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Chapter Nineteen: *Phaeoceros proskaueri* sp. nov., a New Species of the *Phaeoceros hallii* (Austin) Prosk.—*Phaeoceros pearsonii* (M. Howe) Prosk. Complex and the Systematic Affinities of *Paraphymatoceros* Hässel

Barbara J. Crandall-Stotler,[1] Raymond E. Stotler,[1] William T. Doyle,[2] and Laura L. Forrest[3]

[1] Department of Plant Biology, Southern Illinois University, Carbondale, IL 62901-6509, USA; Missouri Botanical Garden, St. Louis, MO 63166-0299, USA
[2] Department of Ecology and Evolutionary Biology, University of California, Santa Cruz, California 95064 USA; University Herbarium, University of California, Berkeley, California 94720-2465 USA
[3] Royal Botanic Garden Edinburgh, Inverleith Row, Edinburgh EH3 5LR, Scotland, UK

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Dedication

This paper is contributed in honor of our respected colleague John J. Engel on the occasion of his 40 years of scientific publications and his distinguished career at the Field Museum.

Abstract

The bryophyte flora of the Pacific Northwest includes three previously known species of tuber-producing hornworts that occupy seasonally dry habitats. Except for *Phymatoceros bulbiculosus* (Brot.) Stotler, W.T. Doyle & Crand.-Stotl., which was recently segregated

from *Phaeoceros* Prosk. on the basis of a combination of morphological and molecular studies, no comprehensive studies of these taxa have been undertaken since their naming. As a consequence, confusion exists regarding their taxonomic identity. This study combines field observations with morphological and molecular data to evaluate the systematic status of *Phaeoceros hallii* (Austin) Prosk. and *Phaeoceros pearsonii* (M. Howe) Prosk. These data support the recognition of a new species, *Phaeoceros proskaueri* Stotler, Crand.-Stotl. & W. T. Doyle, in this complex of related taxa. In addition, morphological studies of the lectotype of *Anthoceros tuberosus* Taylor (= *Phaeoceros tuberosus* (Taylor) Prosk.), which is formally designated here, and the holotype of *Paraphymatoceros diadematus* Hässel demonstrate that neither is related to *P. hallii*, *P. pearsonii* or *P. proskaueri*. *Phaeoceros tuberosus* is aligned with *P. laevis* (L.) Prosk. and *Paraphymatoceros hallii* (Austin) Hässel is placed as a synonym of *Phaeoceros hallii*. *Paraphymatoceros diadematus* is postulated to be congeneric with *Phaeomegaceros* Duff et al., and the following new combinations are made: *Paraphymatoceros fimbriatus* (Gottsche) Stotler, *Paraphymatoceros coriaceus* (Steph.) Stotler, *Paraphymatoceros hirticalyx* (Steph.) Stotler, and *Paraphymatoceros skottsbergii* (Steph.) Stotler.

Introduction

The foothills and valleys of the coastal and central mountains of Washington, Oregon, and California support several species of hornworts that perennate through the long dry summers by forming tubers at the conclusion of their abbreviated winter to early spring reproductive growth cycles (Doyle & Stotler, 2006). Included among these are *Phaeoceros hallii* (Austin) Prosk. and *P. pearsonii* (M. Howe) Prosk., both characterized by somewhat flattened, apical, and/or marginal tubers and *Phymatoceros bulbiculosus* (Brot.) Stotler, W. T. Doyle & Crand.-Stotl. [including *Anthoceros phymatodes* M. Howe (Crandall-Stotler et al., 2006)], which bears stalked ventral tubers. Recent studies have confirmed that the latter taxon is not only morphologically distinct from other tuber-producing species of *Phaeoceros* Prosk. (Crandall-Stotler et al., 2006) but, in fact, comprises a separate lineage in hornwort phylogeny (Duff et al., 2007). *Phaeoceros hallii* and *P. pearsonii*, which occupy similar habitats as *Phymatoceros* Stotler, W. T. Doyle & Crand.-Stotl., are resolved as sister taxa in a lineage that is weakly supported as part of the *Phaeoceros* clade (Duff et al., 2007: fig. 4). Although recognized as distinct taxa in systematic treatments since Howe (1898), diagnostic descriptions of these two species have typically focused on features of mature sporophytes and spores (e.g., Hässel de Menéndez, 1989; Hasegawa, 1991), leaving many details of their gametophyte morphologies unrecorded. As intimated by Proskauer (1957) and Schuster (1992), an overreliance on spore morphology confounds species identification, especially since spore wall architecture can change during late stages of maturation (Crandall-Stotler et al., 2006). Our studies of gametophytes as well as sporophytes in numerous herbarium collections of anthocerotes from California, coupled with extensive field monitoring of populations, have convinced us that there is in the *P. hallii – P. pearsonii* complex a previously undescribed tuber-bearing element that shares features with each of these species but is nonetheless distinct from both. This study was undertaken to clarify the systematic circumscriptions of *P. hallii* and *P. pearsonii* and evaluate the taxonomic status of this element, employing a total evidence approach.

Taxonomic History

Austin (1875) recognized 14 species in his treatment on the hornworts of North America, nine of which were newly described. Five of the new species, namely *Anthoceros fusiformis* Austin, *A. hallii* Austin, *A. oreganus* Austin, *A. stomatifer* Austin, and *A. sulcatus* Austin, were based completely or in part on collections made by Elihu Hall in Oregon. With each of the specimen packets, Austin wrote "R. Mt. Hall" or what perhaps was "R. Mts. Hall." We observed this notation with the types of *A. hallii*, *A. oreganus*, and *A. sulcatus*, and Howe (1898) recorded it for *A. fusiformis* and *A. stomatifer*. Regardless, Austin never made mention of this in any of his

publications, and we presume that it was simply an identifier, since Hall was a well-know Rocky Mountain naturalist. Ewan (1950) pointed out that Hall collected plants in the Rocky Mountains of central Colorado in 1862 and that his specimens were poorly labeled, with some specimens bearing only the name Hall. It is highly unlikely that Austin would have received any Hall specimens from Colorado because they predated his study of bryophytes, and it is not likely that any of the hornworts that Austin described were from the Rocky Mountains. Although the abbreviation could stand for Round Mountain, located in central Oregon and not far from known Hall collecting sites in Salem and Silverton, Oregon, there is no evidence for that either. Unfortunately, "R. Mt." prompted the annotation "Rocky Mts." to be written on these packets by B. Carrington or W. Pearson, who together purchased the Austin herbarium following his death, and this locality error, Rocky Mountains, has been perpetuated in the literature (e.g., Howe, 1898: 18; Hässel de Menéndez, 1989: 718; Schuster, 1992: 772).

Specimens from three locations were cited by Austin (1875) in his treatment of *Anthoceros hallii*. These included a "fertile plant on the ground" from Silverton, Oregon, and sterile material from dripping rocks, Salem, Oregon, collected by E. Hall. He also reported specimens from "swamps, Marvin [sic = Marin] County, California" collected by H. Bolander with thalli said to be "plicate-costate or lamellate; the lamellae bearing elliptical tubers underneath." In a revision of North American hornworts, Howe (1898) selected the fertile Silverton, Oregon, collection, now in the herbarium of the University of Manchester, Manchester Museum (MANCH), as the lectotype specimen for this name. The sterile material from Salem, Oregon, is likewise in MANCH, but the Bolander specimen is not, nor is it cited in either Howe (1898) or Hässel de Menéndez (1989: 731).

In a discussion of *Anthoceros sulcatus*, Austin (1875) wrote of Hall specimens that he had initially aligned them with the genus *Notothylas* Sull. ex A. Gray and that he had applied the manuscript name of *Notothylas hallii* to them in 1874. That initial generic placement was no doubt prompted at least in part by the rather short capsules that are indehiscent or very imperfectly so. With further study, not only did he reconsider, he concluded "*Notothylas Hallii MSS*" to represent the connecting link between the two genera, prompting his reduction of *Notothylas* to *Anthoceros* L. That, of course, necessitated a name change to prevent a later homonym, whereby he chose the epithet "*sulcatus*," in reference to the capsules that were scored with longitudinal furrows. His only specimen citation was "On moist earth, Salem, Oregon, *E. Hall*." Although Austin (1875) contrasted this species only with *N. orbicularis* (Schwein.) A. Gray, Howe (1898: 11) made mention of studying original specimens with the name "*Notothylas hallii*" (= *A. sulcatus*) and considered them to clearly represent *A. hallii*. Five separate packets of the original material are extant in MANCH.

During his studies of North American hornworts, Howe (1898) described *Anthoceros pearsonii* M. Howe, which he depicted as the commonest yellow-spored taxon of the Pacific coast. It was contrasted with *A. hallii* by the development of capsules that are normally four times longer, by the production of more perfect pseudoelaters, and by distinct spore surface markings. Several populations were recorded, with *Howe 16*, from Mill Valley, Marin County, California, designated as the holotype.

An additional species name that became associated with this complex is *Anthoceros bolanderi* Steph. named by Stephani in 1916. Although Frye and Clark (1947) wrote that they were not able to distinguish *A. bolanderi* from *A. pearsonii*, they did not make a formal reduction. That was done by Hässel de Menéndez (1989: 731), who listed the type "U.S.A. California, Mendocino City, Bolander (ex Herb. Gottsche) (G)" as identical to the entry by Bonner (1962: 171) in *Index Hepaticarum*. Because of a probable printer error, "Hab.," (an abbreviation for place of growth) which routinely followed all of Stephani's entries in the six volumes of *Species Hepaticarum* (Stephani, 1898–1924), is missing after the description of *A. bolanderi*. Unfortunately, there is no certain way to know the collection location other than making the assumption that because it was named in honor of Henry Bolander, that it is likely a Bolander collection, thus likely from California. A collection in the Conservatoire et Jardin Botaniques, Genève (G) with these data in the hand of Stephani represents the original material. Therefore, the specimen referred to in G by Hässel de Menéndez (1989: 731) should be considered the lectotype. A portion of that material also exists at UC.

When Proskauer (1951) segregated the genus *Phaeoceros* from *Anthoceros*, he made a number of new combinations, one being *P. hallii*. At the

same time, he listed *Anthoceros phymatodes* as a synonym of *P. hallii*, on the basis of evidence from cultured plants. It has been recently shown, however, that *A. phymatodes* not only is not related to *P. hallii*, but represents a novel genus described as *Phymatoceros* (Crandall-Stotler et al., 2006). Recently, another interpretation has been proposed regarding *P. hallii*. Hässel de Menéndez (2006) named the hornwort genus *Paraphymatoceros* Hässel on the basis of a new species, *Paraphymatoceros diadematus* Hässel, from Chile. In that same publication she removed *P. hallii* from *Phaeoceros* and transferred it to *Paraphymatoceros*, while retaining *P. pearsonii* in *Phaeoceros.* This once again brings into question the systematic affinities of *P. hallii*. Consequently, although the focus of this study is the delimitation of species within the *P. hallii* complex, we also treat *Paraphymatoceros*.

Materials and Methods

MATERIALS—Morphological features were scored for numerous freshly collected specimens from California and Oregon and a broad sample of herbarium collections, including type specimens of *Anthoceros hallii*, *A. sulcatus*, *A. pearsonii*, *A. tuberosus* Taylor, *A. bolanderi*, and *Paraphymatoceros diadematus*, with a combination of optical and scanning electron microscopy (SEM). A list of specimens examined in addition to those cited in the taxonomic section, is included in the appendix to this manuscript.

