A NEW ANCESTRAL DIPLOID SPECIES OF *ECHINOCEREUS* (CACTACEAE) ENDEMIC TO SOUTHWESTERN UTAH AND NORTHWESTERN ARIZONA (U.S.A.)

Blake Wellard

University of Utah Garrett Herbarium
Natural History Museum of Utah
301 Wakara Way
Salt Lake City, Utah 84108, U.S.A.
blakewellard@gmail.com

ABSTRACT

Echinocereus relictus B. Wellard is a newly described diploid (2n = 22) species that is the presumed ancestor of the tetraploid (2n = 44) *E. engelmaniii*. *Echinocereus engelmannii* subsp. *engelmannii* represents both diploid and tetraploid cytotypes. Diploid individuals are endemic to a narrow section of southwestern Utah and northwestern Arizona and have noted similarity with the formerly endangered *E. engelmannii* var. *purpureus*. The distributional range of the diploid and tetraploid plants was mapped using cytogeography. Morphological data collected from ten populations (5 diploid, 5 tetraploid) were correlated with ploidy and analyzed using discriminant function analysis. The cytogeographic and discriminant function analysis results support the distinction of the diploid as new species which is a likely ancestor to the widely distributed tetraploid *Echinocereus engelmannii* subsp. *engelmannii*. *E. relictus* (syn. *E. engelmannii* var. *purpureus*) occurs in a rapidly expanding urban area, and its conservation status needs to be evaluated.

RESUMEN

Echinocereus relictus B. Wellard es una especie nueva diploide (2*n* = 22) que se presume como ancestro de la tetraploide (2*n* = 44) *E. engelmaniii*. *Echinocereus engelmannii* subsp. *engelmannii* presenta ambos citotipos diploide y tetraploide. Los individuos diploides son endémicos de una sección estrecha del suroeste de Utah y noroeste de Arizona y tienen clara similitud con la anteriormente “en peligro” *E. engelmannii* var. *purpureus*. El rango de distribución de las plantas diploides y tetraploides se mapeó usando la citogeografía. Los datos morfológicos colectados de diez poblaciones (5 diploides, 5 tetraploides) se correlacionaron con la ploidía y se analizaron mediante un análisis discriminante. Los resultados de los análisis citogeográficos y discriminantes soportan la distinción del diploide como una nueva especie que probablemente sea el ancestro del tetraploide con distribución amplia *Echinocereus engelmannii* subsp. *engelmannii*. *E. relictus* (syn. *E. engelmannii* var. *purpureus*) ocurre en un área urbana que se expande rápidamente, y su estatus de conservación necesita ser evaluado.t

INTRODUCTION

Echinocereus Engelm. is the third most diverse genus of Cactaceae. Its 44 to 71 species are defined by short, cylindric, single or branching stems possessing few to many ribs with large, laterally-borne, diurnal flowers (Taylor 1985; Blum et al. 1998; Baker 2012). Phylogenetic analysis suggests *Echinocereus* belongs to the tribe Pachycereeae, within the subtribe Echinocereinae (Hunt 2006; Sánchez et al. 2014) and is endemic to western North America where it grows in xeric grasslands, woodlands, and deserts from northern Mexico to South Dakota (Taylor 1985). *Echinocereus* possesses many of the same taxonomic challenges faced by other members of Cactaceae, including geographically isolated populations interspecific hybridization, polyploidy, parallel evolution, and the poor quality or lack of herbarium specimens (Baker & Johnson 2000; Rebman & Pinkava 2001; Reyes-Agüero et al. 2007; De Groot 2011; Majure 2012).

Echinocereus engelmannii subsp. *engelmannii* (Parry & Engelm.) Lem. of section *Erecti* is a xeric adapted [also occurs in higher elevations, including chaparral] species discovered by Charles Christopher Parry, a British-American botanist, around 1849–1850 A.D. and has long been known to be a tetraploid (2*n* = 44) (Stockwell 1935; Pinkava & McLeod 1971; Pinkava et al. 1992; Cota & Wallace 1995). This study follows the treatments by Baker (2012), and Blum et al. (1998) which separate *E. engelmannii* into subsp. *engelmanii* and subsp. fasciculatus. Any future reference here to *E. engelmannii* is referring to subsp. *engelmanni*. The species is distributed throughout the Mojave and Sonoran Deserts, from Baja California and through much of the Great Basin. Numerous infraspecific taxa have previously been recognized in *E. engelmannii*, but perhaps the most

J. Bot. Res. Inst. Texas 10(2): 345 – 359. 2016

notable is *E. engelmanii* var. *purpureus* L.D. Benson, described from north of St. George, Utah. Benson's concept of *E. engelmannii* var. *purpureus* included plants with a narrow stem diameter, 4–10 stems per clump, short central spines, and central spines of dark-purplish red (Benson 1982). *Echinocereus engelmannii* var. *purpureus* was considered to be rare and was subsequently listed as endangered on 11 Oct 1979 but was delisted by the US Fish and Wildlife Service on 27 Nov 1989 due to taxonomic confusion, and the entity was consequently synonymized with *E. engelmannii* var. *chrysocentrus* (Engelm. & J.M. Bigelow) Rümpler.

During ongoing chromosome studies at Arizona State University, a plant identified as *Echinocereus engelmannii* var. *purpureus* from northwestern Arizona was found to be diploid (2*n* = 22) (Pinkava et al. 1998). Additional diploid chromosome counts were conducted in the following years by "Arbeitsgruppe Echinocereus der Deutschen Kakteen-Gesellschaft" with samples from both northwestern Arizona and the St. George, Utah, vicinities, which further supported the presence of diploids in the region (M. Lange pers. comm. 2015). The discovery of diploids is significant because they may represent a distinct ancestral species of the tetraploid *E. engelmannii,* and it also suggests var. *purpureus* may be a part of this new species. These diploids plants are known as "RP 75" in the horticultural trade.

Cytological data is an effective means in Cactaceae to define species boundaries and geographic range, to establish phylogenetic relationships, and for correlating ploidy to morphology (Baker & Pinkava 1987; Parfitt 1987; Baker & Johnson 2000; Baker 2006; Baker & Cloud-Hughes 2014; Stock et al. 2014). Cytological studies, correlated with morphology and geography, have effectively separated diploid from polyploid species (e g , *E. nicholii* (L.D. Benson) B.D. Parfitt (2*n* = 22) from *E. engelmannii* (2*n* = 44)) and polyploid from diploid or other polyploid species (e.g., *E. yavapaiensis* M.A. Baker (2*n* = 66) from *E. arizonicus* Rose ex Orcutt (2*n* = 22) and *E. coccineus* Engelm. (2*n* = 44)) (Baker 2006).

The primary purpose of this study was to investigate whether materials described as *E. engelmanii* var. *purpureus* are part of the variation of a distinct diploid species that may be an extant ancestor to *E. engelmannii*. This question was investigated by (1) mapping the distribution of diploid and tetraploid cytotypes using mitotic chromosome counts; (2) determining whether the type locality for var. *purpureus* was diploid or tetraploid; (3) collecting morphometric data from diploid and tetraploid populations for multivariate analysis; and (4) assessing cytology, morphology and distribution data to determine if the diploid cytotype represented a distinct species.

