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Untangling the Systematics of Salt Marsh Dodders: *Cuscuta pacifica*, a New Segregate Species from *Cuscuta salina* (Convolvulaceae)

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Abstract—The salt marsh dodders, *Cuscuta salina,* have been historically delimited as a morphologically variable assemblage of inbreeding forms that parasitize hosts growing in alkaline or saline habitats from western North America. This morphological diversity has been traditionally classified into three varieties: *salina*, *major*, and ***papillata***. A morphometric analysis of floral characters and a molecular study using both plastid and nuclear DNA sequences strongly support the segregation of a new species, ***Cuscuta pacifica*** Costea and M. A. R. Wright, from *C. salina*. The new species corresponds to a lineage that includes varieties *major* and *papillata*, whereas *C. salina* is limited essentially to its type variety. *Cuscuta pacifica* and *C. salina* are sister species that have only a small area of parapatry in lower California, where they are ecologically and reproductively separated. *Cuscuta salina* occurs mostly in inland vernal pools and salt flats of Arizona, California, Nevada, Utah, Baja California, and Sonora, and grows primarily on *Frankenia* and *Suaeda*. *Cuscuta pacifica* can be found in salt marshes from the south-central Pacific coast of California north into British Columbia, parasitic especially on *Salicornia* and *Jaumea carnosa*. *Cuscuta salina* var. ***papillata*** (Yunck.) Costea and M. A. R. Wright, parasitic on hosts that grow in coastal interdunes, falls within the range of variation of *C. pacifica*, where it is transferred.

Keywords—conservation, *Cuscuta salina*, ITS, marsh dodders, morphometric analysis, new species, *rbcL*, taxonomy, *trnL-F*, 26S rDNA.

Cuscuta salina Engelm., the salt marsh dodder, is a morphologically variable assemblage of forms that belongs to the *C. californica* complex, one of the 15 major clades of the subg. *Grammica* (Stefanović et al. 2007). Members of the *C. salina* group range in distribution from Baja California and mainland Mexico north through the western United States into British Columbia, Canada (Yuncker 1932; Costea et al. 2006a; Costea and Stefanović 2009). Salt marsh dodders inhabit alkaline or saline habitats (e.g. coastal marshes and inland salt flats) in which they act as keystone species and ecosystem engineers (Pennings and Callaway 1996; Callaway and Pennings 1998). Following the monographic treatments of the genus by Yuncker (1921, 1932, 1965), *C. salina* was circumscribed to include three varieties: *salina*, *major* Yunck., and *papillata* Yunck. Based on plastid and nuclear sequences from a limited sampling of individuals belonging to these taxa, recent phylogenetic studies have shown that varieties *salina* and *major* each form a distinct group, sister to one another (Stefanović et al. 2007; Costea and Stefanović 2009). This preliminary finding is consistent with the potential segregation of the *C. salina* group into two species, which however requires a detailed examination of numerous additional collections from across their entire distributional range. The objectives of our current study are to: 1) provide further evidence in favor of recognizing two species within the *C. salina* group, and 2) update the taxonomy of this difficult species group. We present here new evidence from a morphometric analysis, ecological data, and plastid (*trnL-F* and *rbcL*) and nuclear (ITS and 26S rDNA) sequences that expand upon our previous molecular phylogenetic studies of *Cuscuta*.

Materials and Methods

Herbarium Specimens—We have searched for relevant specimens in over 100 herbaria in connection with the upcoming treatments of *Cuscuta* for the second edition of the Jepson Manual and Flora of North America Project. Over 700 collections were identified, annotated, and examined for basic morphology. From these collections, a total of 68 specimens of *C. salina*, representing 33 var. *salina*, 33 var. *major*, and two var. *papillata*, were included in the morphometric analysis (Appendix 1). A subset of 11 specimens was used for the molecular phylogenetic analyses. Multiple accessions of both var. *salina* (5 individuals) and var. *major* (6 individuals) were sampled to cover the geographical range, as well as the diverse morphology of this group. We were able to locate only two collections of var. *papillata* (Appendix 1). Both of these were scored for morphometric analysis, but neither could be sampled for molecular studies due to the age and insufficient quantity of herbarium material available. Based on our previous more inclusive analyses (Stefanović et al. 2007; Costea and Stefanović 2009), we selected *C. suksdorfii* Yunck. as an outgroup. Six specimens of this species were included for comparison in the morphological studies; three additional ones were used for molecular analyses (Appendix 1).

Morphology and Morphometric Analysis—Flowers, capsules, and seeds were rehydrated and examined as indicated in Costea and Stefanović (2009). Numerous photographs illustrating details of the floral and fruit morphology for all taxa, including their type collections, are made available on the Digital Atlas of *Cuscuta* website (Costea 2007-onwards). Micromorphology did not yield useful characters in a previous examination of the *C. californica* complex (Costea et al. 2006a), and consequently such characters were not reexamined here.

Four OTUs (operational taxonomic units), corresponding to the three currently accepted varieties of *C. salina* (Yuncker 1965) and *C. suksdorfii*, were included in the morphometric analysis. These are further referred to as 'salina', 'major', 'papillata', and 'suksdorfii' in the analysis. Previous descriptions of the taxa (Engelmann 1859; Yuncker 1921, 1932, 1942, 1965; Beliz 1986; Costea et al. 2006a) were reviewed to produce an initial list of morphological characters. Herbarium specimens were then examined and new potentially useful characters were added to the character list (Appendix 2). A total of 53 continuous, binary, and multistate characters were formulated and scored for all 74 specimens (Appendix 2). Two ordination analyses, principal components analysis (PCA) and canonical variates analysis (CVA), as well as a clustering technique (UPGMA) were performed using NCSS (Hintze 2007). Principal components analysis was used to examine variation independent of OTU assignment (Peirson et al. 2006). Rotation methods were not employed because these reduce the variance accounted for by each orthogonal component axis (Bowley 1999). Important characters used to delimit taxa were excluded during repetitions of PCA to test whether the taxa are phenetically cohesive in the absence of those characters (Peirson et al. 2006). Canonical variate analysis (CVA) was then performed on the data set. As with the PCA, repetitions of the CVA were performed excluding specific delimiting characters to test the phenetic cohesion of the taxa when these characters are left out (Peirson et al. 2006). A reduced list of characters was distilled using the optimization method of Ballard et al. (2001) and Peirson et al. (2006), but results were largely similar to those given by the unoptimized data set and are not shown. UPGMA was conducted on the unoptimized data set using Euclidean distances and scaled by standard deviation. The

conservation status was reassessed using Nature Serve (2008) ranks and criteria.

Molecular Phylogenetic Analyses—To infer the phylogenetic relationships among the members of the *C. salina* group, multiple sequences from two plant genomes were used. We targeted a noncoding *trnL-F* region and the *rbcL* gene from the plastid genome (ptDNA). We also obtained sequences from the internal transcribed spacer (ITS) region of nuclear ribosomal DNA (rDNA) as well as a ~950 bp portion at the 5′ end of the large subunit (26S rDNA). In addition to the DNA samples used in previous studies (Stefanović et al. 2007; Costea and Stefanović 2009), total genomic DNA was isolated from newly obtained specimens as well (Appendix 1). DNA extractions, polymerase chain reaction (PCR) reagents and conditions, amplicon purifications, cloning, and sequencing procedures follow Stefanović et al. (2007) and Costea and Stefanović (2009). The sequences generated in this study have been submitted to GenBank (accession numbers GQ254875–GQ254890). Newly obtained sequences were incorporated into previously aligned matrices from all four regions (Costea and Stefanović 2009), using Se-Al v.2.0a11 (Rambaut 2002), and deposited in TreeBASE (study number S2126). Gaps in the alignments were treated as missing data; however, we coded gaps as binary characters and appended them to the sequence matrix.