MORPHOLOGICAL METHODS—Freshly collected plants from *Doyle 11*,*366* (*P. hallii*) and *Doyle 11*,*328*, *11*,*357*, and *11*,*422* (*Phaeoceros proskaueri* sp. nov.) were fixed in formalin–acetic acid–ethanol (FAA), dehydrated in a graded series of tertiary-butyl alcohol (TBA), and embedded in Paraplast for serial sectioning (Jensen, 1962). Sections were cut at 8 μm, stained with contrasting Iron Alum Safranin O and 1% Fast Green FCF in 70% ethanol and mounted in Histoclad. Axenic cultures were established from collections of *P. pearsonii* [*Doyle 11*,*377* & *Doyle 11*,*429*], *P. hallii* [*Doyle 11*,*366*], and *P. proskaueri* sp. nov. [*Doyle 11*,*339*], employing the methods of Hatcher (1965) for comparisons with field populations and to provide soil-free material for transmission electron microscopy (TEM) and molecular studies. All phenological data were extracted from field populations sampled over several growing seasons.

Small samples of thalli from the types of *A. tuberosus* and *A. hallii* were restored for SEM following the methods of Hofmann et al. (1996), but with graded replacement of FDA by 100% ethanol before critical point drying (Crandall-Stotler et al., 2006). Freshly collected plants and samples from axenic culture for SEM study were fixed in 2% glutaraldehyde/2% paraformaldehyde in 0.1 M sodium cacodylate at pH 7.2 at 4°C, overnight, postfixed in 2% aqueous OsO_4 for three hours and dehydrated through a graded ethanol series. Both types of samples were critical point dried in a Tousimis Samdri-750 CPD, using CO_2 as the transition fluid, and then mounted on stubs covered with sticky tape. Spore samples were dispersed from air-dried capsules directly onto stubs covered with sticky tape. Samples were coated with 400–450 Å gold-palladium in a Denton Desk II sputter-coater. Specimens were viewed and images captured with either a Hitachi H500 SEM (UC Santa Cruz) or S570 SEM (SIUC).

For TEM, thalli from the axenic culture of *P. proskaueri* sp. nov. (*Doyle 11*,*339*) were fixed four hours at 25°C in 2% glutaraldehyde/2% paraformaldehyde in 0.1 M sodium cacodylate buffer, pH 7.2. After three rinses in buffer, they were postfixed in buffered 2% OsO_4, 3.5 hours at 25°C. They were dehydrated through a graded ethanol series to 100% ethanol, which was replaced through a graded series with 100% propylene oxide (PO). Slow infiltration of Spurr's resin involved the following steps: 1) 24 hours in 25% (v/v) Spurr's resin in PO, 2) 36 hours in 50% (v/v) Spurr's resin in PO, 3) 48 hours in 75% (v/v) Spurr's resin in PO, 4) two 24-hour changes of 100% Spurr's resin. After a third change of resin, the specimens were placed in molds and cured for 24 hours at 65 ºC. Thin sections were poststained for five minutes each with 2% uranyl acetate and basic lead citrate and viewed on a Hitachi H500 TEM.

MOLECULAR METHODS—DNA from the axenic culture of *Phaeoceros proskaueri* (*Doyle 11*,*339*) was extracted, amplified, and sequenced for the chloroplast gene *rbc*L (GenBank EU283415), following the protocol given in Forrest and Crandall-Stotler (2004). Additional hornwort *rbc*L sequences were downloaded from GenBank and manually aligned in PAUP* 4.0b10 (Swofford, 2002). The GenBank numbers for these sequences are given in Figure 7. The resulting matrix comprised 32 taxa and 1135 included characters, of which 354 were variable and 228 were

parsimony-informative. Maximum parsimony analyses were run in PAUP* under Fitch parsimony, with 10,000 random addition replicates, saving 25 trees per replicate. Eight most parsimonious trees were found, with a length of 541 steps. Bootstrap (BS) analysis was performed with the use of a heuristic search strategy, with 1000 replicates, each with 25 random addition replicates and saving no more than 25 trees per replicate. Maximum likelihood was implemented in GARLI 0.951 (www.bio.utexas.edu/faculty/antisense/garli/Garli.html; Zwickl, 2006) with 200 BS replicates and the General Time Reversible model (Yang, 1994) with a gamma rate distribution, which was selected as the best fit to the data by Duff et al. (2007). Three independent runs with the default parameters were used to identify the likeliest topology; each run found the same tree (Fig. 7) with Ln –46331.27762. The resulting tree was rooted on *Leiosporoceros dussii* (Steph.) Hässel, following its sister placement to all other hornworts in Duff et al. (2007).

Morphological Treatment

Morphological evidence unambiguously supports the recognition of three species in the problematic *P. hallii* group—*P. hallii*, *P. pearsonii*, and *P. proskaueri*, a new species named herein. All three are endemic to the Pacific Coast of North America, with *P. hallii* and *P. pearsonii* being broadly distributed from Washington to southern California and *P. proskaueri* known to date only from California. They occupy comparable habitats, sometimes even forming contiguous patches in a single locale, and have rather similar facies. Their elongate, more or less strap-shaped thalli bear abundant lateral or terminal tubers that typically persist on the soil surface. In contrast to *Phymatoceros*, which is sometimes intermixed with one or more of these species, the tubers are never ventral in position and rarely become deeply buried by the downward growth of a subtending stalk. A number of features separate them, as a group, from more widely distributed elements of the *P. laevis* (L.) Prosk. complex, the most consistent of which are seen in distal spore wall ornamentation. In the *P. hallii* group, the distal spore face bears a species-dependent, variable number of elongate, rounded ridges and/or mammillae, but in the *P. laevis* group, the distal spore face is spinose. The proximal spore face in both groups can be finely vermiculate, often with clustered or scattered papillae. Spore morphology, as well as many other characters, also clearly separates *P. hallii* from *Paraphymatoceros* (Table 1). Although there are traits that suggest close affinity among the species of the *P. hallii* complex, each of them possesses a unique assemblage of characters, as detailed below.

Phaeoceros hallii (Austin) Prosk.—*Phaeoceros hallii* grows in moist, shaded habitats, over rocks in creeks or cascades, or on soils that are slow to dry, as in seeps or along creek banks, often mixed with grass, mosses, or both. As exemplified by the lectotype and paratype specimens of *A. hallii* (*Hall 26* & *35*, respectively) and *A. sulcatus* (*Hall 25*), thallus form is dependent on the moisture level of the habitat, a fact that likely influenced Austin (1875) to name two species. Specimens referred to *A. hallii* by Austin (1875) have elongated, flattened thalli with numerous tubers terminating most of the branch apices; fertile plants from the Silverton locality (*Hall 26*) bear 1 or 2 sporophytes at the thallus apices. The presence of diatoms on the thalli confirms that these plants were growing in very wet habitats, as indicated also by the notations, "springy places," "dripping rocks," or "cascades" on the packets. In contrast, the type specimens of *A. sulcatus* growing "on moist earth, Salem, Oregon," (Austin, 1875: 27) have much shorter, somewhat thicker thalli that bear very few tubers and are crowded with short sporophytes. Indeed, most populations of *P. hallii* express this latter morphology. Except for these differences in thallus form, which appear to be environmentally induced, the type specimens of *A. hallii* and *A. sulcatus* are anatomically identical and referable to a single species as originally proposed by Howe (1898). This includes the sterile specimens on dripping rocks (*Hall 35*), which Howe (1898: 11) mistakenly suggested were likely his new species, "*A. pearsoni*."

Thalli are caespitose as they germinate from the dormant tubers but become more or less prostrate and intertwined in irregular mats with continued growth. They are linear, 1.3–3.0 mm in width, and irregularly branched, with the branches narrower than the main axis. Late in the growing season, dorsiventrally flattened, orbicular, dark green to blackish tubers, to 1.5 mm in diameter, terminate many of the lateral branches, as well as the main axes of thalli without sporophytes (Fig. 1.2, Table 1). As the plants dry, the wing margins inroll, imparting what Howe (1898: 10) described as a "plicate-costate" appearance to the thallus.

TABLE 1. Morphological features of the *Phaeoceros hallii* complex and *Paraphymatoceros diadematus*. Items marked with an asterisk (*) were taken from Hässel de Menéndez (2006).

Taxon/Character	*Phaeoceros proskaueri*	*Phaeoceros hallii*	*Phaeoceros pearsonii*	*Paraphymatoceros diadematus*
Thallus width	Males 0.3–0.8 mm; females 1.5–2.0 mm	1.3–3.0 mm	2.5–5.0 mm	0.7–3.0 mm
Thallus margin	2-stratose, abrupt constriction	3 or 4-stratose, abrupt constriction	2 to 4-stratose, gradual taper	1-stratose, gradual taper
Plastid structure	Spheroidal to spindle-shaped; pyrenoid absent	Plate-like to spindle-shaped; pyrenoid present	Spheroidal to plate-like; pyrenoid absent	Pyrenoid absent*
Tuber morphology	Terminating "dwarf" lateral branches; subspheroidal, rarely flattened, < 1 mm	Terminating apices, flattened, to 1.5 mm	Marginal or apical, flattened, to 2.5 mm	Submarginal–ventral, flattened, to 2.0 mm
Sexuality	Dioicous, dimorphic	Monoicous,weakly protandrous	Monoicous, strongly protandrous	Dioicous*
No. of antheridia/ chamber	1	2 to 4	1	4 or 5*
Capsule length (cm)	0.7–1.75 cm	0.4–0.6 cm	2.0–4.0 cm	4.8–7.0 cm
Columella	Rigid, brown	Flaccid, hyaline	Rigid, brown	Rigid, brown
Spore Maturation	Nonsynchronous	Synchronous	Nonsynchronous	Nonsynchronous
Dehiscence	1 longitudinal slit, valves adherent at the apex	Indehiscent, or with 1 short slit; valves adherent at the apex	1 longitudinal slit, slightly twisted; rarely 2-valved; valves adherent at the apex	2-valved, helically twisted; apically free
Spore color at dehiscence	Deep brown to fuscous	Bright yellow to yellow-orange	Tawny to light blackish brown	Tawny to medium blackish brown
Pseudoelaters in median part of capsule	Brown, mostly 2- or 3-celled, to 110 μm long	Pale yellow to tan, mostly 1-celled, <60 μm long	Brown, 3- or 4-celled, to 200 μm long	Blackish brown, 3- or 4-celled, to 400 μm long

The main axis is 6 to 9 cells thick for most of its width, narrowing abruptly to 3 or 4 cells in thickness within 100 μm of the thallus margin. Internal cells are thin-walled, isodiametric, and 35–48 μm in diameter, with slightly larger mucilage cells dispersed among them. Cells of the dorsal epidermis are rhombic pentagonal to hexagonal (rarely quadrate) and 38–46 μm in diameter, with cells near the thallus midregion, elongated to twice their width (Fig. 1.3). Each dorsal epidermal cell bears a single, pleomorphic plastid that can assume a flattened, amoeboid-like outline, or become spindle-shaped to spheroidal, depending on environment. Ventral epidermal cells, which are smaller than the dorsal cells, averaging 25 μm in diameter, typically have only spindle-shaped to subspheroidal plastids, as do all cells of immature thalli grown in axenic culture and cells of developing tubers. A spheroidal pyrenoid-like zone of starch grains is clearly visible in the center of each plastid in the amoeboid or spindle phases (Fig. 1.3), but not in the subspheroidal phase, at least in optical microscopy. Although this structure resembles the pyrenoids of the *P. laevis* complex, only ultrastructural studies can determine with certainty whether this region possesses pyrenoid organization or is simply a centralized aggregation of starch grains.