MATERIALS AND METHODS

Cytological Data

Mitotic chromosome number determinations were made from adventitious root tips formed by field collected stems that were grown in the University of Utah Biology Greenhouse. Root growth was induced by briefly soaking the cut stems in liquid rooting hormone (Dip n' Grow) containing 1.0% indole 3 butyric acid and 0.5% 1-naphthaleneacetic acid. Root tips were collected from early to mid-morning and placed in 2 mM (0.002 M) of 8-hydroxyquinoline (0.029 g/100 ml of distilled water), a mitotic arrest solution, for 5–8 hr at room temperature (~20 °C) before refrigerating in 3:1 95% EtOH and glacial acetic acid (~4–5 °C) for 2–48 hr. Following fixation, roots were transferred to 70% EtOH for at least 2 hr before being hydrolyzed at room temperature for 10 minutes in 1:1 concentrated HCL and 95% EtOH. Roots not used in hydrolysis were kept in 70% EtOH and refrigerated for later use. After hydrolysis, a root tip was dissected in a few drops of 60% glacial acetic acid on a microscope slide to obtain meristematic tissue and a coverslip was placed over the dissected root tip to prevent the sample from desiccating. Prepared slides were gently heated with an alcohol lamp until warm. After heating, the slide was placed on a solid surface, and excess acetic acid was removed by covering the slide with bibulous paper and gently pressing the edges of the coverslip. While keeping the slide stable, roots were then squashed by applying thumb pressure for at least 5 sec. Slides were then placed on dry ice for at least 10 min, and then the coverslip was removed before gently heating again to dry before staining for 12–20 min with 2% Giemsa stain (Gurr's R66). Stained slides were examined using oil immersion at 1000× for cell division. A minimum of 10 cells per sample were analyzed. Images of chromosomes were captured with a digital camera.

Morphometrics

Morphometric data were collected from five diploid and five tetraploid populations during the growing season of March through May 2014. This study defines a population similarly to Baker and Cloud-Hughes (2014): a group of plants of the same species that are most likely to breed amongst each other, rather than exchanging genes with other populations. A map of the morphometric populations is presented in Figure 1.

Morphometric characters were sampled from 33 to 36 individuals (353 total) from each population. Only individuals which possessed a minimum of five stems per clump were selected for analysis. A list and description of morphometric characters used in statistical analysis is presented in Table 1. Stem characters were sampled in the field and five spine clusters were removed from each plant for analysis in the lab. Flower measurements were only sampled from diploid plants for descriptive purposes. Scanned images of the holotype of *E. engelmannii* var. *purpureus* (UTC 174600) were also obtained for additional comparison.

Statistical Analyses

Twenty-three quantitative morphometric characters were selected for comparison, and the data were averaged before importing into SPSS® 12 (IBM, Inc., Armonk, NY) for analysis. Discriminate function analysis (DFA) was used to assess categorical placement of the diploid and tetraploid plants based on morphology, with ploidy being used as the grouping variable (dependent variable) and morphometric measurements used as the predictor variables (independent variables). Wilk's Lambda of the DFA was used to test whether the dependent variable of the discriminant function (ploidy) is important or not in predicting differences between the plants. MANOVA was used to determine any significant differences of mean values of characters between diploid and tetraploid plants.

RESULTS

The cytological and morphometric results support that the diploid cytotype, and the materials formally described as *E. engelmannii* var. *purpureus,* represent a distinct species that is likely an extant ancestor to the widespread tetraploid *E. engelmannii*. The new species will be called *Echinocereus relictus*.

Cytogeography

A total of 63 mitotic chromosome counts were made between October 2013 and January 2015 from stems collected in situ from 47 localities (Table 2). Forty-one of the chromosome counts were diploid ($2n = 22$), including counts from the type locality of *E. engelmannii* var. *purpureus*, and 22 were tetraploid ($2n = 44$). A map of georeferenced chromosome counts within the study area is presented in Figure 1. No chromosome counts were found that deviated from the established Cactaceae base number of $n = 11$.

Field observations and chromosome counts suggest that the populations of *E. relictus* and *E. engelmannii* mostly occupy allopatric geographic areas (Fig. 1). *Echinocereus relictus* occupies about 150,000 ha and is primarily distributed along the Virgin River near the Utah and Arizona borders, north along both sides of the Beaver Dam Mountains, and along the Santa Clara River to near Veyo, Utah. *Echinocereus engelmannii* within the study area is primarily distributed west of the Beaver Dam Mountains, particularly in Beaver Dam Wash and along the Virgin River east of St. George, Utah, to Zion National Park and along the Hurricane Cliffs.

Morphological Analysis

DFA correctly grouped 98.4% (311 out of 316) of the pre-classified taxa, and Wilk's Lambda of the DFA was significant at the $P = 0.001$ level (Table 3). Multivariate analysis (MANOVA) suggested that 19 of the 23 morphological characters are significant at $P = 0.01$ level, and these results are presented along with descriptive statistics (Table 4). One of the tetraploid populations (Woodbury Study Area) was excluded in the statistical analysis after it was discovered to be mixed with diploids (Fig. 1). A photo comparison is presented in Figure 2.

DISCUSSION

This study design follows what has become the standard in Cactaceae and other angiosperm groups, employing comprehensive field studies including ecology, biogeography, genetics, and morphology to make informed

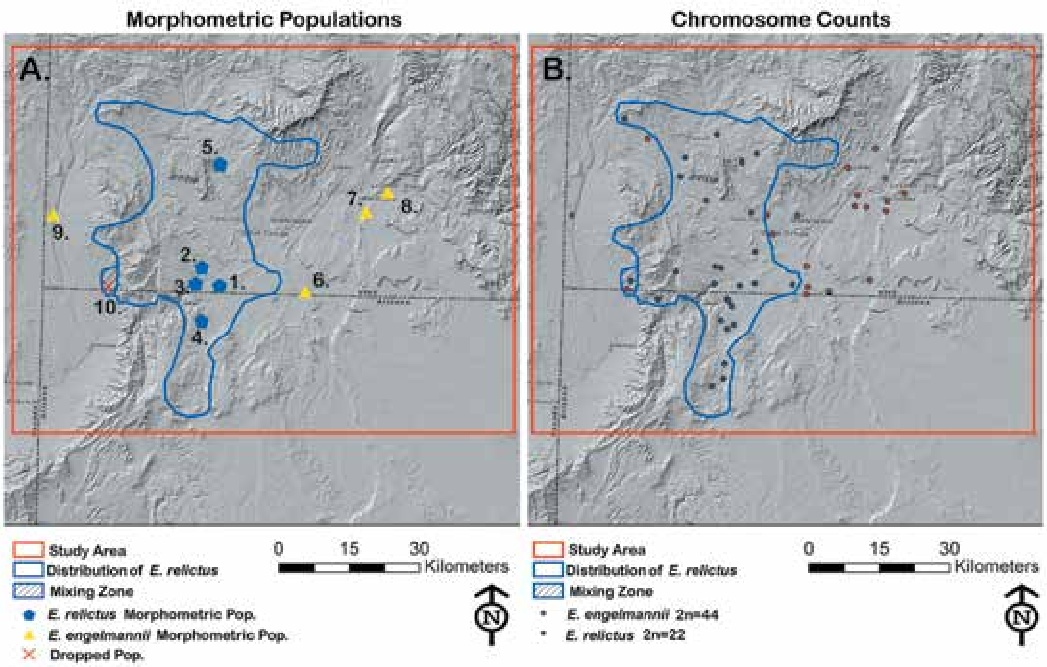

Fig. 1. A. Locations of Morphometric populations within the study area. *Echinocereus relictus* 1–5: (1) South of Sun River; (2) Blooming/Val Springs; (3) Virgin Gorge, Utah; (4) Black Rock Canyon, Arizona; (5) Diamond Valley/Snow Canyon. *Echinocereus engelmannii* 6–9: (6) Fort Pearce/Warner Ridge; (7) Hurricane Cliffs; (8) Virgin River near La Verkin; (9) Beaver Dam Wash, (10) Dropped Population Woodbury Study Area. B. Georeferenced chromosome counts; diploid counts (*E. relictus*) are blue, and tetraploid (*E. engelmannii*) are red.

taxonomic decisions (Baker & Pinkava 1987; Parfitt 1987; Judd et al. 2007; Soltis et al. 2007; Baker 2006; Majure & Ribbens 2012; Baker & Cloud-Hughes 2014; Stock et al. 2014; Laport & Ramsey 2015).The results support that *E. relictus* is a morphologically distinct diploid species occurring only in parts of southwest Utah and northwest Arizona that may represent an ancestor to *E. engelmannii*. The taxonomic recognition of *E. relictus* (syn. *E. engelmannii* var. *purpureus*) is not the first time an entity has been segregated from *E. engelmannii* and elevated to full species status. *Echinocereus nicholii* (L.D. Benson) B.D. Parfitt was formerly considered to be a variety of *E. engelmannii*. However, *E. nicholii* was discovered to be diploid and was elevated to full species status based on ploidy and morphological differences (Parfitt 1987; Pinkava et al. 1992). More recently, Baker (2006) described *Echinocereus yavapaiensis* M.A. Baker ($2n = 66$) and Stock et al. (2014) described *Opuntia diploursina* A.D. Stock, N. Hussey & M.D. Beckstrom ($2n = 22$) based on distinct morphology, ploidy, and geographic distribution. Therefore, the maintenance of *E. relictus* as a diploid cytotype or as a subspecies of the tetraploid *E. engelmannii* is not justified from a systematic or taxonomic standpoint because it obscures the evolutionary relationship of *E. relictus* with its putative descendent, *E. engelmannii,* and its placement among other species within the genus. The high classification percentage of the DFA for *E. relictus* at 98.3% and an overall correct classification of 98.6% further supports the recognition of this new species. *Echinocereus relictus* may for the first time represent a direct ancestor to the widely distributed *E engelmannii* in which, at present, the evolutionary origins are still unresolved (Sánchez et al. 2014).