Parsimony searches were initially conducted separately for sequences from two genomes, followed by analyses of the combined dataset. In all cases, the searches were done with coded indels. Matrix characters were treated as unordered and all changes were equally weighted. Given the moderate number of OTUs, we performed a Branch-and-Bound search using PAUP* v.4.0b10 (Swofford 2002), ensuring recovery of all of most parsimonious (MP) trees. Support for clades was inferred by nonparametric bootstrapping (Felsenstein 1985), also using the Branch-and-Bound algorithm.

Results

Morphometric Analysis—Results of the PCA are illustrated in Fig. 1. The first principal component (27.79% of the variation) separated 'salina' and 'suksdorfii' from 'major' almost completely, while the second component (13.2% of the variation) separated 'suksdorfii' from all the others. 'Papillata' was weakly separated from 'salina' and 'major' on the third component, which represented only 6.76% of the variation. The first component is largely a reflection of flower and/or perianth size (see Supplemental Appendix 1 for a listing of variable loadings onto the axes). Characters making a large contribution to the first component are the flower width and calyx and corolla lobes widths at the base. The second component mainly reflects aspects of the infrastaminal scales (IFS), anthers, and the gynoecium. For example, characters making a large contribution to the second component are the number of fimbriae on the IFS, the width of IFS including or not the fimbriae, the length of the fimbriate region, of the longest fimbria, of the fimbria on the lower and upper half of the IFS, the anther length and width, the calyx lobe length vs. calyx tube length ratio, corolla tube length, the length of the longer style, and the calyx tube length. Characters making a large contribution to the third component include the presence/absence of calyx papillae, the calyx lobe length, the angle formed by the margins of the calyx lobes at the tips, the pedicel length, the number of seeds in the capsules, and the ratio of IFS length vs. corolla tube length. The CVA (Fig. 2) displayed a strong pattern of group separation. 'Papillata' clustered with 'major' on the first canonical variate, and with 'suksdorfii' on the second canonical variate. The third variate strongly separated 'papillata' from the others. The variable-variate correlations are presented in Supplemental Appendix 2. The canonical variates accounted for 50.9%, 36.7% and 12.4% of the variation, respectively.

The UPGMA cluster analysis also revealed a clear separation of three groups: the first includes *C. suksdorfii*, the second comprises 'salina', and the third grouped together 'major' and 'papillata' (Fig. 3). The cophenetic correlation coefficient of the analysis was 0.66.

Molecular Phylogenies—In preliminary analyses, clades recovered based on data from the nuclear genome were congruent with the tree structure recovered using data from the chloroplast genome (trees not shown). Hence, we combined all data and present only these analyses here. The total-evidence parsimony analysis resulted in 14 MP trees [length = 74; consistency index (CI) = 0.97; retention index (RI) = 0.99], one of which was randomly selected to illustrate the inferred relationships as well as branch lengths (Fig. 4). Consistent with previous findings using limited sampling (Stefanović et al. 2007; Costea and Stefanović 2009), the topology resulting from the combined datasets also revealed

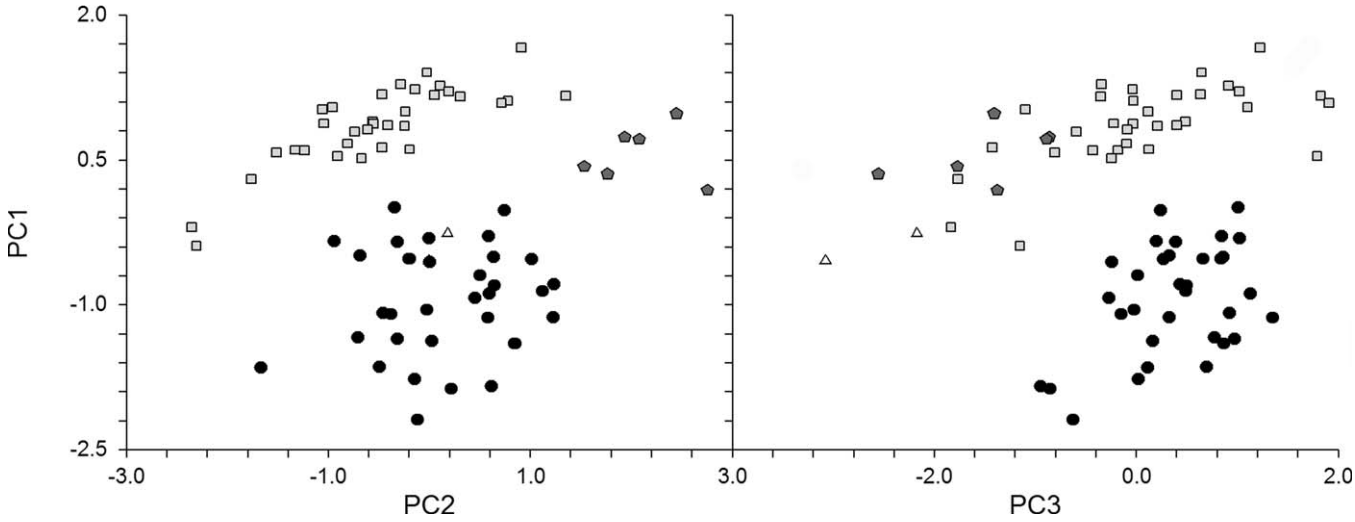

Fig. 1. Principal components analysis (PCA) demonstrates a clear separation of 'salina', 'major', and 'suksdorfii' on the first two component axes (27.79% and 13.2% of the variation, respectively); 'papillata' clusters within 'major' on the first two component axes but is weakly separated from 'suksdorfii' and 'salina' on the third axis (6.76% of the variation). 'Major' – black circles; 'salina' – grey squares, 'papillata' – white triangles, 'suksdorfii' – grey pentagons.