Rhizoids, which are abundant on thalli growing over soil, typically adhere to each other in

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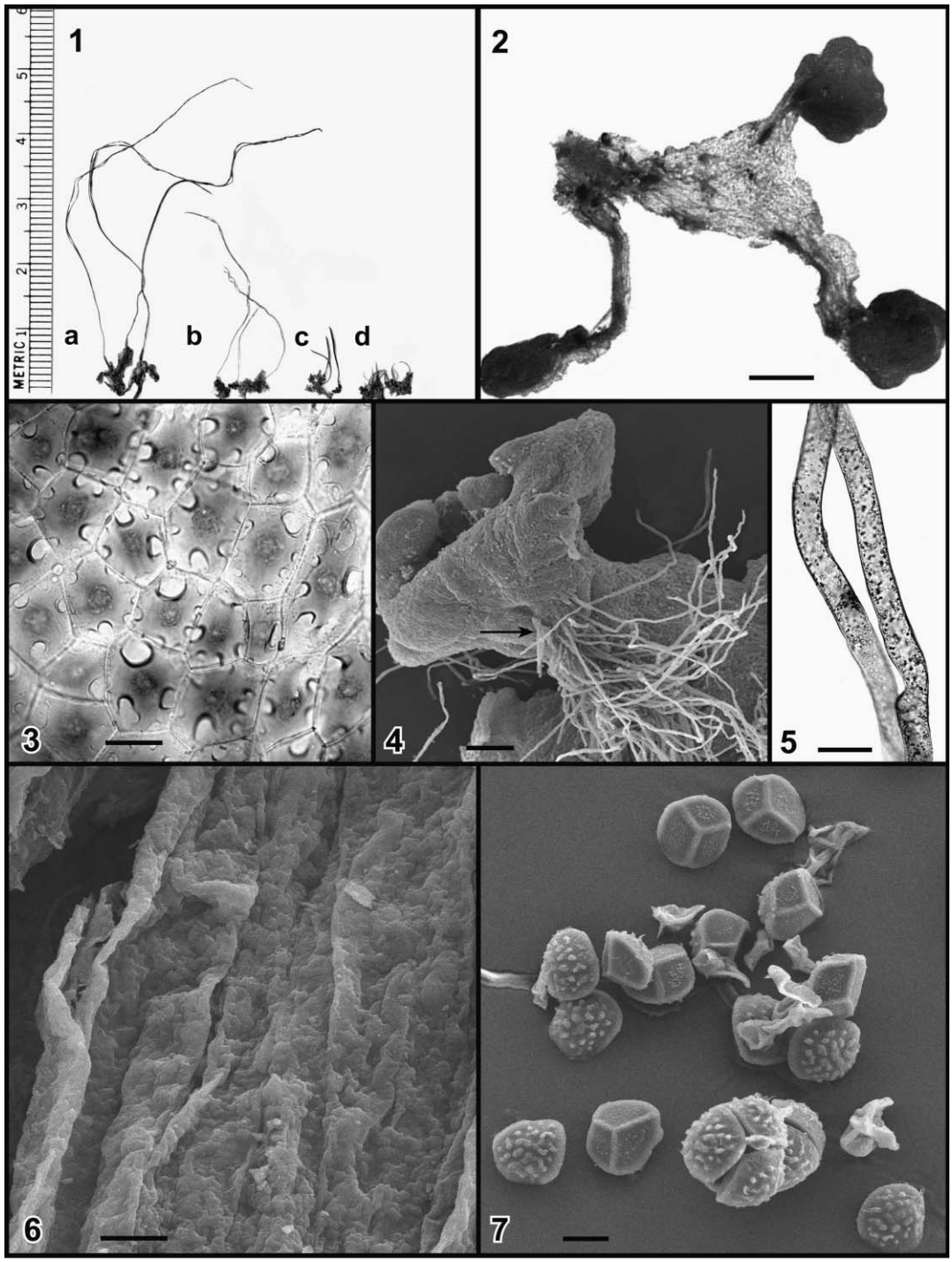
FIG. 1. *Phaeoceros hallii* (Austin) Prosk. **1.** Comparison of sporophyte-bearing plants of *Paraphymatoceros diadematus* Hässel (**a**; from the holotype), *Phaeoceros pearsonii* (M. Howe) Prosk. (**b**; from *Doyle 11415*), *Phaeoceros proskaueri* Stotler, Crand.-Stotl. & W. T. Doyle (**c**; from *Doyle 7268*) and *P. hallii* (Austin) Prosk. (**d**; from *Doyle*



8445), all natural size. **2.** Portion of a thallus with terminal tubers (1-mm scale). **3.** Surface view of the dorsal epidermis, showing cells with irregularly shaped plastids, with central pyrenoids (30-µm scale). **4.** SEM of a chemically preserved thallus, ventral view, showing clumped, adherent rhizoids at the arrow (250-µm scale). **5.** Granulate rhizoids, viewed with a compound microscope (35-µm scale). **6.** SEM of a group of air-dried, adherent, granulate rhizoids (8-µm scale). **7.** SEM of spores and pseudoelaters (30-µm scale). [**2** from *Doyle 11390*; **3**, **5** from *Doyle 11366*; **4** from *Doyle 11363*; **6** from the lectotype of *A. hallii*; **7** from the lectotype of *A. sulcatus*]

bundles or plate-like masses (Fig. 1.4). They are up to 20 μm in diameter, pale brown, and densely granulate in transmitted light (Fig. 1.5), an attribute noted by Austin (1875) as diagnostic of *A. sulcatus*. In addition, smooth, narrower, nonfascicled rhizoids are also present, especially on young thalli and near thallus apices. Although granulate—bundled rhizoids are a striking feature of *P. hallii*—we have observed similar rhizoids, sometimes intermixed with rhizoids that are smooth walled, or nongranulate, in many other hornworts, including other species of *Phaeoceros*, *Notothylas*, *Paraphymatoceros*, and *Phaeomegaceros* Duff et al. Transverse sections of these distinctive rhizoids demonstrate that the granular appearance is due to deposits on the inner surface of the rhizoid cell wall, much like the tubercles or pegs of pegged rhizoids in complex thalloid liverworts. As a consequence, in SEM micrographs, the outer wall of the rhizoid appears smooth in noncollapsed, chemically fixed material (Fig. 1.4), but irregularly roughened in air-dried material in which surface wall collapse exaggerates the inner, noncollapsed deposits (Fig. 1.6). Despite their widespread occurrence in hornworts, these granulate rhizoids have not been previously described. Schuster (1992) does indicate that the rhizoids of *Anthoceros* sensu R.M.Schust. (= *Phaeoceros*) are smooth or punctate (p. 743) and those of *Aspiromitus* Steph. sensu R.M.Schust. (= *Anthoceros*) are smooth or weakly roughened (p. 781), without further discussion, but most authors describe hornwort rhizoids as unicellular and smooth (e.g., Renzaglia & Vaughn, 2000).

As first reported by Howe (1898), *P. hallii* is monoicous, and slightly protandrous. Antheridial chambers are frequently borne on different branches than the archegonia, with up to 6 chambers per branch, but can occasionally be observed on the main thallus, somewhat posterior to a cluster of developing sporophytes. The chambers are 350–400 μm in diameter and contain 2 to 4 antheridia. When mature, the rupture of the chamber roof forms a collar 2 to 4 cells high that persists around the chamber cavity.

Sporophytes are usually numerous, occurring in clusters near thallus apices (Fig. 1.1). Involucres are no more than a third the length of the capsules and are cylindrical, but with the mouth slightly flared. Capsules are erect to slightly bent, less than 6 mm long, and yellow to light brown when mature. Stomates are scattered throughout the epidermis, frequently in pairs, with the guard cells brown in dried collections. In median transverse section, the capsule wall is 4 to 6(7) cells thick. The radial walls of the quadrate epidermal cells are thickened, the 2 to 4(5) layers of assimilatory cells are thin-walled, isodiametric, and 35–40 μm in diameter, and the innermost cells that line the spore sac are narrowly rectangular, averaging 8 μm in depth, with their inner tangential walls slightly thickened. A hyaline columella, consisting of up to 30 columns of elongate, fragile, thin-walled cells that collapse before spore release, extends the entire length of the spore sac. The spore sac ends 250–300 μm below the constricted, knob-like tip of the capsule.

Sporophytes of *P. hallii* resemble those of *Notothylas* in having growth from the basal meristem cease early in ontogeny, even though moisture conditions are suitable for continued growth. Sporogenesis is more or less synchronous, so mature spores fill the entire spore sac at capsule opening. With drying, the capsule shrinks, becomes longitudinally ridged and appears acutely 4-angled. Numerous undehisced capsules, bulging with mature yellow orange spores, are present in most collections, which suggests that capsules are either nondehiscent or very slow to dehisce. In the few cases in which dehiscent capsules have been observed, a single longitudinal suture begins to open about a third of the way down from the capsule tip. The epidermal cells bordering the suture are narrower than the neighboring epidermal cells but are otherwise indistinguishable, and no clearly defined suture is visible in undehisced capsules. Mature, undehisced capsules frequently break off near their insertion in the thallus; this could allow for spore release and dispersal without dehiscence.

Spores are bright yellow to yellow-orange, anisopolar and 42–60(–63) μm in equatorial diameter. Both proximal and distal surfaces are covered with a matrix of fine, interwoven vermiculate thickenings. The distal face is additionally ornamented with 20 to 35 mammillae or short rounded ridges that are 3.5–4.0 μm in width and 3.0–3.5 μm in height (Figs. 1.7, 6.1–6.4). The proximal face has a distinct trilete ridge, and each triangular facet bears a central cluster of 20 to 30 (rarely fewer) papillae, 1.0–1.2 μm in diameter. In optical microscopy, the spore appears rimmed by a thin wing, or cingulum, more than 2.0 μm wide. Pseudoelaters are pale yellow to tan, 15–20 μm wide, mostly 1-celled and usually only slightly longer than broad (Fig. 1.7).

Phaeoceros pearsonii (M. Howe) Prosk.—As initially suggested by Howe (1898), thalli of *P. pearsonii* are quite variable in size and form, with smaller, more highly branched forms occupying drier, more exposed sites. Thalli are typically larger, rather crispate and more spreading than those of *P. hallii* (Fig. 1.1, Table 1), with thallus lobes up to 5 mm wide in robust forms. Individual thalli become flabellate with branching (Fig. 2.2), with branches near the apex diverging subdichotomously. The margin of the thallus is often irregularly crenate to shallowly incised, especially in plants bearing sporophytes. Flattened, elongate, dark green tubers, up to 2.5 mm in width and of variable length, are formed at the thallus apices as well as along the thallus margins (Fig. 2.1, 2.2). In contrast to the orbicular tubers of *P. hallii*, the tubers of *P. pearsonii* are irregular in outline, sometimes including an apical dichotomy and extending for some distance down the margin from the apex (see Howe, 1898: pl. 322). Tubers formed on thin branches can become descendent with downward growth of the branch, but most remain near the soil surface when the subtending thallus dies.

In transverse section, the thalli gradually taper from 6 to 11 cells thick in the middle to 2 to 4 cells thick near the margin, with a single cell projecting at the thallus margin proper. In general, thalli growing in more exposed habitats comprised more cell layers than those from shaded habitats. Interior cells are elongate, 38.5–42.0 μm wide × 65–75 μm long, and larger and more vacuolate than the epidermal cells. Mucilage cells are widely scattered throughout the thallus. Cells of the dorsal epidermis are rhombic to elongated, 19–28 μm in diameter, and up to 70 μm long. The single large plastid in each dorsal epidermal cell is orbicular to angled and internally homogeneous, lacking a pyrenoid (Fig. 2.3), as also reported by Bartlett (1928) and Duff et al. (2007). Indeed, this difference in plastid structure immediately differentiates thalli of *P. pearsonii* from those of *P. hallii*. Cells of the ventral epidermis sometimes have 2 orbicular plastids, or plastids that are dumbbell-shaped, and interior cells of the midrib occasionally have 4 plastids (Bartlett, 1928; Doyle & Stotler, 2006).

Rhizoids are both nongranulate and granulate, with a fairly equal mix of both on any thallus. They are hyaline, fairly abundant on older parts of the thallus, and never fasciculate or adherent.