Biogeography

Packrat midden data collected from areas along the Colorado River and from Vulture Cave in the Grand Canyon suggest that the distribution of *Echinocereus* along with other desert succulents and shrubs has remained largely unchanged for the past 30,000 years (Mead & Phillips 1981; Cole 1990). The low elevation portions of

Table 1. A list and description of morphometric characters used for statistical analysis.

Stems	Description	Abbreviation
1 Number of Stems	Total number of Stems in a clump	NMS
	Characters Measured Five Times For Each Individual	
2 Stem Length	Average Length of the 5 tallest stems	STEML
3 Number of Ribs	Number of longitudinal ribs of stem	NRIBS
4 Stem Diameter Apex	Diameter of stem just below meristem	STEMDIAA
5 Stem Diameter Mid	Diameter of stem at the estimated midlength	STEMDIAM
6 Stem Diameter Base	Diameter of stem just above the base	STEMDIAB
7 Width Between Ribs	Maximum horizontal distance between adjacent ribs	RIBWDTH
8 Length Between Areoles	Average length between three consecutive areoles on the same rib	LNGTHARE
9 Height of Rib to Areole	The height of the rib to the base of the areole	RIBH
	Spines	
10 Number of Central Spines	Total number of central spines per spine cluster	CENSPNUM
11 Length of Abaxial Central Spine	Average length of the lowermost central spine (abaxial spine)	CENSPLNGTH
12 Central Spine Angle	Angle of abaxial central spine with 90^0 equal to zero; angles less than 90 means the spine curved upward, angles greater than 90 means the spine curved downward	CENSPANG
13 Abaxial Central Spine Curvature	Maximum perpendicular departure from a straight line of abaxial central spine	CENSPCUR
14 Abaxial Central Spine Width	Central spine width at the proximal end	CENSPWDTH
15 Abaxial Central Spine Thickness	Central spine thickness at the proximal end	CENSPTHCK
16 Number of Radial Spines	Total number of radial spines	RADSPNUM
17 Average Length of Radial Spines	Average length of 5 radial spines (includes the shortest and longest)	RADSPLNGTH
18 Angle of Radial Spines	Angle of lowermost radial spine with 90^0 equal to zero; angles less than 90 means the spine curved upward, angles greater than 90 means the spine curved downward	RADSPANG
19 Radial Spine Curvature	Maximum perpendicular departure from a straight the line of radial spine with the greatest apparent curvature	RADSPCUR
20 Radial Spine Width	Width of radial spines at the proximal end	RADSPWDTH
21 Radial Spine Thickness	Thickness of radial spines at the proximal end	RADSPTHCK
22 Length of Areole	Areole length of a detached spine.	ARELNGTH
23 Width of Areole	Areole width of a detached spine.	AREWDTH

southwest Utah and northwest Arizona presently act as a refuge for *E. relictus*. The region is further supported in its status as a refugium by the presence of several other endemic cacti (e.g. *Pediocactus sileri* (Engelm. ex J.M. Coult.) L.D. Benson, *P. peeblesianus* (Croizat) L.D. Benson, and *O. diploursina)* as well as other vascular plant species (e,g. Arctomecon humilis Coville, and *Astragalus holmgreniorum* Barneby). Regardless of where the exact refugium of *E. relictus* was historically located, it is clear that the species was able to reach the St. George area before *E. engelmannii* when climatic conditions of the Pleistocene became more favorable for migration. During this period; the Pleistocene Last Glacial Maximum (ca. 21,000 years ago); the St. George area was colder and wetter than today, and was likely a woodland community (VanDevender and Spaulding 1979). Today, *E. relictus* sporadically occurs in woodlands at its upper elevation limits, allowing the inference that it may have survived in suitable habitats during stadial periods of the Pleistocene.

The origin of *E. engelmannii* is still uncertain, but it likely arose during an interstadial period of the Pleistocene when previously isolated populations of *E. relictus* came into contact (autopolyploidy) or when the geographic range of other species of *Echinocereus* overlapped with *E. relictus* and hybridized (allopolyploidy). A similar proposal has been made for species of *Opuntia* by Majure, Judd, Soltis, & Soltis (2012). The large geographic range and morphological variability of *E. engelmannii* suggests that the species may have arisen multiple times with a complex ancestry involving both allopolyploid and autopolyploid events.

Echinocereus engelmannii* var. *purpureus

The United States Fish and Wildlife Service's decision to delist *E. engelmannii* var. *purpureus* in 1989 was

Table 2. List of chromosome count localities. Morphometric populations are in bold. Chromosome voucher specimens are deposited in Garrett Herbarium (UT).

Taxon	Locale	Ploidy/# of Counts	Dec. Lat/Long	Elev. m	Coll. #
E. engelmannii	Utah, Washington Co., La Verkin Overlook Road	2n=44 1 Count	37.19902°, -113.24°	1097	519
E. engelmannii	**Utah, Washington Co., Hurricane Cliffs**	**2n=44** 1 Count	**37.16461°, -113.284°**	**1128**	**520**
E. engelmannii	Utah, Washington Co., Fort Pearce, S of wash	2n=44 1 Count	37.00536°, -113.414°	899	521
E. engelmannii	**Utah, Washington Co., Beaver Dam Wash**	**2n=44** 1 Count	**37.13942°, -114.034°**	**847**	**522**
E. engelmannii	Utah, Washington Co., Toquerville	2n=44 1 Count	37.22682°, -113.287°	1006	523
E. engelmannii	**Utah, Washington Co., La Verkin Overlook Road**	**2n=44** 1 Count	**37.19902°, -113.24°**	**1097**	**524**
E. engelmannii	Utah, Washington Co., N of Sullivan's Knoll	2n=44 1 Count	37.17075°, -113.33°	1036	525
E. engelmannii	Utah, Washington Co., Green Springs Parking Area	2n=44 1 Count	37.15164°, -113.494°	945	527
E. engelmannii	Utah, Washington Co., Sandstone Mountain	2n=44 1 Count	37.19341°, -113.352°	871	528
E. engelmannii	Utah, Washington Co., Mills Road	2n=44 1 Count	37.28383°, -113.311°	1176	529
E. engelmannii	Utah, Washington Co., Bonaza Flat Road (Leeds)	2n=44 1 Count	37.24538°, -113.376°	1128	530
E. engelmannii	Utah, Washington Co., Zion Outlets	2n=44 1 Count	37.11427°, -113.552°	914	531
E. engelmannii	Utah, Washington Co., Old Dump Road	2n=44 1 Count	37.15104°, -113.567°	1036	532
E. engelmannii	**Utah, Washington Co., Fort Pearce Historical Site**	**2n=44** 1 Count	**37.00798°, -113.415°**	**968**	**534**
E. engelmannii	Utah, Washington Co., N of Highway 9	2n=44 1 Count	37.17191°, -113.357°	937	526
E. engelmannii	Utah, Washington Co., Fort Pearce, cliff edge	2n=44 1 Count	37.03116°, -113.317°	1021	533
E. engelmannii	Utah Washington Co., Warner Ridge	2n=44 1 Count	37.05455°, -113.47°	945	535
E. engelmannii	Utah, Washington Co., "Flower Hill"	2n=44 1 Count	37.00072°, -113.469°	945	536
E. engelmannii	Utah, Washington Co., Bee Hive Dome	2n=44 1 Count	37.0151°, -113.466°	905	537
E. relictus	Utah, Washington Co., W of Highway 18	2n=22 1 Count	37.25338°, -113.632°	945	540
E. relictus	Utah, Washington Co., E of Gunlock Reservoir	2n=22 1 Count	37.25619°, -113.768°	1189	541
E. relictus	**Utah, Washington Co., NW of Cinder Cone**	**2n=22** 1 Count	**37.24871°, -113.631°**	**1390**	**542**
E. relictus	Utah, Washington Co., S of Gunlock Reservoir	2n=22 1 Count	37.21952°, -113.777°	1390	543
E. relictus	Utah, Washington Co., Ivins near Kayenta	2n=22 3 counts	37.17551°, -113.714°	937	544
E. relictus	**Utah, Washington Co., Blake's Lambing Ground**	**2n=22** 1 Count	**37.01629°, -113.69°**	**991**	**546**
E. relictus	Utah, Washington Co., Basalt slopes old airport	2n=22 1 Count	37.0791°, -113.593°	814	548
E. relictus	**Arizona, Mohave Co., Black Rock Canyon**	**2n=22** 1 Count	**36.94576°, -113.671°**	**1113**	**545**
E. relictus	**Utah, Washington Co., S of Sun River**	**2n=22 4 counts**	**37.01446°, -113.629°**	**802**	**547**
E. relictus	Utah, Washington Co., S of new airport	2n=22 1 Count	37.01924°, -113.502°	802	549