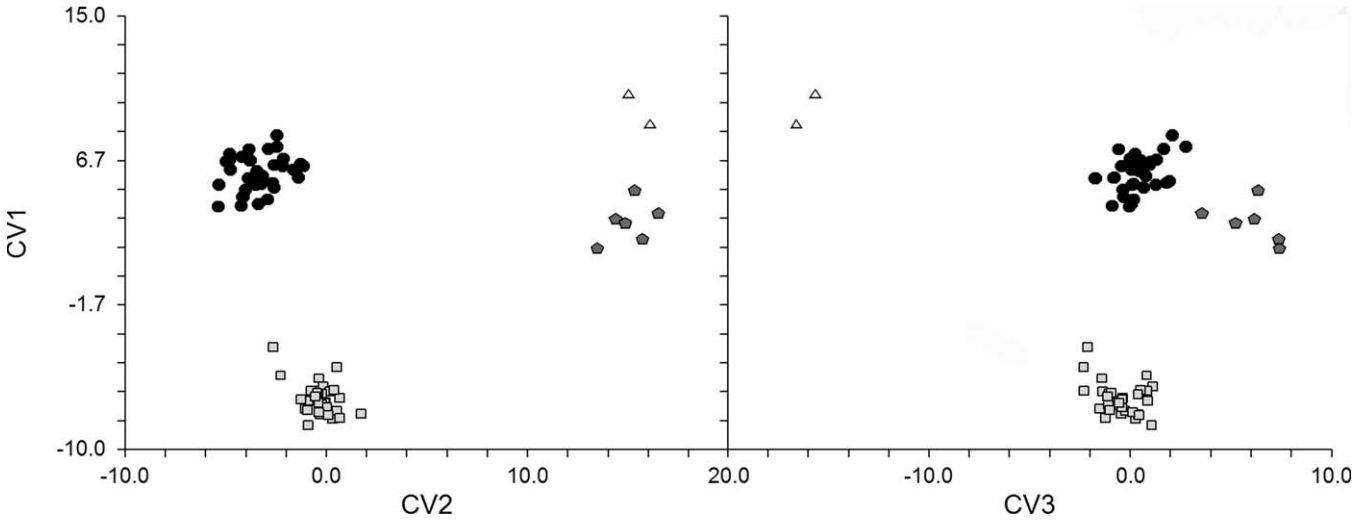

FIG. 2. Canonical variates analysis (CVA) shows a clear separation of all a priori OTU groupings. 'Papillata' clusters with 'major' on the first canonical axis, with 'suksdorfii' on the second canonical axis and is isolated on the third. Canonical variates accounted for 50.9%, 36.7% and 12.4% of the variation, respectively. 'Major' – black circles; 'salina' – grey squares, 'papillata' – white triangles, 'suksdorfii' – grey pentagons.

two major subclades within the *C. salina* group. Based on our current expanded sampling, the two species are reciprocally monophyletic and molecularly distinct from each other, as evidenced by their relative branch lengths and strong bootstrap support (Fig. 4). These molecular results further corroborate our finding based on morphology (e.g. compare with Fig. 3).

Discussion

***Systematics of the Cuscuta salina Group*—**Both morphometric and molecular phylogenetic results confirm the presence of two clearly distinct species within the assemblage of forms that is currently circumscribed as *C. salina*. One corresponds to *C. salina* var. *salina*, and the other to var. *major* plus var. *papillata*. The amount of morphological variation observed in these two lineages is comparable to that found in other species of the *C. californica* complex (Costea et al. 2006a; Costea and Stefanović 2009), and more widely, in subg. *Grammica* (Yuncker 1932; Costea et al. 2006b,c; Costea et al. 2008) as well as subg. *Cuscuta* (Yuncker 1932; García 1998, 1999). Despite the absence of molecular data at present, the morphological similarity of var. *papillata* to var. *major* (Figs. 1–3), along with its sympatric distribution in Mendocino Co., California (where var. *salina* is absent, Fig. 5), indicates that these two taxa are conspecific.

The separation of *C. salina* into two entities, and their recognition at the species level, is further supported by the reproductive biology, geographical distribution, and the ecology of these two lineages. Based on selfing experiments and pollen/ovule ratio analysis, Beliz (1986) found that *C. salina* populations are self-fertilizing, which presumably contributes to the reproductive isolation among them. In addition, varieties *salina* and *major* are essentially allopatric. When they are found parapatrically, in limited geographical areas (Fig. 5), their populations are separated ecologically. Variety *salina* grows on hosts from inland salt flats, alkali flats, and vernal pool habitats in California, Nevada, Utah, and Arizona in the U.S.A., and Baja California, Nayarit, and Sonora in Mexico. Isolated populations of var. *salina* were also found in the interior (but not on the shores) of the Channel Islands in California and other islands off of Baja California. Variety *major*, by contrast, is strictly confined to coastal salt marshes from the south-central coast of California north into British Columbia, Canada. The host range specificity of these taxa is different as well (see below), determined by the distinct plant communities encountered in the ecosystems they inhabit.

***Nomenclature*—**It is evident from the protologue that Engelmann delimited *C. salina* as a mixture of saline dodders "extending to British Columbia (Lyall), and in the interior of Arizona and southern Utah". Essentially, he described *C. salina* by merging and renaming at specific rank (as "*C. salina*, Engelm. n. sp.") two of his earlier varieties (Engelmann 1859), *C. subinclusa* var. *abbreviata* and *C. californica* var. *squamigera*, that he cited in synonymy. The protologue does not clearly mention any specimen, only "C. Wright, Bolander, Kellogg". Therefore, Yuncker selected as a lectotype *Remy s.n.* which was specifically noted in Engelmann's protologue of var. *squamigera* ("J. Remy ! in Hb. Mus. Paris"). According to the International Code for Botanical Nomenclature (ICBN, art 9.17; McNeill et al. 2006), Yuncker's lectotypification of *C. salina* must be followed because: a) the lectotype is in agreement with the protologue, and b) it does not contain parts belonging to more than one taxon. Consequently, the autonymic variety must retain the name *C. salina*, whereas *C. salina* var. *major* requires a specific epithet. "*Cuscuta major*" is not a valid option because it would be a later homonym (even if the earlier homonyms are treated nowadays as synonyms, e.g. *C. major* Koch & Ziz, Cat. Palat. 5. 1813 is *C. epilinum* and *C. major* Gilib., Fl. Lit. Inch. i. 18. 1782 is *C. europaea*). The basionym of *C. subinclusa* var. *abbreviata* is available, but because of the confusion with *C. salina* var. *salina* (see the note under *C. pacifica* var. *pacifica*) and the fact that a new diagnosis would still be required, we prefer to describe the taxon corresponding to var. *major* as a new species, *C. pacifica*. *Cuscuta salina* var. *papillata* is retained as a variety of *C. pacifica*, and a new nomenclatural combination is proposed.