Plants are monoicous, but exceedingly protandrous. Antheridial chambers begin development with the initiation of tuber germination, sometimes forming even within the fleshy tissue of the tuber (Howe, 1898: pl. 322). As the thallus expands out from the tuber, successive rows of antheridial chambers are formed (Fig. 2.4). The chambers are spheroidal, 100–120 μm in diameter and house a single antheridium per chamber (Fig. 2.5). The chamber collar is 2 to 4 cells high and 2 cell layers thick at the base. Archegonia are formed much later in tuberling germination, after maturation and dehiscence of the antheridia on that thallus. In a natural population, some tuberlings would be producing antheridia and others archegonia, thus facilitating fertilization among tuberlings. This does not preclude self-fertilization, however, in that many tubers within a population could have been formed on the same parent thallus. We have not observed empty antheridial chambers on live thalli with mature sporophytes but have observed antheridial production on germinating tubers that are still attached to dead remnants of sporophyte-bearing thalli.

Sporophytes occur singly or in pairs near the thallus apex (Fig. 1.1b). Involucres are up to 5 mm long and narrowly cylindrical, with the mouth entire and appressed to the capsule, never flared as in *P. hallii*. Capsules with nearly mature to mature spores are tan to dark brown and range from 2 to 4 (rarely 5) cm in length, with longer capsules occurring in slower to dry habitats. Of course, length also varies with the time of collection, in that in nature, the basal meristem will continue to generate cells, even after mature spores are present at the capsule apex; thus, capsules collected in late March tend to be shorter than those collected nearer the end of the growing season in June. Stomates are dispersed throughout the epidermis. In the median part of the capsule, the wall consists of an epidermis, 3 layers of plastid-containing, assimilatory cells, and an inner "endodermis-like" layer. The radial walls of the epidermal cells are pigmented and thickened, as also are the radial and inner tangential walls of the endodermis-like layer. The columella consists of 16 columns of thick-walled cells and is visible as a smooth, rigid, pale brown to dark brown central strand at capsule dehiscence. Most capsules dehisce along a single longitudinal suture that begins just below the solid apical knob of the capsule and extends basally, but dehiscence along 2 sutures can also occur. The suture cells are narrower than the surrounding epidermal cells and unpigmented. When the capsule opens along 2 sutures, the split along one of them is often

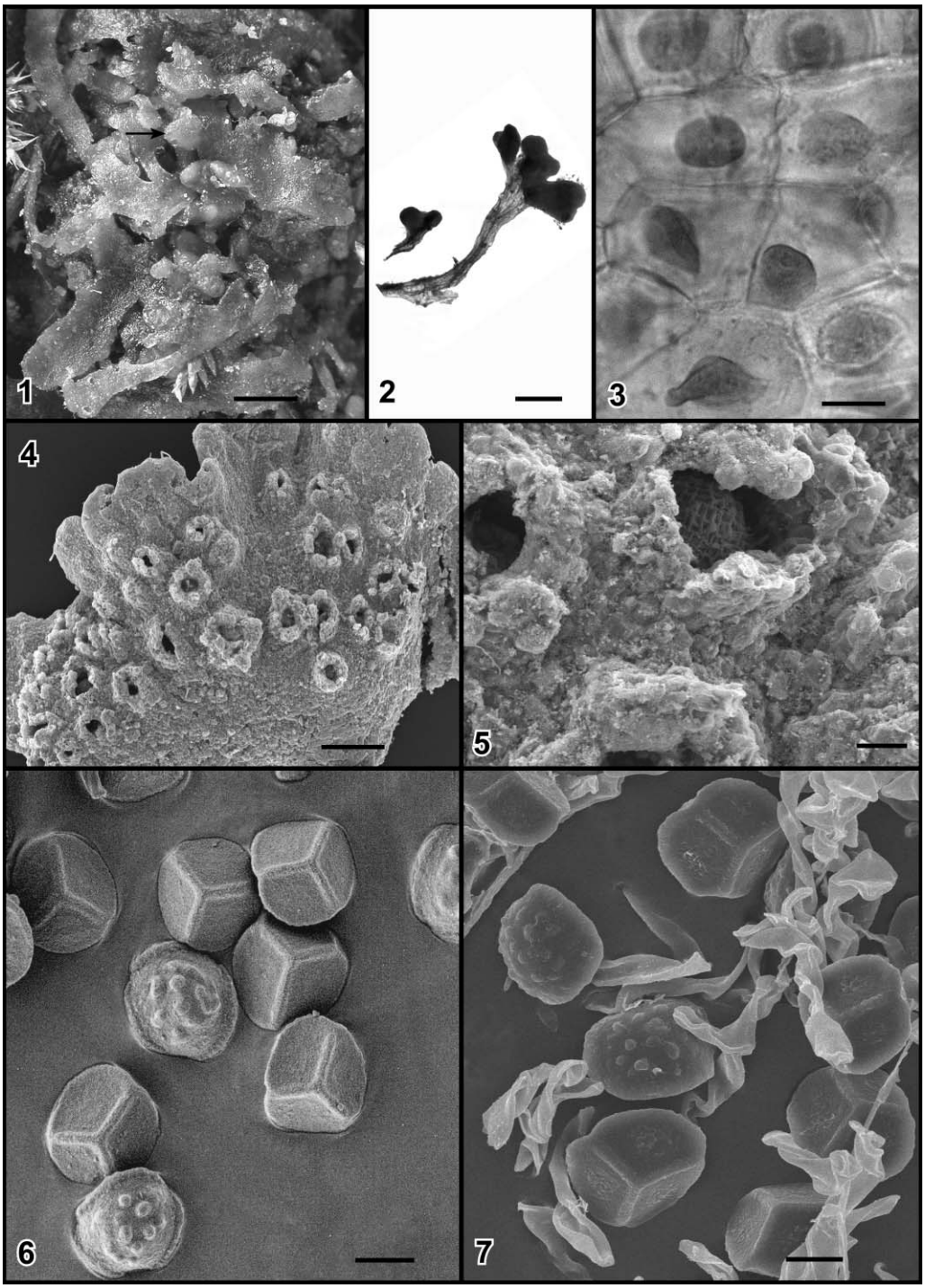


FIG. 2. *Phaeoceros pearsonii* (M. Howe) Prosk. **1.** Habit of young tuber-bearing plants, showing tubers in both lateral (at arrow) and apical positions (2-mm scale). **2.** Thalli showing apical and marginal tubers (3-mm scale). **3.** Surface view of the dorsal epidermis, showing plastids that lack pyrenoids (14-µm scale). **4.** SEM of a thallus soon after its emergence from a tuber, showing numerous rows of antheridial cavities (100-µm scale). **5.** Antheridial cavity with a single antheridium, SEM (50-µm scale). **6.** SEM of spores that are characteristic of the species (20-µm scale). **7.** SEM of spores and pseudoelaters that are light blackish brown and bear a few proximal facet papillae (20-µm scale). [**1** from *Doyle 11325*; **2**, **3** from *Wagner m2269*; **4**, **5** from *Doyle 11420*; **6** from *Doyle 8423*; **7** from *Wagner m1387*]

incomplete. In nature, the valves remain adherent at the apex and slightly twisted.

Sporogenesis is nonsynchronous, so even though a capsule is dehisced, not all spores in it are mature. Fully mature spores in field-dried specimens are usually yellow to tawny, anisopolar and 36–48 μm in equatorial diameter (Fig. 2.6). They resemble the spores of *P. hallii* in having both proximal and distal surfaces covered with a matrix of fine, interwoven vermiculate thickenings. The distal surface is further ornamented with 5 to 15 mammillae or rounded, crescentic projections, but the facets of the proximal face are generally otherwise unornamented. There is a prominent trilete ridge on the proximal face, and a distinct wing or cingulum. In optical microscopy, the proximal facets appear granulose as Howe (1898) described them. Isotype specimens of both *A. pearsonii* [CA] and *A. bolanderi* Steph. [CA] exhibit this pattern of spore wall architecture (Fig. 6.5–6.8), supporting the proposal by Hässel de Menéndez (1989) that they are conspecific. Although most populations possess spores of this type, in a few geographically dispersed populations, mature spores in field-dried capsules develop a blackish brown pigmentation that obscures the vermiculate thickenings and possess a few central papillae on some, or all, of the facets of the proximal face (Figs. 2.7, 6.11, 6.12). This spore morphology seems to occur more frequently in populations growing in exposed habitats (Doyle & Stotler, 2006) but does not define all such populations. Plants with this spore morphology are totally referable to *P. pearsonii* in all other features, leading us to conclude that spore characters by themselves are not always reliable indicators of taxon identity, a conclusion also reached by Proskauer (1957) for the *P. laevis* complex.

Pseudoelaters are brown, 8–12 μm wide, and 3 or 4 cells long in the median part of the capsule or spore tetrad zone. In the zone of spore maturation, the pseudoelaters fragment, so when dispersed, they are typically only 1 or 2 cells long. Individual cells can be up to 80 μm long.

Phaeoceros proskaueri Stotler, Crand.-Stotl. & W. T. Doyle, sp. nov.—This previously undescribed, tuberous species has been collected from numerous localities in California, including the western foothills of the Central and Southern Sierra Nevada and the coastal ranges of the South and Central Coast Regions. It is most common on disturbed, fine-grained soils that dry soon after the end of the winter–spring rains, such as exposed areas on hillsides, abandoned dirt roads, road cuts, trail banks, banks of ephemeral creeks, openings in the chaparral, or among oaks. It sometimes grows with *P. pearsonii*, *P. bulbiculosus*, and *A. fusiformis*, as well as various species of *Asterella* P. Beauv., *Fossombronia* Raddi, *Riccia* L., *Sphaerocarpos* Boehm., and *Targionia* L.

In contrast to both *P. hallii* and *P. pearsonii*, thalli of *P. proskaueri* are unisexual and highly dimorphic, with tuberling populations typically consisting of contiguous clusters of male and female plants (Fig. 3.1, 3.6). Tuber germination occurs in late fall to early winter, shortly after the onset of the rainy season. Soon after emergence from the tubers, male thalli measure less than 400 μm in width, whereas developing female thalli in the same population are more than 900 μm wide. As development continues, thallus segments on male plants reach a maximum width of 800 μm, and those of female plants enlarge up to 2.0 mm in width. Male plants are much more highly branched than females and bear more abundant tubers (Fig. 3.1, 3.2). The tubers are distinctive. They are usually subspheroidal and only slightly dorsiventrally flattened, occasionally more elongate and flattened, and develop mostly at the apices of abundantly produced, very short lateral branches. In early stages of tuber development, these short branches look like quadrate lobes of an incised thallus margin (Fig. 3.1), but they bear apical cells and can grow into normal, elongate lateral branches if tuber formation does not occur. During tuber formation, the apical cell of the branch no longer functions, and the whole branch becomes thickened. Although these tubers can look like they are marginal, they do not originate from marginal cells of the thallus. Tubers formed in this way develop early in the growing season; they are initially pale green, but with the deposit of pigments in the epidermal cell walls, are brown at maturity. Internal tissues are achlorophyllose, test negative for starch, and contain oil droplets and small protein granules. Near the end of the growing season, the apices of leading thallus segments may also become tuberous. In contrast to the early-formed lateral tubers that persist on the soil surface, these apical tubers tend to be more elongate and are sometimes pushed downward by the elongating thallus tissue behind them.