Table 2. *Continued.*

Taxon	Locale	Ploidy/# of Counts	Dec. Lat/Long	Elev. m	Coll. #
E. relictus	Utah, Washington Co., N of Astragalus Dr.	2n=22 1 Count	37.01928°, -113.592°	853	552
E. relictus	Arizona, Mohave Co., S on Black Rock Road	2n=22* 4 counts	36.97399°, -113.646°	884	553
E. relictus	Arizona, Mohave Co., N on Black Rock Road	2n=22 1 Count	36.98728°, -113.658°	914	554
E. relictus	Arizona, Mohave Co., Summit of Cedar Pockets road	2n=22 1 Count	36.98244°, -113.823°	1067	555
E. relictus	Arizona, Mohave Co., S of mine in Black Rock	2n=22 1 Count	36.93843°, -113.64°	1341	557
E. relictus	Arizona Mohave Co., Junction to Low Mountain	2n=22 1 Count	36.92986°, -113.655°	1067	558
E. relictus	Arizona, Mohave Co., Black Canyon Rim overlook	2n=22 1 Count	36.86303°, -113.659°	1287	559
E. relictus	Arizona, Mohave Co., N of Maple Canyon	2n=22 1 Count	36.83505°, -113.661°	1372	560
E. relictus	Arizona, Mohave Co., Wolf Hole Mountain junction	2n=22 1 Count	36.81925°, -113.688°	1453	561
E. relictus	**Utah, Washington Co., Val Springs area**	**2n=22** 1 Count	**37.04659°, -113.674°**	**881**	**564**
E. relictus	Utah, Washington Co., Near Snow Canyon	2n=22 1 Count	37.15486°, -113.605°	1006	566
E. relictus	Utah, Washington Co., N of Diamond Valley	2n=22 1 Count	37.26816°, -113.596°	1539	567
E. relictus	Utah, Washington Co., Sand Cove	2n=22 1 Count	37.30212°, -113.693°	1539	568
E. relictus	Arizona, Mohave Co., Black Rock	2n=22 2 counts	36.9836°, -113.654°	899	569
E. engelmannii	Utah, Washington Co., Cole Springs Road	2n=44 1 Count	37.29061°, -113.859°	1284	572
E. relictus	Utah, Washington Co., SW of Squaretop Mountain	2n=22 1 Count	37.32754°, -113.915°	1284	573
E. relictus	Utah, Washington Co., Ben's Poppy Study Site	2n=22 1 Count	37.04895°, -113.689°	914	565
E. engelmannii	Utah, Washington Co., Lower Hurricane Cliffs	2n=44 1 Count	37.18202°, -113.28°	1067	575
E. relictus	Utah, Washington Co., Mojave Desert Joshua Tree Road	2n=22 1 Count	37.03686°, -113.781°	1346	574
E. engelmannii	Utah, Washington Co., Woodbury Study Area	2n=44 1 Count	37.0008°, -113.898°	832	576
E. relictus	Utah, Washington Co., Woodbury Study Area	2n=22 1 Count	37.01739°, -113.889°	832	577

Table 3. Discriminant function analysis and Wilk's Lambda results (SPSS 22).

	Predicted Group Membership			
Taxon	*E. relictus*	*E. engelmannii*	Total	
E. relictus	174 (98.3%)	3 (1.7%)	177 (100%)	
E. engelmannii	2 (1.4%)	137 (98.6%)	139 (100%)	
Total	176	140	316	
	98.4% of original grouped cases correctly classified.			
	Wilks' Lambda for the Discriminant Function Analysis Results.			
Test of Function(s)	Wilks' Lambda	Chi-square	df	Sig.
1	0.186	509.102	23	0.000

Table 4. MANOVA and descriptive statistic summaries of the morphological characters used in analysis. Statistically significant (P < 0.01) characters are in bold.

Metric #	Character	Average		Median		Mode		Min		Max		Sdev		MANOVA
		E. relictus	*E. engel*	*E. relictus*	*E. engel.*	*E. relictus*	*E. engel.*	*E. relictus*	*E. engel.*	*E. relictus*	*E. engel.*	*E. relictus*	*E. engel.*	Sig.
1	NMS	9.22	9.43	8	8	8	8	5	5	44	29	4.61	4.48	0.604
2	**STEML**	**18.73**	**21.36**	**19**	**21**	**21**	**20**	**6**	**8**	**33**	**43**	**4.72**	**5.18**	**0**
3	**NRIBS**	**12.50**	**11.26**	**12**	**11**	**12**	**11**	**10**	**9**	**16**	**14**	**1.04**	**0.83**	**0**
4	**STEMDIAA**	**4.10**	**5.06**	**4**	**5**	**4**	**4.8**	**2.1**	**3.5**	**6.5**	**7.5**	**0.62**	**0.69**	**0**
5	**STEMDIAM**	**4.91**	**5.86**	**4.9**	**5.9**	**4.7**	**5.9**	**3**	**4**	**7.6**	**8.1**	**0.69**	**0.68**	**0**
6	**STEMDIAB**	**5.20**	**6.20**	**5.2**	**6.2**	**5.2**	**6.5**	**3.3**	**4.1**	**9.2**	**8.9**	**0.75**	**0.72**	**0**
7	**RIBWDTH**	**10.39**	**14.67**	**10**	**14**	**10**	**13**	**5**	**8**	**19**	**22**	**2.44**	**2.30**	**0**
8	**LNGTHARE**	**10.49**	**14.44**	**10.33**	**14**	**10**	**13**	**4**	**9**	**18**	**22.33**	**2.30**	**2.23**	**0**
9	**RIBH**	**4.78**	**6.72**	**5**	**7**	**5**	**7**	**1**	**3**	**9**	**12**	**0.85**	**1.35**	**0**
10	**CENSPNUM**	**4.92**	**5.29**	**5**	**5**	**5**	**6**	**1**	**3**	**9**	**11**	**1.05**	**1.15**	**0**
11	**CENSPLNGTH**	**3.09**	**3.71**	**3**	**3.6**	**2.9**	**3.5**	**1.2**	**1.5**	**6.1**	**7.2**	**0.72**	**0.80**	**0**
12	CENSPANG	133.69	137.10	130	140	140	140	85	80	185	190	19.64	18.85	0.031
13	CENSPCUR	1.33	1.64	1	1	1	1	0	0	10	10	1.24	1.44	0.008
14	**CENSPWDTH**	**1.02**	**1.20**	**1.02**	**1.19**	**1.02**	**1.18**	**0.41**	**0.62**	**1.82**	**1.91**	**0.20**	**0.20**	**0**
15	**CENSPTHCK**	**0.78**	**0.88**	**0.77**	**0.86**	**0.78**	**0.82**	**0.31**	**0.53**	**1.24**	**1.38**	**0.13**	**0.13**	**0**
16	**RADSPNUM**	**12.82**	**12.43**	**13**	**12**	**13**	**12**	**9**	**4**	**19**	**13**	**1.58**	**5.74**	**0**
17	**RADSPLNGTH**	**8.70**	**11.24**	**8.6**	**11**	**8**	**10.2**	**5**	**6.8**	**14.4**	**18.6**	**1.40**	**1.97**	**0**
18	**RADSPANG**	**101.31**	**94.07**	**100**	**90**	**100**	**90**	**80**	**90**	**160**	**155**	**10.19**	**9.29**	**0**
19	RADSPCUR	0.23	0.23	0	0	0	0	0	0	3	3	0.45	0.45	0.943
20	**RADSPWDTH**	**0.42**	**0.58**	**0.41**	**0.56**	**0.42**	**0.56**	**0.13**	**0.27**	**0.77**	**1.01**	**0.09**	**0.12**	**0**
21	**RADSPTHCK**	**0.36**	**0.46**	**0.35**	**0.44**	**0.35**	**0.42**	**0.15**	**0.24**	**0.67**	**0.9**	**0.08**	**0.10**	**0**
22	**ARELNGTH**	**4.40**	**5.42**	**4.4**	**5.4**	**4.2**	**5**	**2.7**	**3.2**	**6.5**	**7.6**	**0.57**	**0.71**	**0**
23	**AREWDTH**	**3.80**	**4.70**	**3.8**	**4.6**	**4**	**4.5**	**2.2**	**2.8**	**5.5**	**6.7**	**0.52**	**0.68**	**0**