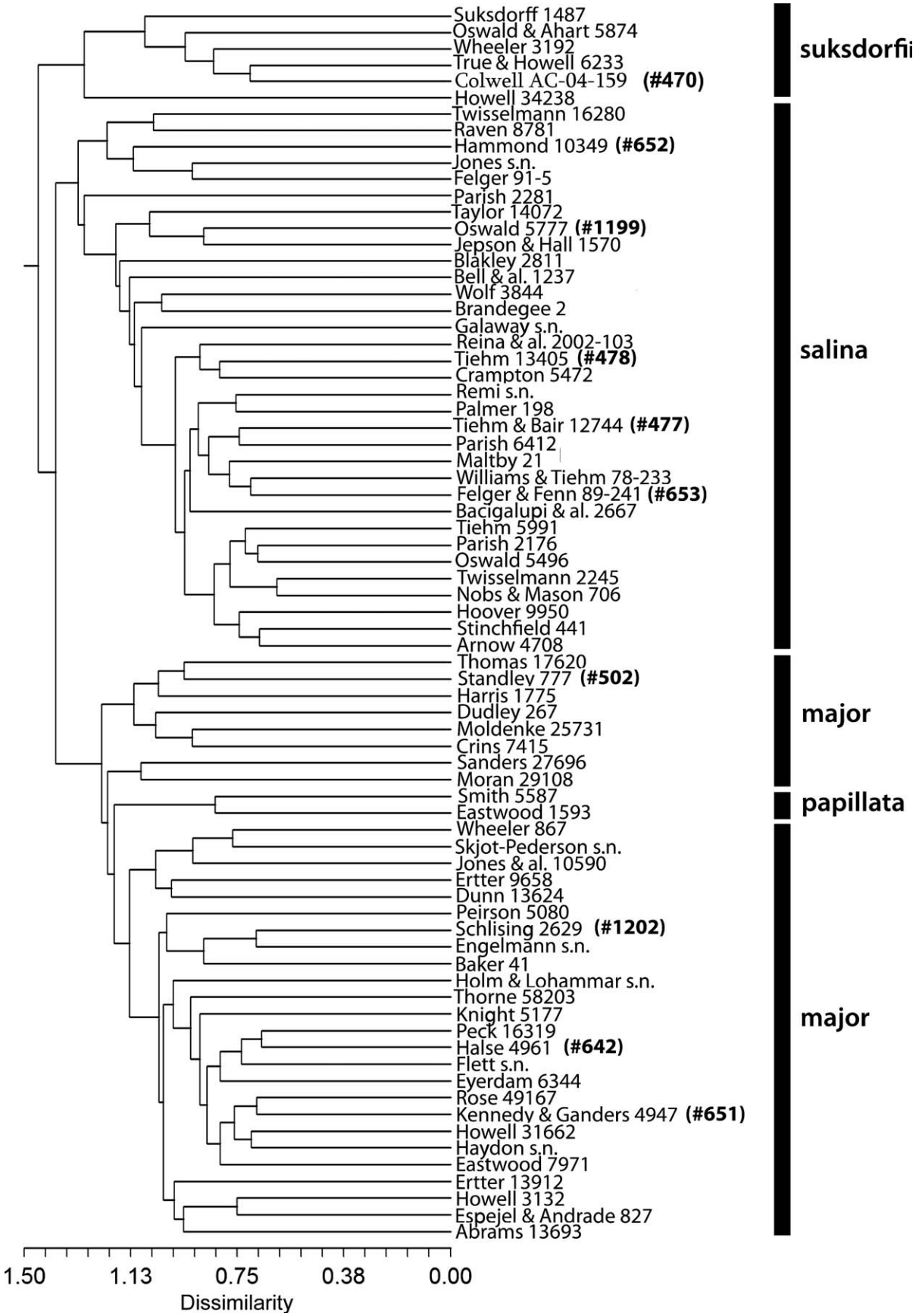

FIG. 3. Phenogram resulting from the UPGMA analysis using Euclidean distances demonstrates clearly delineated clustering of 'suksdorfii', 'salina', and 'major' (with 'papillata' embedded within 'major'). OTU labels correspond to herbarium collections listed in Appendix 1. Numbers in bold refer to DNA extractions used in the molecular study (compare with Fig. 4 and Appendix 1).

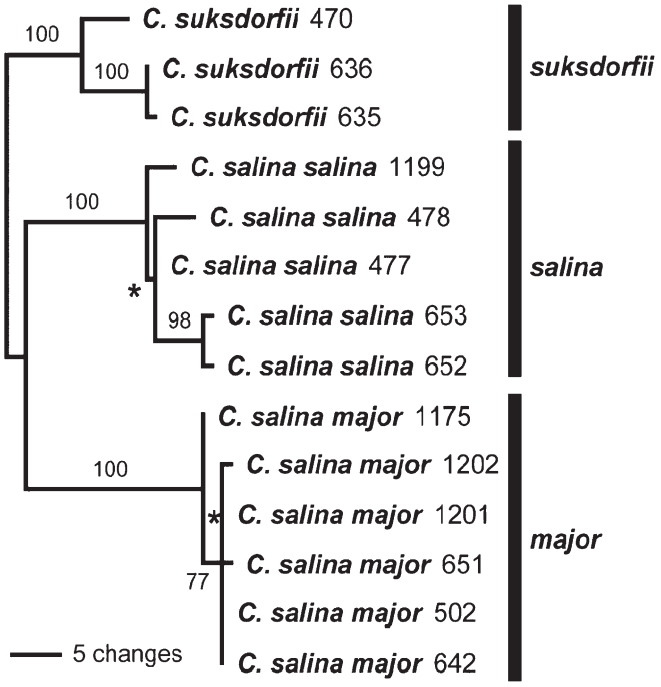

Fig. 4. Phylogenetic relationships among species of the *Cuscuta salina* group derived from maximum parsimony analyses of the combined plastid DNA (*trnL–F* plus *rbcL*) and nuclear DNA (ITS plus 26S) data. Trees are rooted using the closely related *C. suksdorfii*. Numbers following species names correspond to DNA accessions (compare with Fig. 3 and Appendix 1). One of 14 equally parsimonious trees (L = 74; CI = 0.97; RI = 0.99) was chosen to illustrate the amount and distribution of inferred change. Branch lengths are drawn proportionally to the number of changes. Asterisks indicate nodes that collapse in the strict consensus. Bootstrap values are indicated above the branches.

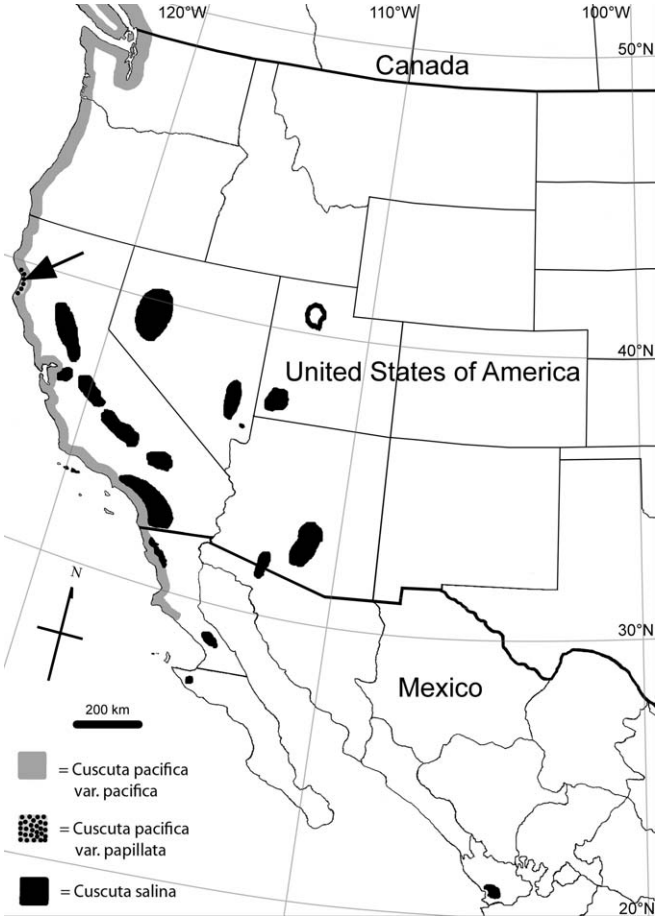

Fig. 5. Geographical distribution of *Cuscuta salina*, *C. pacifica* var. *pacifica* and *C. pacifica* var. *papillata*. Black arrow points toward Mendocino Co, where var. *papillata* is restricted.