The thallus is differentiated into a broad central midrib, which bears the rhizoids, and thinner wings. In transverse section, the midrib is 10 to 16

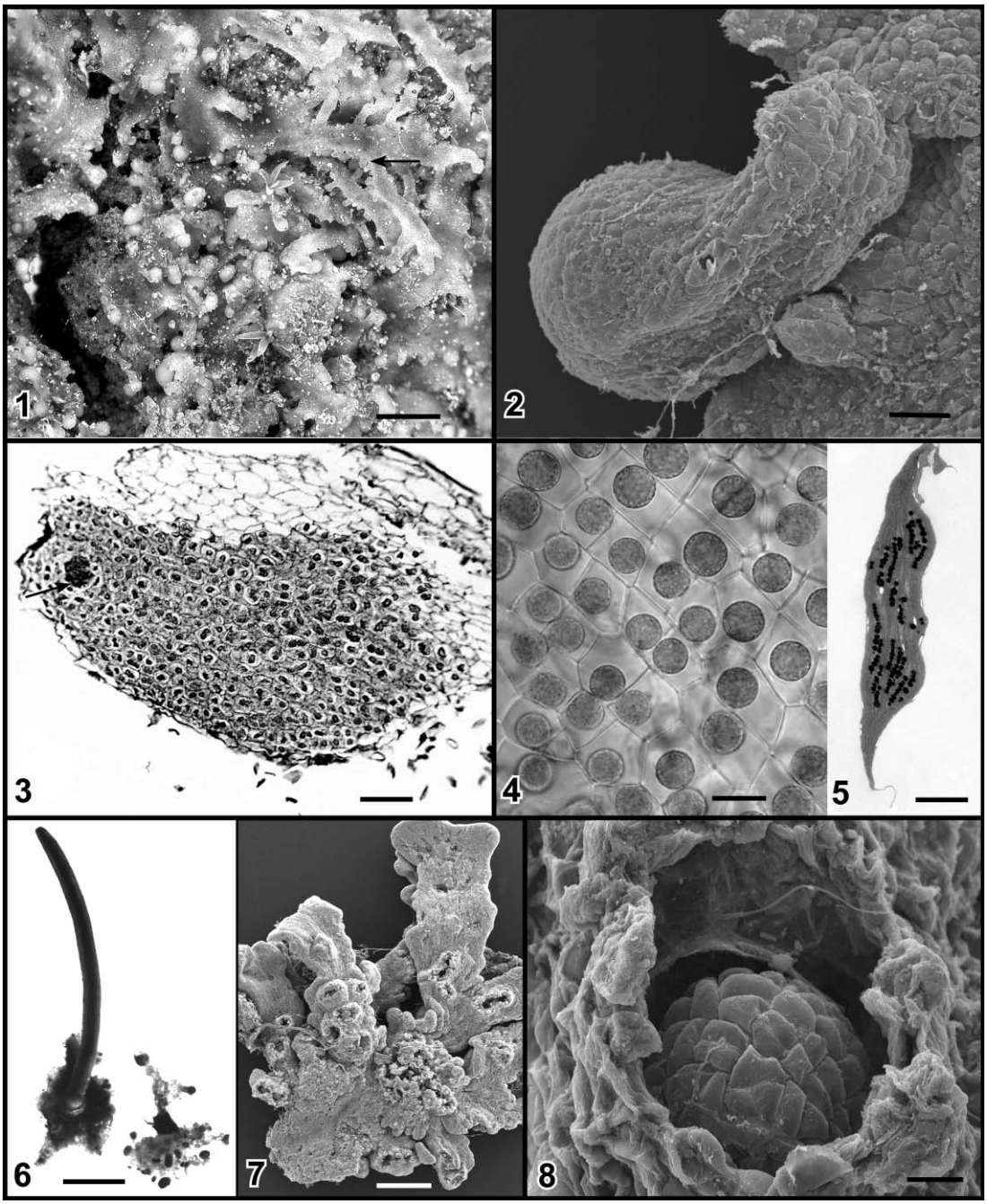


FIG. 3. *Phaeoceros proskaueri* Stotler, Crand.-Stotl. & W. T. Doyle. **1.** Habit of young tuber-bearing plants, showing very short lateral branches beginning to form tubers at their apices (at arrow) (1.5-mm scale). **2.** SEM of a typical tuber (75-µm scale). **3.** Vertical longitudinal section through a developing tuber; the apical cell and meristematic tissues of the young branch are still present, indicated by arrow (60-µm scale). **4.** Surface view of the dorsal epidermis, showing plastids that lack pyrenoids (20-µm scale). **5.** TEM of a single plastid, showing small peripheral starch grains and no pyrenoid (2.6-µm scale). **6.** Mature thalli, male (on the right) and female (on the left), showing the dimorphism of the sexes (2-mm scale). **7.** SEM of a male plant soon after emergence from the tuber, remnants of which are in the lower left (525-µm scale). **8.** Antheridial cavity with a single antheridium (25-µm scale). [**1**, **3** and male plant in **6** from *Doyle 11357*; **2** from *Doyle 11328*; **4**, **7**, **8** from *Doyle 11431*; **5** and the female plant in **6** from *Doyle 11339*]

cells thick and constricts abruptly to the wings, which are 4 to 10 cells thick. Near the thallus margins, the wings taper to 2 cells in thickness. This thinner area of the wing margin turns upward when the thallus dries. A few scattered mucilage cells, as well as the *Nostoc* colonies, are restricted to the midrib. There is no consistent difference in thallus thickness between males and females, but the wing portion of the thallus is much narrower in male plants so the midrib occupies a larger proportion of the thallus. Interior cells of the midrib are longitudinally elongated, 30.4–38.0 μm in diameter and up to 120 μm in length; interior cells of the wing are similar in size, but are radially elongated. Both dorsal and ventral epidermal cells are quadrate in transverse section, 20–30 μm in diameter, with dorsal cells rhombic pentagonal or rectangular in surface view (Fig. 3.4). Plastids are orbicular in surface view, but oval to spindle-shaped in section and definitely lack pyrenoids (Fig. 3.5). Small, dispersed starch grains and strands of osmiophilic spherules are within the plastid matrix.

Rhizoids are abundant on the thallus midrib and are mostly of the granulate type, with non-granulate rhizoids occurring only near the thallus apex and on developing tubers. Although granulate, the rhizoids are hyaline and never adherent or fascicled as in *P. hallii*.

Male thalli dichotomize right after emergence from their tubers. The two branches grow unequally, with one remaining small and close to the tuber and the other expanding to form the major axis of the plant. As in *P. bulbiculosus* (Crandall-Stotler et al., 2006), antheridial chambers form very soon after tuber germination, occurring in clusters at the base of both branches (Fig. 3.7). Individual chambers are broadly cone-shaped, 200–300 μm across at the base, and elevated above the thallus surface even before opening. Each contains a single antheridium (Figs. 3.8, 4.1). Mature antheridia are yellow-orange, up to 200 μm in diameter, with untiered jacket cells and a 4-seriate stalk, up to 3 cell rows in length. The chamber collar, formed from the roof cells of the chamber, is hyaline and up to 5 cells high. Sometimes, the opening of contiguous chambers produces a single collar, giving the appearance of a single elongate chamber with 2 antheridia. The presence of an internal partition between the antheridia, however, verifies that all chambers are developmentally monandrous. After a short period of antheridial production, the male thalli undergo a period of vegetative growth, with extensive branching and tuber production. The antheridial chambers, which are produced only near the base of the male thallus, deteriorate early in the growing season, long before the maturation of sporophytes on the female thalli, and hence are not preserved in most herbarium collections.

Female plants undergo a small amount of vegetative growth after tuber emergence, dichotomizing at least twice before producing archegonia at their broadened thallus apices. At the time of fertilization, female thalli are typically less than 1.0 mm in width, but still almost 3 times as large as male thalli in the same population. As sporophytes develop, the thalli grow in length and width and initiate the short tuber-forming branches. Tubers are fully developed by the late stages of sporophyte maturation.

Sporophytes occur singly or rarely in pairs at the apices of the female thallus lobes. They are substantially larger than those of *P. hallii*, but much smaller than those of *P. pearsonii* (Fig. 1.1c). The involucres are short, 0.6–1.5 mm high, and narrowly campanulate, with the mouth flared to 0.7 mm wide. Capsules are usually less than 1.0 cm long, but can be up to 1.75 cm in sites where moisture is available later into the season. The capsule wall consists of an outer epidermis, three layers of plastid-containing assimilatory cells and an inner "endodermal" layer (Fig. 4.2). The radial walls of both the epidermal and inner "endodermal" cells are darkly pigmented and thickened. The columella consists of 16 columns of thick-walled, elongate cells and, as in *P. pearsonii*, is visible as a rigid, brown central strand in dehiscing capsules. Stomates occur singly and are scattered throughout the epidermis. Field-matured capsules are blackish brown at the tip, to brown in the median zone of still maturing spores. They open along a single longitudinal suture that is visible as a line of yellow to orange cells, extending basally from just below the knob-like tip of the capsule. In capsules that dehisce when less than 7 mm long, the valves and dehiscence slit are straight, but in longer capsules they are helically twisted.

Sporogenesis is nonsynchronous. Spores that have just emerged from tetrads in the median to basal part of mature capsules are partially covered with remnants of the sporocyte wall and intrasporal septum (Fig. 4.3). Slightly anterior to the tetrad zone, these remnants are no longer visible, and a matrix of vermiculate thickenings can be seen covering both proximal and distal