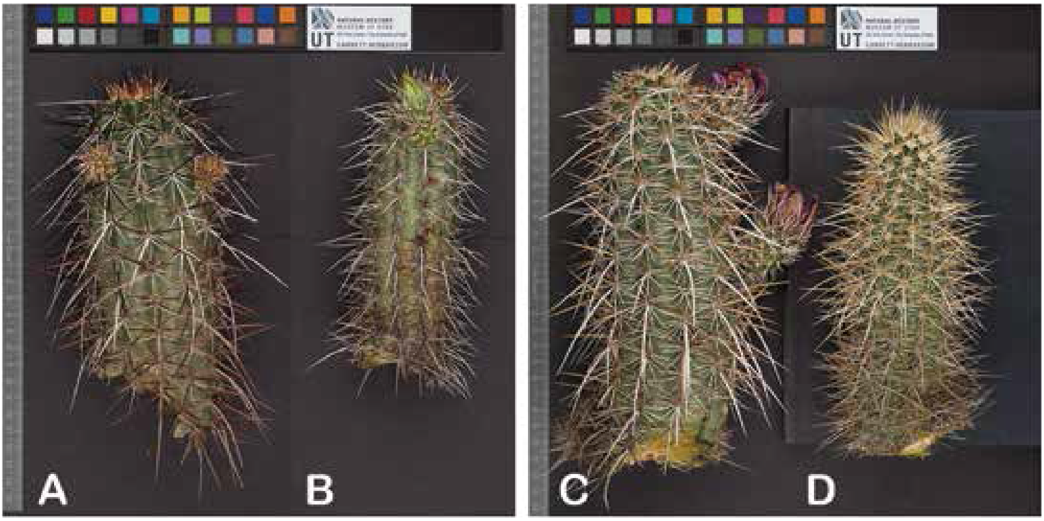

Fig. 2. Morphological comparison of *E. engelmannii* (A & C) vs. *E. relictus* (B & D). A & B. Comparison of *E. relictus* from the type locality (B) and *E. engelmannii* (A) from a nearby population. C & D. Comparison of the mixed population of *E. engelmannii* (C) and *E. relictus* (D) from the Woodbury Study Area. C and D were found 10 m apart.

largely based on a morphological study of *E. engelmannii* in Washington Co., Utah, by Woodbury and England (1988) and comments by other botanists that concluded var. *purpureus* was simply a color phase of *E. engelmannii* var. *chryocentrus* Engelm. ex Rumpler (Miller 1988; Welsh et al. 1993). If *E. engelmannii* var. *purpureus* had been known to be diploid prior to Woodbury and England's study, the researchers could have correlated morphology and ploidy, and the entity would likely have been raised to full species status and not delisted.

The diploid chromosome count from the type locality of *E. engelmannii* var. *purpureus* and the morphology of the type specimen suggests that the variety is best placed as a synonym of *E. relictus*. The variety name "purpureus" would not be valid as a specific epithet because that combination has previously been used and is a synonym of *Echinocereus reichenbachii* (Terscheck) Britton & Rose.

TAXONOMIC TREATMENT

The following key is intended to be used in the field during the growing season and may be incapable of properly identifying herbarium specimens due to the distortion of key characters once materials have been pressed. Data used to separate *E. relictus* from *E. engelmannii* were based on 50,750 field and lab measurements, and all data used in the key were collected using digital calipers and a metric ruler. Key characters were chosen from notable differences observed in the field and from significant characters identified with MANOVA analysis.

FIELD KEY TO *ECHINOCEREUS* OF NORTHWESTERN MOHAVE COUNTY, ARIZONA, AND SOUTHWESTERN UTAH

1. Flowers red, red-orange, pinkish-red, 1.4–6.1 cm wide, tubular, anthers pinkish; hummingbird pollination syndrome; central spines generally terete; plants often forming tightly-clustered mounds.
   2. Spines glabrate; plants often dioecious or gynodioecious; flowers imperfect, pollen-fertile flowers with stamens equal to or surpassing style, pollen-infertile flowers with stamens conspicuously shorter than the style, anthers sterile; tetraploid ($2n = 44$) ____ **E. coccineus** subsp. **coccineus**
   2. Spines mostly papillose-setulose when viewed under 30× magnification; plants synoecious; flowers perfect; diploid ($2n = 22$) ____ **E. mojavensis**
1. Flowers rose-pink to deep magenta, 5–9 cm wide, tepals proximately darker, anthers yellowish; bee pollination syndrome; central spines flattened, angled or sub-terete; plants typically forming semi-open clumps.
   3. Ribs up to 14, averaging 12.5(11–14), width between adjacent ribs and length between areoles along the same rib

usually fewer than 13 mm apart, averaging ca. 10.5 mm (5–15.5 mm), lateral and lowermost radial spines averaging fewer than 9 mm (5.5–11.5 mm); plants restricted to northwestern Mohave Co., Arizona, and west-central Washington Co. Utah; diploid (2*n* = 22) __________ **E. relictus**

3. Ribs up to 13, averaging 11.3(10–13), width between adjacent ribs and length between areoles along the same rib usually greater than 13 mm apart, averaging ca. 14.5 mm (10.2–19 mm), lateral and lowermost radial spines averaging more than 11 mm (8.5–16 mm); widespread distribution; tetraploid (2*n* = 44) __________ **E. engelmannii**

Echinocereus relictus B. Wellard, sp. nov. (**Figs. 3, 4**). TYPE: UTAH: "Limestone Road" S of Sun River Community, 1.25 mi N of Utah/Arizona border, 1 mi W of Port of Entry, 795 m, 37.018198°, -113.632523°, 14 Apr 2016, *B. Wellard 578 with K. Wellard* (HOLOTYPE: UT; ISOTYPE: ASU).