Taxonomic Treatment

Key to Salt Marsh Dodders

1. Inflorescences corymbiform cymes; pedicels (0.5–)1–5 mm long; flowers 2.5–4.5 mm long; corolla tube ± cylindric, corolla lobes triangular-lanceolate spreading to reflexed, not or barely overlapping; infrastaminal scales 80–90% of the length of the corolla tube, with 25–45 fimbriae; capsule 1-seeded ..... *C. salina*
1. Inflorescences dense umbellate to subglomerulate cymes; pedicels 0.5–2 mm long; flowers 3.5–6 mm long; corolla tube campanulate, corolla lobes broadly ovate to ovate-rhombic, overlapping; infrastaminal scales 50–70% of the length of the corolla tube, with 10–25 fimbriae; capsule 1–2-seeded 2
   2. Papillae present on pedicels and calyx ..... *C. pacifica* var. *papillata*
   2. Papillae absent on pedicels and calyx ..... *C. pacifica* var. *pacifica*

Cuscuta salina Engelm. in W. H. Brewer, S. Watson, & A. Gray, Bot. Calif. 1: 536. 1876.—TYPE: U.S.A. Utah, Rio Virgen, on *Suaeda*, saline soil, Nov 1885, *Remy s.n.* [lectotype: MO!; isolectotypes: P!, fragment NY!, Yuncker (1932)].

Cuscuta californica Hook. & Arn. var. *squamigera* Engelm., Trans. Acad. Sci. St. Louis 1: 499. 1859. *Cuscuta squamigera* (Engelm.) Piper, Contrib. U. S. Natl. Herb. 11: 455. 1906. *Cuscuta salina* var. *squamigera* (Engelm.) Yunck., Illinois Biol. Monogr. 6, pt. 2–3: 71, Fig. 126. 1921.—TYPE: U.S.A. Utah, Rio Virgen, on *Suaeda*, saline soil, Nov 1855, *J. Remy s.n.* (holotype: P!; isotype: MO!, fragment NY!).

Inflorescences: corymbiform cymes of 2–16 flowers, confluent; pedicels (0.5–)1–5 mm long; bracts 1 at the base of clusters and 0–1 at the base of pedicels, membranous, ovate to lanceolate, 0.7–1.2 × 0.3–0.5 mm, margins entire, apex acute to acuminate. Flowers: 5-merous, 2.5–4.5 mm long, papillae or dome-like cells present on the corolla lobes; laticifers forming long lines, conspicuous in the perianth, ovary and capsule; calyx 1.5–2.5 mm long, glossy yellow when dried, cylindric to narrow campanulate, equaling corolla tube, divided ca. 1/2 to the base, tube 0.6–1.2 mm long, lobes 0.7–1.5 mm long, equal, ovate-lanceolate to lanceolate, not basally overlapping, margins entire, acute to acuminate; corolla: white when fresh, creamy when dried; 2.2–4.0 mm long, the tube 1.2–2 mm long, cylindric-campanulate to obconical, lobes 1.3–2 mm long, ovate-lanceolate to oblong-lanceolate, equaling the corolla tube, initially erect, later patent or reflexed, not overlapping at the base, margins entire or irregular, apex acute to acuminate or cuspidate (sometimes appearing tridentate); stamens: exserted when flowers are completely open, anthers broadly oblong to elliptical, 0.3–0.7 × 0.3–0.4 mm, filaments

0.3–0.7 mm long (for the morphology of pollen see Costea et al. 2006a); infrastaminal scales: 1–1.8 mm long, 80–90% of the corolla tube length, oblong to slightly obovate, bridged at 0.18–0.45 mm, (20–)25–45 fimbriae 0.03–0.20 mm long; ovary: ellipsoid, thickened and risen apically, styles evenly filiform, 0.4–0.9 mm long, shorter than the ovary. Capsules: 1.6–2.5 × 1.7–2.2 mm, thickened around the small interstylar aperture, indehiscent or irregularly dehiscent, surrounded or capped by the withered corolla. Seeds: 1 per capsule, ± visible through the pericarp, 1.35–1.57 × 1.25–1.43 mm, ± dorsoventrally compressed, broadly elliptic to subround, hilum subterminal, subround, 0.11–0.14 × 0.7–0.11 mm, vascular scar linear, 0.02–0.05 mm long, oblique; surface of seed coat epidermis alveolate when dried and papillate when hydrated, cells 30–40 µm in diameter. Figure 6 E–G.

Distribution and Ecology—U.S.A.: Arizona, California, Nevada, New Mexico, Utah, Texas. Mexico: Baja California, Sonora. May also occur in the Channel Islands of Southern California and other islands in Baja California. Grows at 70–800 m elevation on herbaceous hosts (e.g. species of *Frankenia, Salsola, Suaeda, Wislizenia*) from inland salt flats, marshes, and ponds. Flowering from March to November.

Conservation Status—G4 (apparently secure, see Costea et al. 2006a).

Cuscuta pacifica Costea and M. A. R. Wright, sp. nov. —TYPE: U.S.A. California: Humboldt Co., Humboldt Bay near Table Bluff, parasitic on *Salicornia ambigua*, salt marsh, 28 August 1941, *C. C. and S. K. Harris 1175* (holotype: NY!; isotypes: B!, DAO!, GH!, IND!, OSC!, RSA!, UC!, US, WLU! and possibly other herbaria because this collection is part of Plantae Exsiccatae Grayanae).

Cuscutae salinae similis, sed inflorescentiae denso-umbellatae ad subglomerulatas; pedicelli 0.5–2 mm longi; flores 3.5–6 mm longi; calyx 1.8–3.3 mm longus, campanulatus ad cupulatum; corolla 2.8–5.4 mm longa, tubo campanulato, lobis late ovatis ad rhombice ovatos, erectis ad effusos, manifeste imbricatis basi; scalae porcae oblongae ad parvum obovatas, 10–25 fimbriis 0.05–0.17 mm longis; semina 1–2 per capsulam.

Inflorescences: dense umbellate to subglomerulate cymes of 2–17 flowers, confluent; pedicels 0.5–2 mm long; bracts 1 at the base of clusters and 0–1 at the base of pedicels, membranous, ovate to lanceolate, 0.7–1.8 × 0.4–0.9 mm, margins entire, apex acute to acuminate. Flowers: 5-merous, 3.5–6 mm long; papillae or dome-like cells present on the corolla lobes and sometimes on the calyx and pedicels; laticifers forming long lines, conspicuous in the perianth, ovary and capsule; calyx 1.8–3.3 mm long, dull brown when dried (rarely yellow), campanulate to cupulate, equaling corolla tube, divided ca. 2/3 to the base, tube 0.6–1.6 mm long, lobes 1.3–2.2 mm long, ± equal, ovate-triangular, slightly overlapping basally, margins entire, acute to acuminate; corolla: white when fresh, generally dark brown when dried (rarely creamy), 2.8–5.4 mm long, the tube 1.5–2.6 mm long, campanulate, lobes 1.7–2.6 mm long, broadly-ovate to rhombic-ovate, equaling the corolla tube, erect to spreading, overlapping at the base, margins entire or irregular, apex acute to cuspidate (sometimes appearing tridentate); stamens: included when flowers are completely open, anthers broadly elliptical to subround, 0.35–0.5(–0.6) × 0.2–0.4 mm, filaments 0.3–0.6 mm long (pollen as in *C. salina*); infrastaminal scales: 0.8–1.6 mm long, 50–70% of the corolla tube length, consisting of oblong to slightly obovate ridges with 10–25 fimbriae, 0.03–0.17 mm long, bridged at 0.25–0.50 mm; ovary and styles: as in *C. salina*. Capsules: 2–3.6 × 1.4–2.1 mm, thickened around the small interstylar aperture, indehiscent or irregularly dehiscent, surrounded by the withered corolla. Seeds: 1–2 per capsule, not visible through the persistent corolla and pericarp, 1.45–1.95 × 1.25–1.43 mm, ± dorsoventrally compressed, broadly elliptic to subround, hilum subterminal, subround, 0.11–0.14 × 0.7–0.11 mm, vascular scar linear, 0.02–0.05 mm long, oblique; surface of seed coat epidermis alveolate when dried and papillate when hydrated, cells 30–40 µm in diameter. n = 14 (Beliz 1986); $2n$ = ca. 30 (Pazy and Plitmann 1995). Figure 6 A–D.