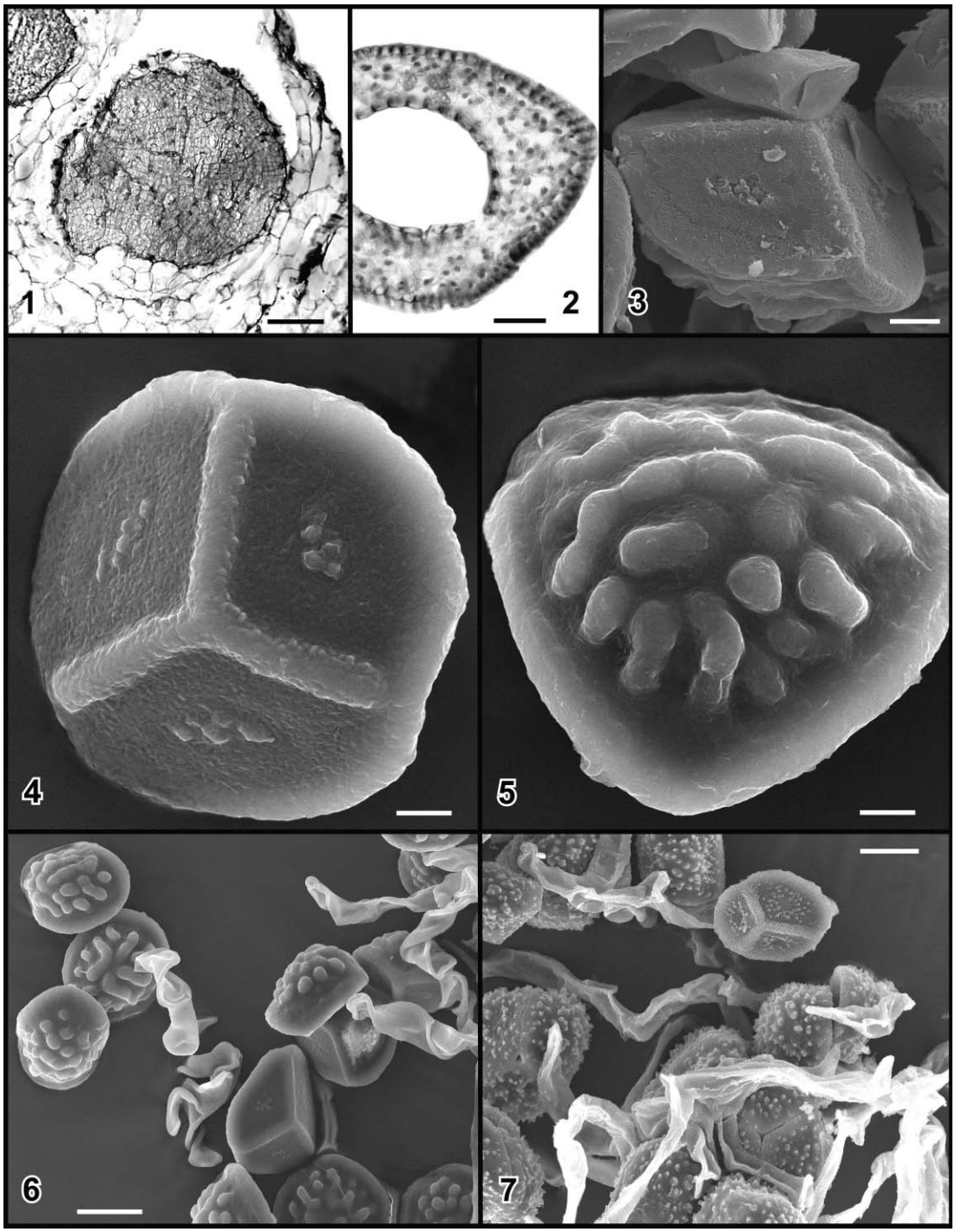


FIG. 4. *Phaeoceros proskaueri* Stotler, Crand.-Stotl. & W. T. Doyle (**1**–**6**) and *Phaeoceros tuberosus* (Taylor) Prosk. (**7**). **1.** Vertical longitudinal section through a single antheridial chamber (50-µm scale). **2.** Transverse section through capsule wall (150-µm scale). **3.** SEM of spore that has just separated from its tetrad; note the remnants of the intrasporal septum and the densely vermiculate spore wall thickenings beneath it (10-µm scale). **4, 5.** SEM of proximal (**4**) and distal (**5**) faces of mature deep brown spores from field-dehisced capsules (5-µm scale). **6.** SEM of dispersed spores and pseudoelaters from near the capsule apex (20-µm scale). **7.** SEM showing spores, spore tetrads and pseudoelaters (20-µm scale). [**1** from *Doyle 11422*; **2**, **4**–**6** from *Doyle 11339*; **3** from *Doyle 11328*; **7** from the lectotype]

faces of the spores, which at this stage are yellow to tawny. The proximal face bears a distinct trilete ridge and a small central cluster of 4 to 10 papillae, 2.5–3.0 μm in height (Figs. 4.4, 6.10). The distal face is conspicuously ornamented with 13 to 30 rounded mammillae or elongate, rounded ridges, 5.0–6.0 μm in diameter, and a cingulum, or wing, is present (Figs. 4.5, 6.9). At this stage, the spores bear some semblance of the spores of *P. hallii*. In fully mature capsules that dehisce in the field, however, spores at the capsule apex are deep brown to fuscous, and the vermiculate matrix of the spore surface is obscured (Fig. 4.4, 4.5). Such fully mature spores are smaller than those of *P. hallii*, 36–49 μm in diameter. As in *P. bulbiculosus*, there appears to be a deposition of some type of coating material over the spore wall during the final stages of spore maturation (Crandall-Stotler et al., 2006). This late modification of spore wall structure never occurs in *P. hallii*, but does occur to a limited extent in a few atypical populations of *P. pearsonii*. Spores of the latter are, nonetheless, distinguished from those of *P. proskaueri* in having fewer mammillae on the distal face and more widely dispersed, smaller proximal face papillae (Fig. 6.11, 6.12).

Pseudoelaters are brown and 2 or 3 cells long in median parts of the capsule, but fragment into 1(2)-celled structures near the capsule apex. Individual cells are 12–18 μm wide and up to 40 μm long.

Clearly, *P. proskaueri* is morphologically distinct from all other tuber-producing hornworts common to western North America (Table 1). It is also decidedly different from *Phaeoceros tuberosus* (Taylor) Prosk. (= *Anthoceros tuberosus* Taylor), a species distributed in western Australia, and *P. diadematus* Hässel, a newly described taxon from Chile (Hässel de Menéndez, 2006). The lectotype collection of *A. tuberosus* is dioicous and dimorphic, with male thalli up to 1.5 mm wide and female thalli to 2.8 mm wide. Small tubers, less than 0.4 mm in diameter, are formed, sometimes abundantly, at the apices of elongate lateral branches. They are never ventral in origin as described by Ashworth (1896), and except for their much smaller size, resemble the tubers of *P. hallii*. The few male plants in the collection bear groups of empty antheridial chambers near the tips of elongate thallus lobes. Sporophytes are 1.0–1.3 cm long, yellow to brownish orange, and possess stomates in the capsule wall. Capsule dehiscence occurs along 2 sutures that are straight to slightly twisted. The spores are yellow to yellow orange, even at the tips of completely opened capsules, and are covered with a dispersed network of vermiculate thickenings. The proximal surface bears a prominent trilete ridge and up to 30 dispersed papillae on each facet; the distal surface bears numerous conical verrucae that are 3–4 μm high (Fig. 4.7). Pseudoelaters are pale yellow, mostly 3- or 4-celled and often branched. This assemblage of characters suggests that the affinities of *P. tuberosus* lie with *P. laevis*, not the *P. hallii* complex.

According to Hässel de Menéndez (2006), *P. diadematus* is also a dioicous taxon. Males are described as having 4 or 5 antheridia per antheridial cavity (Hässel de Menéndez, 2006), but we were unable to find antheridial chambers in the specimens we examined, including the type. In two of the collections, we did find two size classes of thalli, suggesting that the sexes are dimorphic. Thalli with immature sporophytes range from 1.5 to 1.9 mm in width, whereas those in the same collection without sporophytes are 0.7–0.8 mm in width. Collections bearing these smaller thalli were made in late August (*Villigrán 1017*) and mid-September (*Villigrán 1029*), whereas the holotype (*Villigrán 1115*), which bears mature, dehisced capsules, was collected in early November. The maximum width of sporophyte-bearing thalli in this latter collection is 3.0 mm. Thalli are very thin, 3 to 4 cells thick at the thallus midline, tapering to only 1 cell thick within 6–8 cells of the thallus margin. We have not studied live material, but Hässel de Menéndez (2006) reports that the chloroplasts lack pyrenoids and occur singly in the cells. Rhizoids are granulate, sometimes adherent in plate-like masses, and 12–19 μm in diameter; they frequently have broadly flared, funnel-shaped tips. In the *P. hallii* complex, the rhizoids are always bluntly rounded at their tips, never flared, but we have observed similar flared rhizoids in species of *Phaeomegaceros*.

Tubers are formed ventrally from submarginal cells near the dichotomizing apical notch of the main thallus, at other points along the thallus border, or at the tips of elongate lateral branches (Fig. 5.1). The thallus margin extends dorsally beyond the tuber by 2 or 3 cells. Tubers are elongate, to 2.0 mm long and 1.0 mm wide, dark brown at maturity, and covered with rhizoids ventrally. They are never stalked or descendent.

Capsules in the holotype are dark brown, up to 7.0 cm long and dehiscent for about two-thirds their length (Fig. 1.1). They are split along two

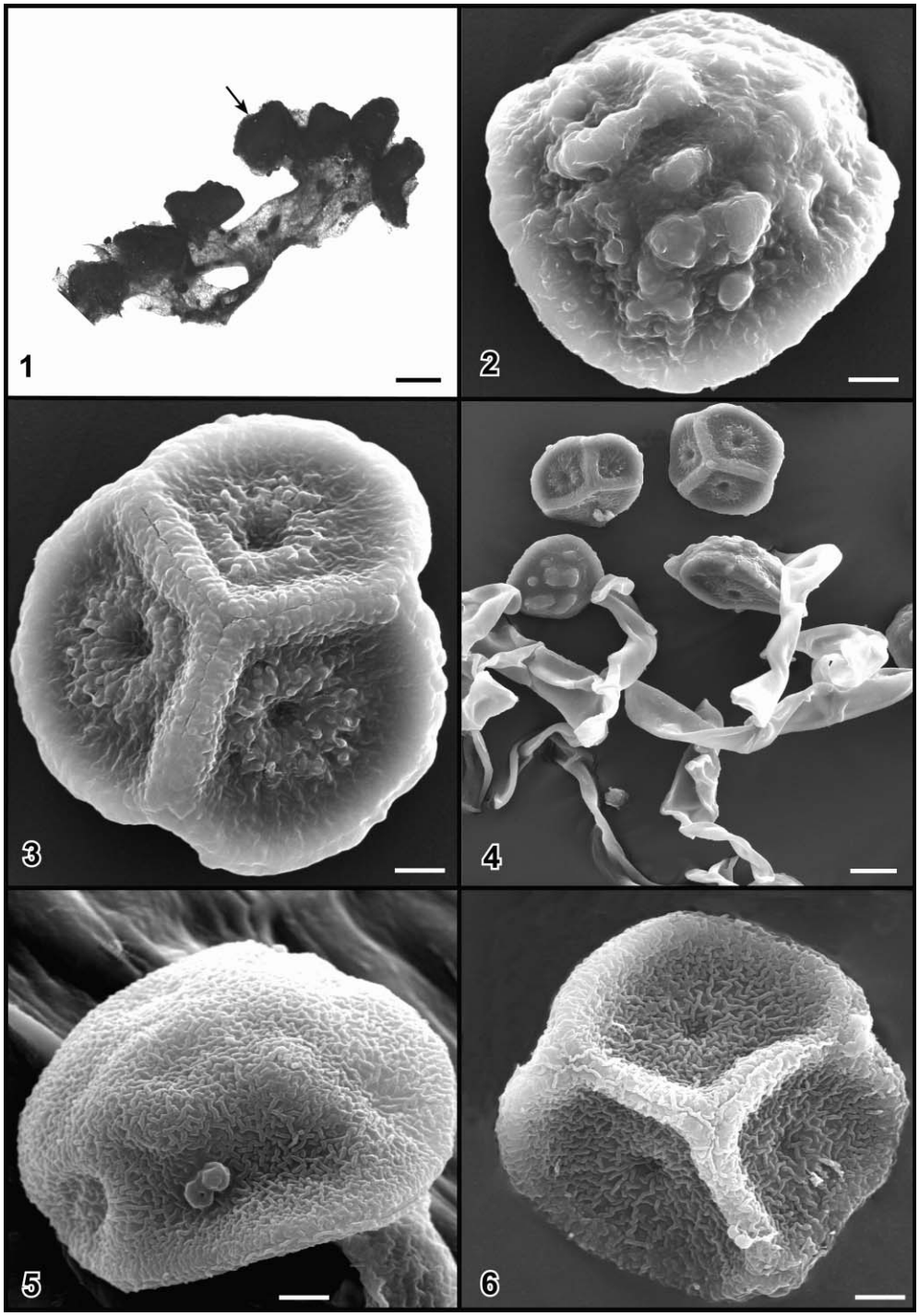


FIG. 5. *Paraphymatoceros diadematus* Hässel (**1**–**4**) and *Paraphymatoceros coriaceus* (Steph.) Stotler (**5**, **6**). **1.** Tuber-bearing thallus, ventral view; at the arrow note the small rim of marginal cells extending beyond the tuber (1-mm scale). **2, 3.** SEM of distal (**2**) and proximal (**3**) faces of mature spores from a dehisced capsule (6-μm scale in **2**, 5-μm scale in **3**). **4.** SEM of mature spores and pseudoelaters from a dehisced capsule (20-μm scale). **5, 6.** SEM of distal (**5**) and proximal (**6**) faces of mature spores (5-μm scale). [**1**–**4** from the holotype; **5**, **6** from *von Konrat s.n.*]

lines, and the valves are helically twisted and apically free. The capsule wall is 5 to 7 cell layers thick, consisting of an epidermis with thickened, pigmented outer tangential and radial walls, 3 to 5 layers of plastid-containing assimilatory cells, and an inner "endodermis" with thickened radial and inner tangential walls. Cells of both the epidermis and "endodermis" are rectangular in transverse section, whereas the assimilatory cells are isodiametric to quadrate. Stomates are widely scattered throughout the epidermis. The columella consists of 16 columns of thick-walled cells and protrudes as a rigid brown strand in dehiscing capsules.