Echinocereus engelmannii var. *purpureus* L.D. Benson

Echinocereus relictus possesses several characters that distinguish the species morphologically from its closest relative *E. engelmannii*. Some of the most notable include: greater number of ribs, shorter spacing between ribs, shorter spacing between areoles, shorter radial spines, shorter abaxial central spine, smaller radial and central spine diameters, shallower rib height to the base of the areole, and narrower stem diameter (Table 5).

Plants unbranched when young, ultimately producing 1–45 branches, forming a compact or somewhat open clump. **Stems** mostly erect or somewhat decumbent, cylindric or tapering apically, (6)10–30(33) cm tall, stem diameter apex (2.1)2.9–5.2(6.5) cm, average 4 cm, stem diameter at midpoint (3)3.4–6.2(7.6) cm, average 4.9 cm, stem diameter base (3.3)3.8–6.6(9.2), average 5.2 cm; ribs (10)11–14(16), average 12.5 ribs, crests slightly undulate, horizontal distance between ribs (5)5.5–15(19) mm, average 10.5 mm; areole (4)5–15.5(18) mm apart along the same rib, average 10.4 mm, height of rib to base of areole (1)3.6–5.8(9) mm, average 4.8 mm, areole diameter (2.7)3.4–5.4(6.5) mm long × (2.2)2.9–4.7(5.5) mm wide, average 4.4 mm long × 3.8 mm wide. **Spines** 16–22 per areole, straight or curved to twisted with polymorphic spine color of whitish to gray, yellow-gold or dull yellow, reddish-brown, or dark blackish-purple spines; spine angle (negative < 90° < positive) radial spines (80°)85°–120°(160°), average 101°, central spines (85°)100°–180°(185°), average 133°; radial spine curvature 0–3 mm, average 0.25 mm, central spine curvature 0–4 (10) mm, average 1.3 mm; radial spines 10–16 per areole, length (5)5.8–11.25(14) mm, average 8.5 mm, proximal radial spine diameter (0.13)0.23–0.58(0.77) mm wide × (0.15)0.2–0.51(.67) mm thick, average 0.42 mm wide × 0.36 mm thick; central spines 3–7 per areole, length (1.2)1.5–4.5(6.1) cm, average 3.1 cm, proximal abaxial spine diameter (0.41)0.63–1.3(1.82) mm wide × (0.31)0.53–1.02(1.24) mm thick, average 1.02 mm wide × 0.78 mm thick; divergent-porrect or rarely recurved, abaxial central spine generally whitish, rarely colored, often flattened or sharply angled, sometimes terete-subterete. **Flowers** 5–9 cm long × 3–9 cm wide, average 7.3 × 5.8 cm; flower tube 10–20 × 10–32 mm; hypanthium 9–20 × 12–32 mm; tepals bright rose-pink to magenta, darker proximally, 30–52 × 5–20 mm, average 40.4 × 11.4 mm, apices rounded, attenuate or somewhat fimbriate or incised; stamen 18–24 mm long, anthers yellow, filaments yellowish-whitish, not motile; ovary 7–28 × 9–20 mm, nectar chamber 3–11 mm wide; style 19–28 × 1.5–4 mm; stigma 9–13 lobes. **Fruits** ovoid-spherical, 20–40 mm long × 20–40 mm wide, red-orangish and often splitting at maturity, pulp white-reddish-white, fruit spines detach at maturity. **Seeds** 1–1.5 mm, black, pitted. **Ploidy** chromosome count 2*n* = 22.

Ploidy.—Diploid 2*n* = 22 based on chromosome counts from 32 individuals from several localities throughout the study area.

Phenology.—Flowers late March to mid-May depending largely on elevation, habitat, temperature, and precipitation. Fruits typically mature about 6–8 weeks later in June and July.

Elevation.—ca. 700–1600 m.

Distribution.—Distributed in northwestern Mohave County, Arizona, and Washington County, Utah (Fig. 1).

Habitat and floristic association.—Rock outcrops and crevices, washes, open plains, hills, valleys, and arid mountain slopes. Warm desert shrub, black brush, and piñon-juniper communities.

Conservation.—Some habitat destruction of *E. relictus* is inevitable and has already occurred due to expanding urban areas in the vicinity of St. George, Utah. Natural and anthropogenic disturbance and habitat destruction are the greatest threats to the species survival. Conservation status needs to be evaluated.

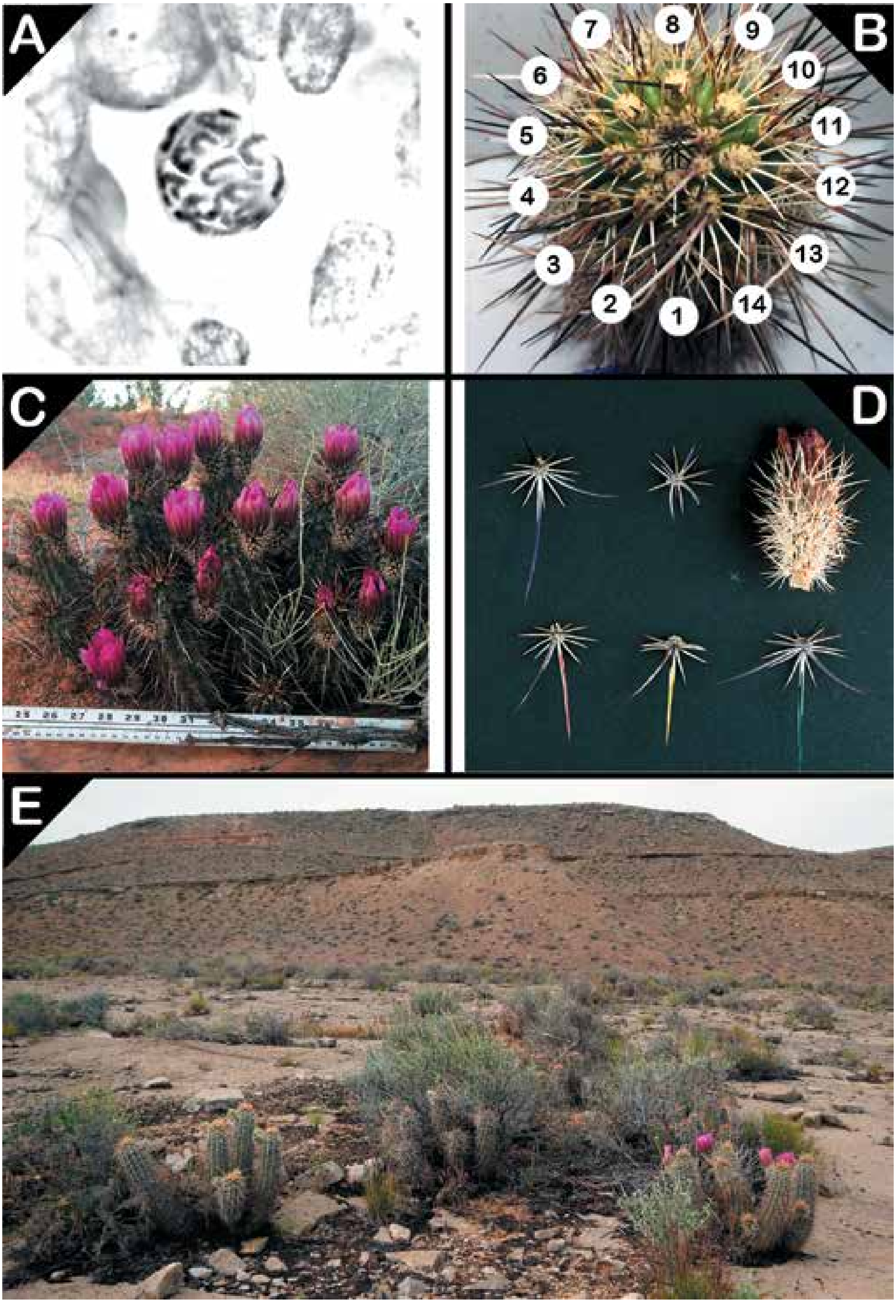

Fig. 3. Morphological features of *E. relictus*. A. Diploid (2n = 22) chromosome count from near the type locality. B. Upper range of rib number for *E. relictus*. C. *E. relictus* that would have formerly fit the concept of *E. engelmannii* var. *purpureus*. D. General spine morphology from the type locality. E. Habitat of *E. relictus* at the type locality.