Etymology—The specific epithet references to the Pacific coastal habitat and geographical distribution of this species.

Cuscuta pacifica var. pacifica

Cuscuta salina var. *major* Yunck., Illinois Biol. Monogr. 6: 161. 1921.—TYPE: U.S.A. California: Santa Clara Co., Palo Alto, frequent on *Salicornia* in the marshes, 14 Sep 1901, *Baker 41* (holotype: NY!; isotypes: CAS!, GH!).

Cuscuta subinclusa var. *abbreviata* Engelm., Trans. Acad. Sci. St. Louis 1: 500. 1859.—TYPE: U.S.A. California [Solano Co.]: Mare Island in San Francisco Bay, on *Arthrocnemum*, *Wright s.n.* (holotype: MO!, type 2757815; isotype GH!, 00267828).

Note—The collection currently registered at MO as "holotype 2757814", barcode "MO-694322", on *Grindelia* cannot be a type of *C. subinclusa* var. *abbreviata* because it was cited by Engelmann under "typical" *C. subinclusa*. A note handwritten by Engelmann on this specimen mentions that "among the loose flowers may be a few from *Arthrocnemum Cuscuta*, from the same locality." A similar note can be found on the specimen that has *Arthrocnemum* as a host: "among the loose flowers may be a few of *C. subinclusa* on *Grindelia* from the same locality." Wright had apparently sent Engelmann a mixture of the two dodders from the same locality, *C. subinclusa* ("typical") and *C. subinclusa* var. *abbreviata*, but on different hosts (*Grindelia* and *Arthrocnemum*, respectively). Most likely Engelmann separated them into two envelopes/specimens, warning that, however, a few flowers may still be mixed among them. Subsequent authors (Yuncker 1921, 1932; Costea et al. 2006a) considered the *Wright s.n.* specimen on *Arthrocnemum* to be *C. salina* var. *salina*. A reexamination of this collection revealed that it is *C. pacifica* (var. *pacifica*).

Distribution and Ecology—Canada: British Columbia. Mexico: Baja California. U.S.A.: California, Oregon, Washington (Fig. 5). Grows on hosts from coastal salt marshes and tidal flats (sea level), especially on *Jaumea carnosa* and *Salicornia virginica*. Flowering between June and October.

Conservation Status—G4 (apparently secure; Costea et al. 2006a).

Cuscuta pacifica var. **papillata** (Yunck.) Costea and M. A. R. Wright, comb. nov. *Cuscuta salina* var. *papillata* Yunck., Bull. Torrey Bot. Club 69: 543. 1942.—TYPE: U.S.A., California, Mendocino Co.: Fort Bragg, 8–16 Aug 1912, *Eastwood 1593* (holotype: GH!; holotype fragment NY!).

Variety *papillata* is characterized by papillae on the calyx and pedicels. The varietal rank is preserved for this form because similarly papillate plants are currently accepted in *C. californica* complex (e.g., *C. californica* var. *papillata*). Papillae,

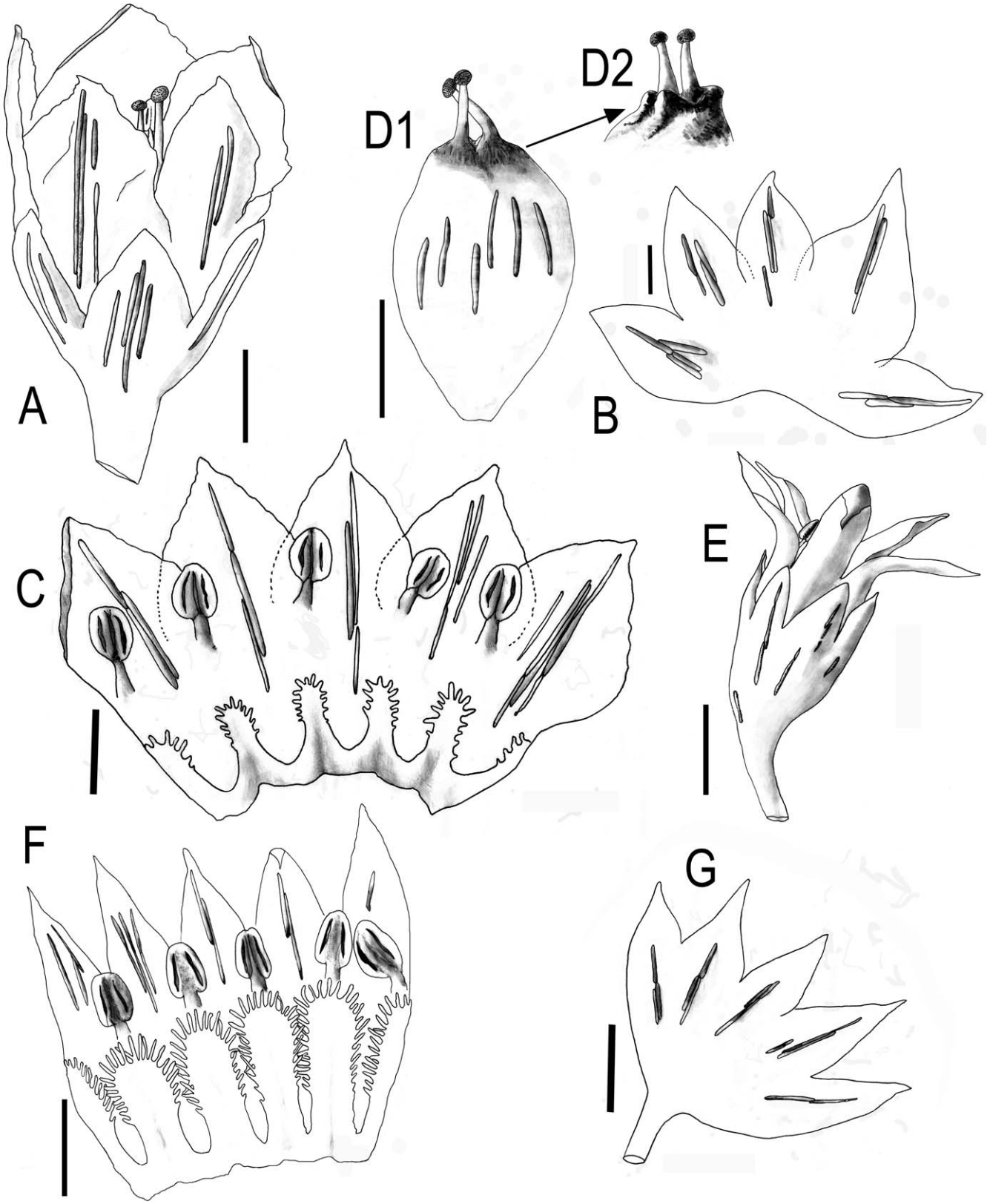

FIG. 6. *Cuscuta pacifica* Costea and M. A. R. Wright (*Harris & Harris 1175*), A. Flower. B. Dissected calyx, dorsal side. C. Dissected corolla (opened, ventral side). D1–D2. Gynoecium. *Cuscuta salina* (*Bacigalupi & al. 2667*). E. Flower. F. Dissected corolla. G. Dissected calyx. Scale bars = 1 mm.

dome-shaped cells or an intergradation between these may occur on the corolla lobes of both *C. salina* and *C. pacifica*, but they apparently are not taxonomically significant. Such plants with obvious papillae on the corolla lobes (but not on the calyx and pedicels) have been included in *C. salina* var. *papillata* in the past (e.g., Costea et al. 2006a).