Spores are mostly yellow to tawny but can be medium blackish brown near the capsule tip, and 40–50 μm in equatorial diameter. The distal surface is roughened with small papillae that gradate to short vermiculate thickenings and 2 to 12 large rounded protuberances or ridges, 5–6 μm in both height and diameter (Fig. 5.2). The proximal face is roughened with coarse, vermiculate thickenings and bears a prominent trilete ridge; each facet bears a central depression that averages 5.0 μm in diameter (Fig. 5.3). A ring of somewhat higher, coarser thickenings radiate out from the depressions, gradating into the finer vermiculate strands near the edges of the facet. Hässel de Menéndez (2006) describes the proximal facets as having "a crown of baculae surrounding a depressed area," but in SEM micrographs, the "crown" appears to be made up of coarser, fused extensions of the vermiculate background, not rod-shaped baculae. Pseudoelaters are blackish brown, frequently branched, 3 or 4 cells long in the median part of the capsule, with individual cells 9.0–10.0 μm wide and 80–100 μm long. They typically fragment into shorter, 1- or 2-celled structures in the zone of dehiscence.

Hässel de Menéndez (2006: 210) included *P. hallii* in *Paraphymatoceros*, noting that it produces tubers and referencing an illustration of spores from Hässel de Menéndez, 1989: fig. 13. However, the spore proximal face in *P. diadematus*, which is the type species of the genus, is very different from that of *P. hallii*, as well as *P. pearsonii* and *P. proskaueri* (compare Figs. 5 and 6). There is, in fact, more resemblance to the proximal face architecture found in some species of *Phaeomegaceros* (Fig. 5.5, 5.6). For example, in *Phaeomegaceros coriaceus* (Steph.) Duff et al., each facet of the proximal face is covered with vermiculate thickenings and bears a central depression that is ringed by radiating thickenings; in contrast to *Paraphymatoceros*, however, the distal spore face is also densely vermiculate and is marked with a few shallow depressions and 2 to 4 protuberant ridges (see also Campbell, 1982: fig. 4). Spores with proximal face depressions, or fovea, also occur in *Phaeoceros foveatus* J. Haseg., a southeast Asian species that is closely related to *Phaeomegaceros fimbriatus* (Gottsche) Duff et al. (Hasegawa, 2001: Fig. 2; Villarreal & Renzaglia, 2006), as well as *Phaeoceros himalayensis* (Kash.) Prosk. (Asthana & Srivastava, 1991: pl. 48) and *Notothylas dissecta* Steph. (Hässel de Menéndez, 1976: fig. 1). Furthermore, in the latter two species, the spores are initially yellow but become brown before natural dehiscence (Proskauer, 1967; Hässel de Menéndez, 1976).

In addition to producing foveate spores with an underlying matrix of vermiculate thickenings, other characters supporting the relationship between *Paraphymatoceros* and *Phaeomegaceros* include the following: the absence of a pyrenoid in the chloroplasts, rhizoids with flared tips, a dioicous sexual condition, a capsule wall with epidermal stomates and 5 or more layers of assimilatory cells, branched pseudoelaters, and two apically free, helically twisted capsule valves. According to Duff et al. (2007), all species of *Phaeomegaceros* are monandrous, but Campbell and Hasegawa (1993) report 4 antheridia per cavity in *Phaeomegaceros hirticalyx* (Steph.) Duff et al., as is also the case in *Paraphymatoceros* (Hässel de Menéndez, 2006). Morphological evidence suggests that *P. diadematus* is remote from the *P. hallii* group and supports its placement with the currently recognized species of *Phaeomegaceros*.

Molecular Evidence and Phylogeny

In both maximum parsimony (MP) and maximum likelihood (ML) analyses of *rbc*L sequence data, *P. proskaueri*, *P. hallii*, and *P. pearsonii* form a well-supported lineage, designated as the *P. pearsonii* clade, within a *Phaeoceros* + *Notothylas* clade (Fig. 7), with BS values of 99% (MP) and 96% (ML). Within the *P. pearsonii* clade, *P. proskaueri* is resolved as sister to *P. hallii*, with *P. pearsonii* sister to the *P. proskaueri* + *P. hallii* clade. The relationship of the *P. pearsonii* clade to the *Notothylas* clade or the *Phaeoceros carolinianus* (Michx.) Prosk. + *P. laevis* clade is unresolved in MP. In ML, the *Notothylas* clade is

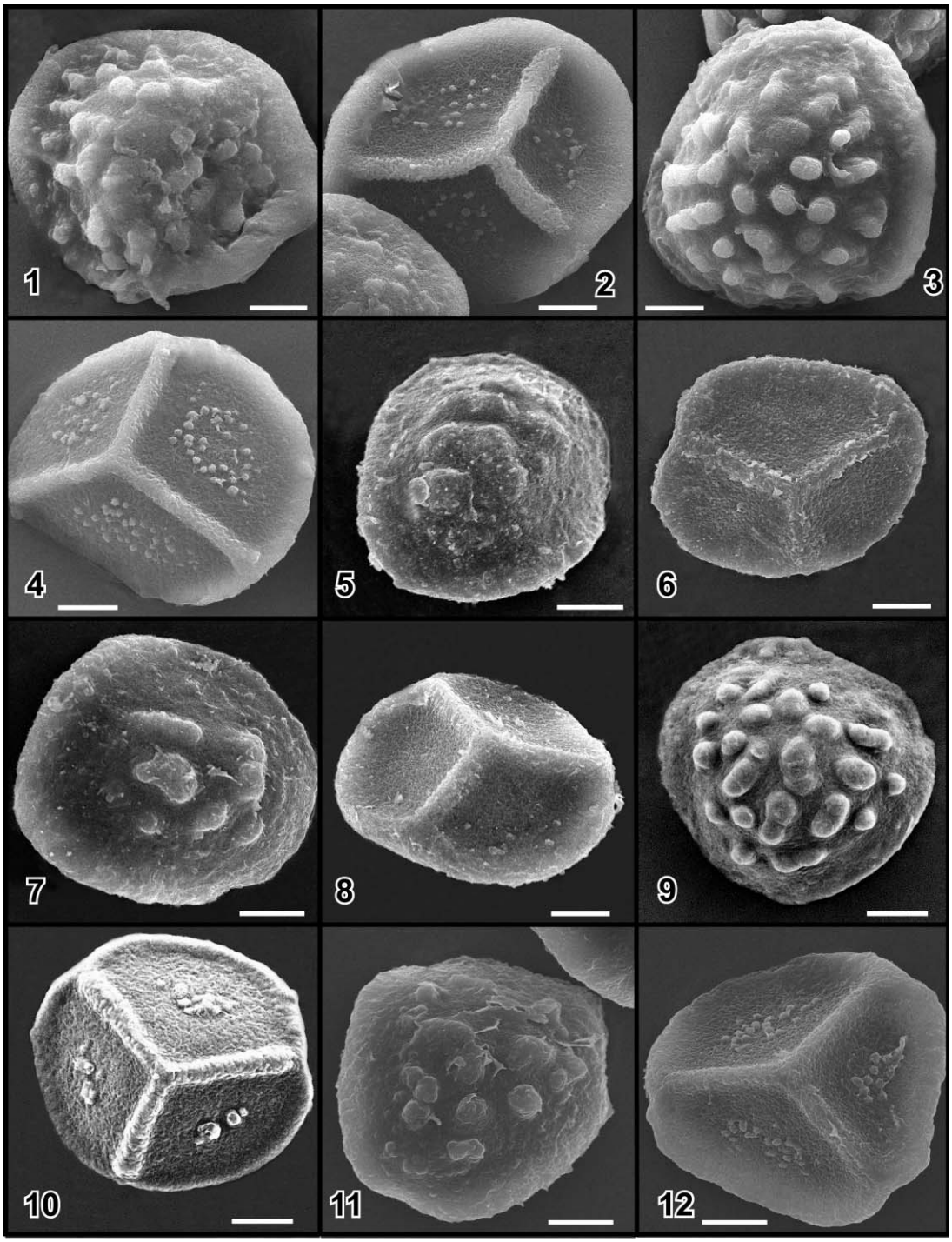


FIG. 6. SEM images of spore wall architecture in type specimens of the *Phaeoceros hallii* species complex (**1**–**10**) and spore variation in *Phaeoceros pearsonii* (M. Howe) Prosk. (**11**–**12**). **1, 2.** Lectotype of *Anthoceros hallii* Austin [MANCH]. **3, 4.** Lectotype of *Anthoceros sulcatus* Austin [MANCH]. **5, 6.** Isotype of *Anthoceros pearsonii* M. Howe [UC]. **7, 8.** Isotype of *Anthoceros bolanderi* Steph. [UC]. **9, 10.** Holotype of *Phaeoceros proskaueri* Stotler, Crand.-Stotl. & W. T. Doyle [UC]. **11, 12.** Atypical spores that are blackish brown and have several proximal facet papillae (from *Wagner m1387*) (all scales are 10 μm).

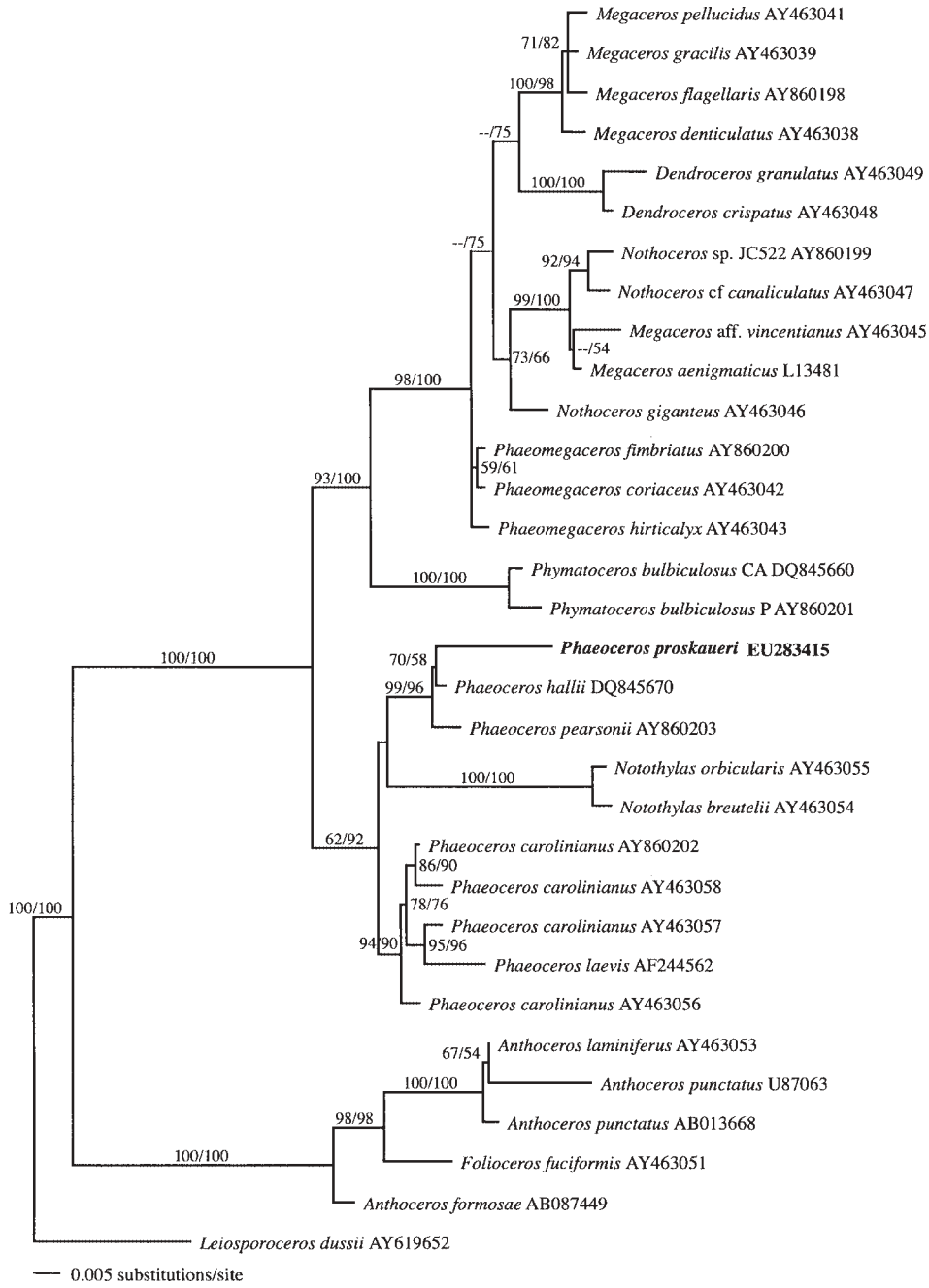


FIG. 7. Likelihood phylogram for hornworts based on chloroplast rbcL sequence data, generated using the general time reversible model. The first number above or to the right of branches is the MP bootstrap value and the second represents likelihood bootstrap support. Slender branches were not recovered by parsimony analyses.

recovered as sister to the *P. pearsonii* clade, but with less than 50% BS support. These results are congruent with the MP and Bayesian reconstructions of Duff et al. (2007).