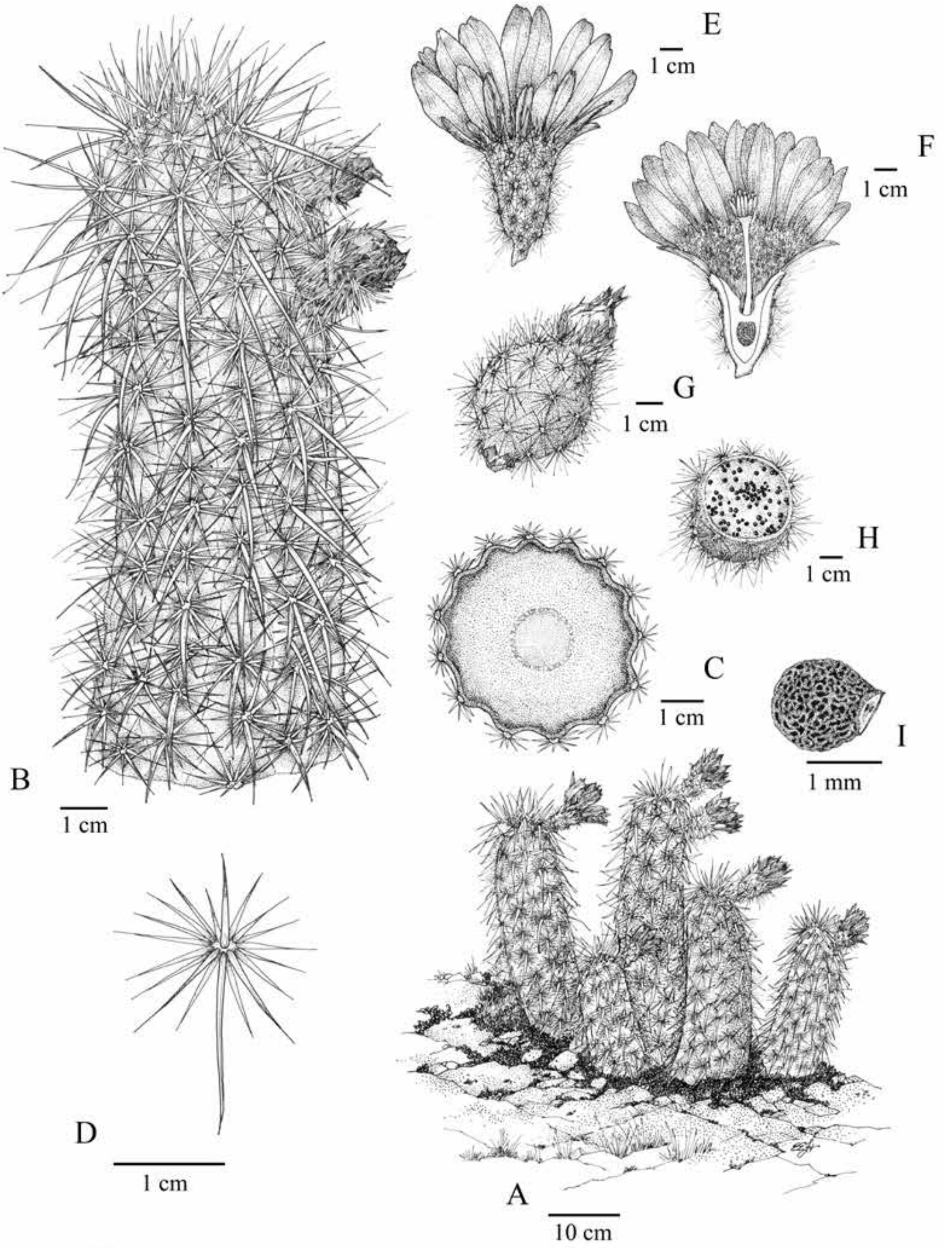

Fig. 4. Illustration of *Echinocereus relictus*. A. Habit. B. Single stem with flowers. C. Stem cross section with 14 ribs. D. Areole and spines. E & F. Flower longitudinal section. G. Fruit. H. Fruit cross section. I. Seed.

Table 5. Comparison of morphometric characters of *E. relictus* and *E. engelmannii*. Diagnostic characters used in the key are bolded.

Character	*E. relictus*	*E. engelmannii*
NMS	1–45	3–60
STEML	10–30 cm	12–30
NRIBS	**11–14**	**10–13**
STEMDIAA	2.9–5.2 cm	4–6.8 cm
STEMDIAM	4–6 cm	4.7–7.5 cm
STEMDIAB	3.8–6.5 cm	5.1–7.8 cm
RIBWDTH	**5.5–15 mm**	**10.1–19 cm**
LNGTHARE	**5–15.5 mm**	**10.2–19 mm**
RIBH	**3.6–5.8 mm**	**4–9.1 mm**
CENSPNUM	3–7	3–7
CENSPLNGTH	1.5–4.5 cm	2.5–5.1 cm
CENSPANG	85°–120°	105°–165°
CENSPCUR	0–4 mm	0–4 mm
CENSPWDTH	0.5–1.4 mm	0.8–1.45 mm
CENSPTHCK	0.53–1.02 mm	0.65–1.10 mm
RADSPNUM	10–16	10–16
RADSPLNGTH	**5.5–11.5 mm**	**8.5–16 mm**
RADSPANG	85°–120°	85°–105°
RADSPCUR	0–3 mm	0–3 mm
RADSPWDTH	0.23–0.58 mm	0.39–0.79 mm
RADSPTHCK	0.2–0.51 mm	0.33–0.62 mm
ARELNGTH	3.4–5.4 mm	4.3–6.6 mm
AREWDTH	2.9–4.7 mm	3.3–6 mm
Ploidy	**2n=22**	**2n=44**

Etymology.—The specific epithet, *relictus*, references how this species survived in a refugium in the northeast Mojave Desert. *Relictus* also references the ancestral diploid state of the species which makes it a plausible ancestor to *E. engelmannii*.

Specimens examined: **U.S.A. ARIZONA. Mohave Co.:** Black Rock Canyon accessed from BLM Road 1009, 2.6 mi NW of the Gypsum Mine in Black Rock Arizona, 36.9458°, -113.671°, 28 Apr 2014, *B. Wellard 545* (UT). **UTAH. Washington Co.:** ½ mi NW of Diamond Valley Cinder Cone, ¼ mi N of Highway 18, 37.2487°, -113.631°, 5 May 2014, *B. Wellard 542 & L. Looby* (UT); Val Springs West of Bloomington, accessed from Navajo Dr, 37.04659°, -113.6740°, 26 Apr 2014, *B. Wellard 564 & L. Looby* (UT); 16 Apr 2015, *B. Wellard 570 & L. Looby* (UT); Blake's Lambing Grounds near the Virgin River, 37.01629°, -113.68996°, 28 Apr 2014, *B. Wellard 546* (UT); S of Sun River community accessed along frontage road just W of Port of Entry, 37.01446° -113.6287°, 6 Nov 2013, *B. Wellard 547 & A. Dean Stock* (UT); "Limestone Road" S of Sun River Community, 1.25 mi N of Utah/Arizona border, 0.5 mi E of Virgin River, 37.019245°, -113.635852°, 16 Apr 2016, *B. Wellard 579 & K. Wellard*.