Distribution and Ecology—U.S.A.: endemic to California, Mendocino Co. Grows close to sea level on hosts such as

Lupinus variicolor that can be found in interdune depressions on the coastal plateau (Smith and Wheeler 1990–1991). It flowers from July to October.

***Conservation Status*—**Our extensive search at the end of July 2008 at the type collection site failed to recover this taxon although the suitable hosts were present. Given the apparent rarity of var. *papillata*, the conservation status T2 ('Imperiled') is proposed.

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Appendix 1. List of herbarium specimens examined for morphometric and molecular analyses of the *Cuscuta salina* group. Country, locality details, date, collectors, and herbaria in which the specimens are deposited are provided for all spacimens. In addition, for material used in molecular analyses, DNA extraction and GenBank accession numbers (*trnL-F*; *rbcL*; ITS; 26S rDNA) are given in the square parentheses. Abbreviations of herbaria follow Index Herbariorum.

***Cuscuta pacifica* var. *pacifica*:** CANADA. British Columbia: Boundary Bay, 16 Aug 1987, *Crins 7415* (DAO); Greater Vancouver Regional District, 5 Aug 1959, *Holm and Lohammar s.n.* (UCR); Surrey, Crescent Beach, 49°03′43″N, 122°52′38″W, 3 Aug 1997, *Kennedy and Ganders 4947* (UBC) [#651; EF194500, EU883463, EF194711, EU883513]. U.S.A. California: Alameda Co., Oakland Beach, San Francisco Bay, Jul 1880, *Engelmann s.n.* (CAS); N of Toll Plaza, San Francisco Bay, W Oakland, 23 Sep 1994, *Ertter 13912* (UC); Contra Costa Co., Pt. Pinole Regional Shoreline, 30 Sep 1990, *Ertter 9658* (UC); Humboldt Co., salt marsh on Humboldt Bay near Table Bluff, 28 Aug 1941, *Harris & Harris 1175* (type of *C. pacifica*, B); Los Angeles Co., just W of Malibu, 8 Sep 1948, *Nobs & Smith 648* (UC); Marin Co., Almonte marsh, 6 Sep 1918, *Eastwood 7971* (CAS); Monterey Co., near Monterey, Jul 1893, *Dudley 267* (CAS); Orange Co., 2.5 mi. from Balboa, 29 Aug 1924, *Peirson 5080* (CAS); Newport Bay, 25 Jun 1932, *Wheeler 867* (CAS); San Diego Co., Agua Hedionda Ecological Reserve, 33°08′36″N, 117°18′47″W, 30 Apr 2004, *Sanders 27696* (UCR); San Francisco Co., San Francisco, 1 Jul 1956, *Howell 31662* (CAS); San Mateo Co., South San Francisco, 24 Aug 1949, *Rose 49167* (NY); Atherton, 23 Oct 1927, *Skjot-Pedersen s.n.* (AAU); Santa Barbara Co., Goleta Beach, 18 Aug 1960, *Dunn 13624* (CAS); Santa Barbara Co., Santa Cruz Island, 14 Jun 2006, *Colwell s.n.* (TRTE) [#1175; GQ254879; GQ254883; GQ254887; GQ254875]; Santa Clara Co., Palo Alto, 14 Sep 1901, *Baker 41* (type of *C. salina* var. *major*, MO); Palo Alto, 18 Jun 1972, *Moldenke 25731* (NY); Palo Alto Yacht Harbour, 6 Oct 1974, *Thomas 17619* (CAS); Sonoma Co., Lower Tubbs Island, 23 Sep 1985, *Knight 5177* (CAS); Bodega Head, 27 Jul 1963, *Schlising & Keaton 2629* (CHSC) [#1202; GQ254881; GQ254885; GQ254889; GQ254877]; Ventura Co., Coast Highway near Pt. Mugu, 26 Jun 1935, *Abrams 13693* (CAS); Pt. Mugu, 8 Oct 1927, *Howell 3132* (JEPS); Solano Co., Grizzly Island Rd. 5.5 mi south of Hwy 12 near Suisun City, 19 Oct 1998, *Oswald 9666* (CHSC) [#1201; GQ254880; GQ254884; GQ254888; GQ254876]. Oregon: [no locality and date] *Haydon s.n.* (CAS); Lincoln Co., Waldport, 30 Jul 1995, *Halse 4961* (NY) [#642; EF194498, EU883462, EF194709, EU883512]; Tillamook, 10 Aug 1930, *Peck 116319* (DS); Washington: Gray's Harbor Co., ocean shores at S end of peninsula, 6 Aug 1982, *Standley 777* (NY) [#502; EF194499, EU883461, EF194710, EU883511]; Jefferson Co., 12 Aug 1944, *Eyerdam 6344* (MO); near Port Townsend, 10 Oct 1937, *Jones & al. 10590* (CAS); Pierce Co., Tacoma, [no date], *Flett 249* (UC). MEXICO. Baja California: Cabo Punta Banda, 10 May 1990, *Espejel & Andrade 827* (MEXU); Cabo Punta Banda, 31°44.5′N 116°47.5′W, 3 Aug 1980, *Moran 29108* (ENCB); N of Punta Banda, ca. 1 mi. N of La Jolla, 25 Apr 1984, *Thorne 58203* (RSA). ***Cuscuta pacifica var. papillata*:**

