotic irregularities related to

incompatible parental chromosomes (Suzuki et al. 1989: 42).

Although low pollen viability counts signal low species fertility, it does not necessarily mean that the pollen (hence species) is sterile. Both pollen viability and pollen germinability (formation of pollen tubes) tests are necessary to determine if pollen is capable of fertilizing an ovule. Sometimes the opposite is observed. Recent studies of a putative hybrid between *Bursera cuneata* and *B. pinnata* (Rico and Reyes-Estanslao, 2019: 7) show that although pollen viability counts are lowest in the putative hybrid, pollen germination (formation of pollen tubes) in the putative hybrid was significantly (statistically) higher than either parent species. In addition, the putative hybrid produces fertile offspring.