ACKNOWLEDGMENTS

This project necessitated much work in the field and lab and would not have been possible without volunteers. I would like to thank the following for their generous time and assistance: A. Dean Stock, Patricia Spallone, Lessie Wood, Meghan McCormick, Christopher Hunsaker, Petr Šimon, Brie Beckham, Liz Looby, Claire Gibbons, Angelika Dampf Stone, Chad Roberts, Mindy Wheeler, and my parents Holly and Kevin Wellard. I would also like to thank: Sue Harley for general advisement and assistance with SEM; Mary Barkworth of USU-UTC for a digitized scan of *E. engelmannii* var. *purpureus*; Jason Alexander of UVSC for general advisement and assistance with nomenclatural code; Elizabeth Johnson, collection manager for UT, for herbarium assistance; Marc Baker for advisement on morphometric sampling methodology and assistance with data analysis; Tony Frates of Utah Native Plant Society for general advisement regarding conservation; Ed Bullock for collecting cytological specimens; Rod Chylsta for observing flowering times; Steve Hegji for photographs and field observations; Michael Lange for providing cytological information; and Elaine Hultgren for all the time spent on the beautiful illustration. I would also like to thank Lindsay Walker, Christie Bills, and Erik Rickart of the Natural History Museum of Utah for their assistance. Thank you to A. Dean Stock and Patricia Spallone for continuous support throughout the project and for mentoring me in cytogenetics. I would also like to extend special thanks to my adviser Mitchell Power for taking me on as a student and my committee members Andrea Brunelle and Simon Brewer for giving me the opportunity to explore my research interests. And finally I would like express my appreciation to Michelle Cloud-Hughes and Marc A. Baker for critical reviews.

REFERENCES

Baker, M. 2006. A new florally dimorphic hexaploid, *Echinocereus yavapaiensis* sp. nov.(section *Triglochidiatus*, Cactaceae) from central Arizona. Pl. Syst. Evol. 258(1–2):63–83. doi:10.1007/s00606-005-0390-9

Baker, M.A. 2012. *Echinocereus* (Cactaceae). In: N. Holmgren, P.K. Holmgren, & J.L. Reveal, eds. Intermountain flora: Vascular plants of the Intermountain West, USA. Volume 2, Part A, Subclasses Magnoliidae-Caryophyllidae. New York Botanical Garden, Bronx, New York, U.S.A.

Baker, M.A. & M.A. Cloud-Hughes. 2014. *Cylindropuntia chuckwallensis* (Cactaceae), a new species from Riverside and Imperial Counties, California. Madroño 61(2):231–243. doi:10.3120/0024-9637-61.2.231

Baker, M.A. & R.A. Johnson. 2000. Morphometric analysis of *Escobaria sneedii* var. *sneedii, E. sneedii* var. *leei*, and *E. guadalupensis* (Cactaceae). Syst. Bot. 25(4):577–587. www.jstor.org/stable/2666722

Baker, M.A. & D.J. Pinkava. 1987. A cytological and morphometric analysis of a triploid apomict, *Opuntia*× *kelvinensis* (subgenus *Cylindropuntia*, Cactaceae). Brittonia 39(3):387–401. doi:10.2307/2807141

Blum, W., M. Lange, W. Rischer, & J. Rutow. 1998. *Echinocereus*: Monographie. Geog. 3(4). Turnhout, Proost.

Cole, K.L. 1990. Reconstruction of past desert vegetation along the Colorado River using packrat middens. Palaeogeogr. Palaeoclimatol. Palaeoecol. 76(3):349–366.

Cota, J.H. & R.S. Wallace. 1995. Karyotypic studies in the genus *Echinocereus* (Cactaceae) and their taxonomic significance. Caryologia 48(2):105–122. doi:10.1080/00087114.1995.10797321

de Groot, S.J. 2011. Collecting and processing cacti into herbarium specimens, using ethanol and other methods. Syst. Bot. 36(4):981–989. doi:10.1600/036364411X604985

Hunt, D. 2006. The new cactus lexicon: Descriptions and illustrations of the cactus family. David Hunt Books, Milborne Port, Dorset, England.

Judd, W.S., D.E. Soltis, P.S. Soltis, & G. Ionta. 2007. *Tolmiea diplomenziesii*: A new species from the Pacific Northwest and the diploid sister taxon of the autotetraploid *T. menziesii* (Saxifragaceae). Brittonia 59(3):217–225. doi: 10.1663/0007-196X(2007)59[217:TDANSF]2.0.CO;2

Laport, R.G. & J. Ramsey. 2015. Morphometric analysis of the North American creosote bush (*Larrea tridentata*, Zygophyllaceae) and the microspatial distribution of its chromosome races. Pl. Syst. Evol. 301(6):1581–1599. doi:10.1007/s00606-014-1179-5

Majure, L.C. 2012. The evolution and systematics of the *Opuntia humifusa* complex. Dissertation, University of Florida, Gainesville, Florida, U.S.A.

Majure, L.C., W.S. Judd, P.S. Soltis, & D.E. Soltis. 2012. Cytogeography of the Humifusa clade of *Opuntia* ss Mill. 1754 (Cactaceae, Opuntioideae, Opuntieae): correlations with Pleistocene refugia and morphological traits in a polyploid complex. Comp. Cytogen. 6(1):53.

Majure, L.C. & E. Ribbens. 2012. Chromosome counts of *Opuntia* (Cactaceae), prickly pear cacti, in the Midwestern United States and environmental factors restricting the distribution of *Opuntia fragilis*. Haseltonia 17:58–65. doi:10.2985/1070-0048-17.1.7

Mead, J.I. & A.M. Phillips III. 1981. The late Pleistocene and Holocene fauna and flora of Vulture Cave, Grand Canyon, Arizona. S.W. Naturalist 26:257–288.

Miller, J.M. 1988. Floral pigments and phylogeny in *Echinocereus* (Cactaceae). Syst. Bot. 13:173–183. www.jstor.org/stable/2419093

Parfitt, B. 1987. *Echinocereus nicholii* (L. Benson) Parfitt, stat. nov.(Cactaceae). Phytologia 63(3):157–158.

Pinkava, D.J. & M.G. McLeod. 1971. Chromosome numbers in some cacti of western North America. Brittonia 23(2):171–176. doi:10.2307/2805433

Pinkava, D.J., B.D. Parfitt, M.A. Baker, & R.D. Worthington. 1992. Chromosome numbers in some cacti of western North America-VI, with nomenclatural changes. Madroño 39:98–113.

Pinkava, D.J., J. Rebman, & M. Baker. 1998. Chromosome numbers in some cacti of western North America - VII. Haseltonia 6:32–41.

Rebman, J.P. & D.J. Pinkava. 2001. *Opuntia* cacti of North America: An overview. Florida Entomol. 84:474–483. www.jstor.org/stable/3496374

Reyes-Agüero, J.A., F. Carlín-Castelán, J.R. Aguirre R., & H.M. Hernández. 2007. Preparation of *Opuntia* herbarium specimens. Haseltonia 13:76–82. doi:10.2985/1070-0048(2007)13[76:POOHS]2.0.CO;2

Sánchez, D., S. Arias, & T. Terrazas. 2014. Phylogenetic relationships in *Echinocereus* (Cactaceae, Cactoideae). Syst. Bot. 39(4):1183–1196. doi:10.1600/036364414X683831

Soltis, D.E., P.S. Soltis, D.W. Schemske, J.F. Hancock, J.N. Thompson, B.C. Husband, & W.S. Judd. 2007. Autopolyploidy in angiosperms: Have we grossly underestimated the number of species? Taxon 56(1):13–30. www.jstor.org/stable/25065732

Stock, A.D., N. Hussey, & M.D. Beckstrom. 2014. A new species of *Opuntia* (Cactaceae) from Mojave Co, Arizona. Cact. Succ. J. (Los Angeles) 86(2):79–83. doi:10.2985/015.086.0201

Stockwell, P. 1935. Chromosome numbers of some of the Cactaceae. Bot. Gaz. 96(3):565–570. www.jstor.org/stable/2471509

Taylor, N.P. 1985. The genus *Echinocereus*. A Kew Magazine monograph. Collingridge, London, England, in association with The Royal Botanic Gardens, Kew, Richmond, England.

Welsh, S.L., N.D. Atwood, S. Goodrich, & L.C. Higgins. 1993. A Utah flora. Brigham Young University, Provo, Utah, U.S.A.

Woodbury, L. & L. England. 1988. Morphological variation in *Echinocereus engelmannii* in Washington County, Utah. Unpublished report. U.S. Fish and Wildlife Service, Salt Lake City, Utah, U.S.A.