U.S.A. California: Mendocino Co., Fort Bragg, 8–16 Aug 1912, *Eastwood 1593* (the type, GH); 8 km north of Gualala, 13 Jun 1979, *Smith 5587* (CAS). ***Cuscuta salina*:** U.S.A., Arizona: Southern Arizona, 1867, *Palmer 198* (MO); Pima Co., Organ Pipe Cactus National Monument, 19 Jul 1989, *Felger & Fenn 89-241* (NY) [#653; EF194496, EU883467, EF194708, EU883517]; Pinal Co., S end of Picacho Reservoir, 9 Apr 1996, *Hammond 10349* (NY) [#652; EF194495, EU883466, EF194707, EU883516]; California: Alameda Co., N of Livermore, 31 Aug 1966, *Hoover 9950* (RSA); Colusa Co., Williams Rd. bridge over 'the Trough', 11 Jul 1916, *Stinchfield 441* (DS); Sacramento National Wildlife Refuge, 31 Aug 1993, *Taylor 14072* (JEPS); Fresno Co., 1 mi. W of Kerman Junction, on California Hwy. 180, 29 Jul 1941, *Bacigalupi & al. 2667* (DS); Mendota Pool, 2 Oct 1948, *Nobs & Mason 706* (UC); 3.2 mi. W of Kerman on road to Mendota, 31 Aug 1955, *Raven 8781* (CAS); Glenn Co., Sacramento National Wildlife Refuge, 9 Jun 1993, *Oswald 5496* (CHSC); 5 Aug 1993, *Oswald 5777* (CHSC) [#1199; GQ254882; GQ254886; GQ254890; GQ254878]; Kern Co., East side of county, on DiGiorgio Rd., 0.6 mi E of Cottonwood Rd., 16 Oct 1969, *Twisselmann 16280* (OSC); Riverside Co., near Elsinore – Temescal Wash, 31 May 1901, *Jepson & Hall 1570* (JEPS); Lake Elsinore, on *Frankenia,* 3 Nov 1891, *Parish 2281* (CAS); San Bernardino Co., San Bernardino Valley, 26 May 1891, *Parish 2174* (CAS); Solano Co., 2 mi N of Dozier Station, 8 Dec 1959, *Crampton 5472* (CAS); Ventura Co., E Anacapa Island, 26 Apr 1959, *Blakley 2811* (JEPS); Nevada: Churchill Co., Lahontan Valley, 8 Sep 1998, *Tiehm & Bair 12744* (NY) [#477; EF194492, EU883464, EF194704, EU883514]; Near spring SW of Sand Mtn, 24 Jul 1978, *Williams & Tiehm 78-233* (RSA); Clark Co., Muddy Mountains, 24 Oct 1979, *Bell & al. 1237* (RSA); Lincoln Co., Caliente, 27 Aug 1912, *Jones s.n.* (RSA); Pershing Co., Lower Lovelock Valley, 31 Aug 2000, *Tiehm 13405* (NY) [#478; EF194493, EU883465, EF194705, EU883515]; Utah: Salt Lake Co., W of Salt Lake City, *Arnow 4708* (NY); Washington Co., St. George, 12 Oct 1935, *Galway s.n.* (UC). MEXICO; Baja California: Ensenada, 28°44′34″N 113° W, 8 Mar 2002, *Reina* & al. *2002-103* (WLU); San Quintin, 24 May 1889, *Brandegee 2* (MO); Nayarit: San Blas, 20 Apr 1867, *Maltby 21* (UNH); Sonora**:** Sonoyta, 28 Apr 1991, *Felger 91-5* (MEXU). ***Cuscuta suksdorfii*:** U.S.A., California: Madera Co., Miller Meadow, 9 Aug 1958, *Howell 34238* (CAS); Mariposa Co., Yosemite National Park, 20 Jul 2004, *Colwell AC-04-159* (TRTE, WLU) [#470; EF194503, EU883473, EF194714, EU883524]; Nevada Co., 1.5 mi S of Sagehen Creek, 11 Aug 1970, *True & Howell 6233* (CAS); Placer Co., ca 1/4 mile E of Yuba Gap and Highway 80, 22 Jul 2002, *Ahart 9885* (JEPS) [#635; EF194501, EU883474, EF194712, EU883525]; ca. 2 mi of Humbug Summit - Lost Lake, 22 Jul 1989, *Oswald & Ahart 3949* (CHSC) [#636; EF194502, EU883475, EF194713, EU883526]; Plumas Co., ca. 0.5 mi N of the S boundary of Lassen Volcanic National Park, 25 Aug 1993, *Oswald & Ahart 5874* (CHSC); Siskiyou Co., Siskiyou Mts., Head E Fork Horse Creek, S9 T47 N, R10 W, 21 Aug 1934, *Wheeler 3192* (type of *C. suksdorfii* var. *subpedicellata*, NY). Washington: Skamania Co., on an island of a mountain lake, 24 Sep 1891, *Suksdorf 1487* (type of *C. suksdorfii*, NY).

Appendix 2. Characters used in the morphometric analysis of the *Cuscuta salina* group.

***Continuous characters*—1.** Stem diameter (mm). **2.** Flower diameter measured at the base of the corolla lobes (mm). **3.** Pedicel length (mm). **4.** Bract length (mm). **5.** Calyx length measured from the receptacle to the tips of the calyx lobes, on the abaxial surface <mm>. **6.** Length of exserted part of corolla (above tips of calyx lobes to tips of corolla lobes, mm). **7.** Calyx tube angle, measured at the base of calyx, with reference to the central floral axis (rad). **8.** Calyx lobe divergence angle, with reference to the central floral axis (rad). **9.** Calyx tube length, measured on the adaxial surface (mm). **10.** Calyx lobe length along the adaxial midline (mm). **11.** Calyx lobe maximum width (mm). **12.** Calyx lobe width at base (mm). **13.** Calyx lobe, angle formed by margins at the apex (rad). **14.** Corolla tube length along the adaxial surface (mm). **15.** Corolla lobe length along the adaxial midline (mm). **16.** Corolla lobe maximum width (mm). **17.** Corolla lobe width at base (mm). **18.** Corolla lobe, angle formed by margins at the apex (rad). **19.** Overlapping of corolla lobes measured on dissected corollas, on the adaxial face (mm). **20.** Staminal filament length (mm). **21.** Anther length (mm). **22.** Anther width (mm). **23.** Infrastaminal scales (IFS): length of fimbriate portion (mm). **24.** Afimbriate portion of the IFS length (measured at the base of the IFS) (mm). **25.** Interscale bridge length (mm). **26.** Maximum width of IFS including the fimbriae (mm). **27.** Maximum width of IFS not including the fimbriae (mm). **28.** Fimbriae length on the whole IFS (mm). **29.** Fimbriae length on the lower half of the IFS (mm). **30.** Fimbriae length on the upper half of the IFS (mm). **31.** Longest fimbria length (mm). **32.** Number of fimbriae per IFS (nr). **33.** Ovary length (mm). **34.** Length of the longer style (mm). **35.** Length of the shorter style (mm). **36.** Stigma diameter **(**mm). ***Ratio characters*—37.** Maximum width of corolla lobes vs. width at the base of corolla lobes. **38.** Calyx length vs. exserted corolla length. **39.** Calyx lobe length vs. calyx tube length. **40.** Corolla lobe length vs. corolla tube length. **41.** IFS length vs. corolla tube length. **42.** IFS length vs. IFS width not including fimbriae. **43.** Number of fimbriae on the upper half of IFS vs. number of fimbriae on lower half of the IFS. ***Qualitative characters*—44.** Corolla lobe apex (and margins under the apex): 0. entire, 1. irregular, 2. tridentate. **45.** Laticifers: 0. absent, 1. few present, 2. many present. **46.** Papillae on the corolla lobes: 0. absent, 1. present. **47.** Dome shaped epidermal cells on the corolla lobes: 0. absent, 1. present. **48.** Papillae on the calyx and/or pedicel: 0. absent, 1. present. **49.** Seeds per capsule: 0. one seed only, 1. one or two seeds on the same plant (multiseeded capsules). **50.** Fusion of IFS with the corolla tube: 0. 80–100% of the IFS length fused, 1. 50–70% of the IFS length fused. **51.** Corolla lobe orientation at full anthesis: 0. patent to reflexed, 1. erect to spreading. **52.** Stamens at full anthesis: 0. included, 1. exserted. **53.** Flower colour observed on dried material: 0. cream-yellow (light), 1. brownish (darkened).