As can be seen from the branch length on the phylogram (Fig. 7), the level of genetic divergence between *P. proskaueri* and *P. hallii* supports the recognition of *P. proskaueri* as a distinct species of the *P. pearsonii* clade, as predicted by our morphological studies. The defining characters of the clade include perennation through apical or marginal tubers; thalli more or less strap-shaped

and not forming rosettes; capsule dehiscence along a single longitudinal slit, usually without helical twisting of the wall; spores yellow when immature, sometimes becoming darkened with secondary deposits late in development; spore walls with a vermiculate network of thickenings, overlaid distally by rounded protuberances or ridges and proximally often with a central group of papillae. *Phaeoceros hallii* has plastids with a distinct pyrenoid-like central region and 2–4 antheridia in each antheridial cavity, whereas *P. proskaueri* and *P. pearsonii* lack pyrenoids in their plastids and are monandrous. Obviously, neither the lack of a pyrenoid nor monandry are necessarily lineage-defining characters, being expressed in representatives of numerous lineages, including *Phymatoceros* (Crandall-Stotler et al., 2006), *Paraphymatoceros* (Hässel de Menéndez, 2006), *Phaeomegaceros* (Villarreal & Renzaglia, 2006), and other elements of the subclass Dendrocerotidae Duff et al. (Duff et al., 2007).

Morphological data support the notion of a relationship between the *P. pearsonii* clade and some species of *Notothylas*, as first discussed by Austin (1875). The capsules of *P. hallii* resemble those of *Notothylas* in being short, synchronous in sporogenesis and often indehiscent, but differ in having stomates in the capsule wall and an erect rather than horizontal orientation. Spore wall architecture in *N. orbicularis* is very similar to that of *P. pearsonii* (Schuster, 1992: fig. 1055) and the spores of *N. breutelii* (Gottsche) Gottsche become fuscous at maturity. Worldwide, *Notothylas* is morphologically heterogeneous and it is likely that with further sampling, the genus will be resolved as paraphyletic. Nonetheless, the clade containing *N. orbicularis* is either nested in or sister to the genus *Phaeoceros* (Duff et al., 2007), and the affinities of the *P. pearsonii* clade are with it and the genus *Phaeoceros*.

Taxonomy

NOTOTHYLADACEAE (Milde) Prosk., Phytomorphology 10: 10. 1960.

PHAEOCEROTOIDEAE Hässel, J. Hattori Bot. Lab. 64: 81. 1988.

PHAEOCEROS Prosk., Bull. Torrey Bot. Cl. 78: 346. 1951.

Phaeoceros hallii (Austin) Prosk., Bull. Torrey Bot. Club 78: 347. 1951.

Anthoceros hallii Austin, Bull. Torrey Bot. Club 6: 26. 1875. Lectotype (designated by M. Howe, Bull. Torrey Bot. Club 25: 11. 1898): USA, Oregon [Marion County], 26, Springy Places, Silverton, *Hall* [*26*] (MANCH—EM74235/21217!; isolectotypes MANCH (2!) EM74234/21216, EM74241/21224; Paratype: USA, Oregon [Marion County], Dripping rocks, Salem, *Hall* [*35*] (MANCH (2!) EM74232/21214, EM74237/21219).

Anthoceros sulcatus Austin, Bull. Torrey Bot. Club 6: 27. 1875. Lectotype, designated here: USA, Oregon [Marion County], 25, moist earth, Salem, Oregon, *Hall* [*25*] (MANCH—EM74240/21223!; isolectotypes MANCH (4!) EM74233/21215, EM74236/21218, EM154642/21221, EM74239/21222.

Notothylas hallii Austin ms., Bull. Torrey Bot. Club 6: 27. 1875.

Paraphymatoceros hallii (Austin) Hässel, Phytologia 88: 209. 2006. syn. nov.

Phaeoceros pearsonii (M. Howe) Prosk., Bull. Torrey Bot. Club 78: 347. 1951.

Anthoceros pearsonii M. Howe, Bull. Torrey Bot. Club 25: 8 + pl. 322–323. 1898. "*pearsoni*." Holotype: USA, California, Marin County, Mill Valley, 7 May 1892, Howe 16 (NY—non vidi); isotype: (UC!).

Anthoceros bolanderi Steph., Sp. Hepat. 5: 975. 1916. Lectotype (designated by G. Hässel de Menéndez, Candollea 44: 731. 1989): USA, California, Mendocino City, *Bolander* (ex Herb. Gottsche) (G—non vidi; isotype: UC!)

Phaeoceros proskaueri Stotler, Crand.-Stotl. & W. T. Doyle, sp. nov.

Plantae thalloides, dioicae, dimorphae; thallus parvus, crassus, angustus, linearis, ramosus irregulariter, tuberibus numerosis subsphaeroideis in ramis brevissimis lateralibus; cellulae chloroplasto uno; chloroplasti sine pyrenoidibus; androecium monandrum; capsula stomatophora, columella brunnea, rigida; sporae brunneae vel fuscae ubi maturis; pseudoelateres pallidi-brunnei, 2- vel 3-cellularis.

Plants prostrate, gregarious, forming mats; thallus segments linear, irregularly branched, with the branches subparallel to somewhat spreading and linear-oblong to narrowly lingulate, with a broad, thick central midrib that bears rhizoids ventrally, with the margins irregular and slightly upturned, constricted to 2 cells; plastids 1 per

cell, spheroidal to spindle-shaped, lacking a pyrenoid; rhizoids hyaline, abundant, mostly granulate, nongranulate (smooth) near the apex or on developing tubers, never adherent or fascicled; tubers abundant, at the apices of very short lateral branches, pale green, becoming brown at maturity, subspheroidal, less than 1 mm in diameter, on female plants rarely strongly flattened; plants dioicous, dimorphic; male thalli 0.3–0.8 mm wide; antheridial chambers near the base of the male thalli, with 1 antheridium per chamber; female thalli with sporophytes 1.5–2.0 mm wide; involucres short, 0.6–1.5 mm, narrowly campanulate, with the mouth flared; capsules 0.7–1.75 cm long (usually less than 1 cm), with the columella persistent, dehiscing by a single longitudinal slit along a suture line of yellow to orange cells; spores yellow to tawny when immature, becoming deep brown to fuscous when mature, 36–49 μm in diameter; spore proximal face trilete, with a small central cluster of 4 to 10 papillae on each facet; spore distal face with 13–30 rounded mammillae or elongate, rounded ridges, with a cingulum or wing; pseudoelaters brown, 2 or 3 cells long, fragmenting into 1(2)-celled structures near the capsule apex, each cell 12–18 μm wide, up to 40 μm long.

HOLOTYPE: **CALIFORNIA; Monterey County**. Fort Ord Public Lands–Bureau of Land Management, Barloy Canyon Road, nr Native Plant Reserve and Ft. Ord sign, hills primarily with coastal scrub chaparral, on north-facing slope on silty soil of roadcut, N 36°37′99″, W 121°44′14″, ca. 133 m.s.m., 14 April 2005, *Doyle 11350* (UC; Isotypes: ABSH, BA, BM, FH, G, MANCH, NY, W).

This species is named to honor Dr. Johannes Max Proskauer, who was the world's leading authority on hornworts at the time of his death in 1970. We are pleased that the species to bear his name is an element of *Phaeoceros*, a genus that he erected as a consequence of his doctoral research on the morphology of *Anthoceros* (Proskauer 1951).

Phaeoceros tuberosus (Taylor) Prosk., J. Indian Bot. Soc. 42A: 185. 1964.

Anthoceros tuberosus Taylor, Lond. J. Bot. 5: 412. 1846. Lectotype, designated here: Australia, "*Anthoceros tuberosus* MSS. T[homas]. T[aylor]., Swan River. Mr. James Drummond 1843" (FH!). [Although Proskauer annotated this specimen as the "Lectotype" he never published that choice.]

DENDROCEROTACEAE (Milde) Hässel, J. Hattori Bot. Lab. 64: 82. 1988.

PHAEOMEGACEROTOIDEAE Duff, J. Villarreal, Cargill & Renzaglia, Bryologist 110: 241. 2007.

PARAPHYMATOCEROS Hässel, Phytologia 88: 208. 2006. [*PHAEOMEGACEROS* Duff, J. Villarreal, Cargill & Renzaglia, Bryologist 110: 241. 2007. syn. nov.]

Paraphymatoceros diadematus Hässel, Phytologia 88: 209. 2006. (Holotype: CHILE, R V [Región de Valparaíso], Cachagua, Qda. Aguas Claras, En paredón húmedo junto al sendero, 05-Nov-05, Col. *C. Villagrán 1115*, Det. Hässel de Menéndez 2006, Holotipo (SGO: 153453!); paratypes: R V, Cachagua, Qda. Aguas Claras En paredón húmedo junto al sendero, 18-Sep-05, Col. *C. Villagrán 1029*, Det. Hässel de Menéndez 2006, Topotipo (SGO: 153454!); R V, El Tabo, Quebrada de Córdoba, Ladera de exposición sur. Matorral esclerófilo, 21-Ago-06, Col. *C. Villagrán 1038*, Det. Hässel de Menéndez 2006, (SGO: 153455!). [Hässel de Menéndez (2006) cited Villagrán 1029 and 1038 from Córdoba; this is correct for 1038, but 1029 is from the type locality of Qda. Aguas Claras.]

Hässel de Menéndez (2006) intended to include three species in her genus *Paraphymatoceros*, but she inadvertently transferred *Anthoceros minutus* Mitt., an African species, to the genus *Phymatoceros* Stotler, Crand.-Stotl. & Doyle, not *Paraphymatoceros*. We have not studied that taxon, but her inclusion of *Phaeoceros hallii* in *Paraphymatoceros* is shown here to be misplaced, in that it clearly belongs with *P. pearsonii* and *P. proskaueri*. Although Hässel de Menéndez (2006) placed her new genus into Notothyladaceae subfamily Phaeocerotoideae, our study of *P. diadematus* revealed that it is not only more appropriately aligned with *Phaeomegaceros* but can be regarded as congeneric with that taxon. Because *Paraphymatoceros* has priority, the following new combinations are made.

Paraphymatoceros fimbriatus (Gottsche) Stotler, **comb. nov.** Basionym: *Anthoceros fimbriatus* Gottsche, Ann. Sci. Nat. Bot. sér. 5. 1: 187–188. 1864.

Paraphymatoceros coriaceus (Steph.) Stotler, **comb. nov.** Basionym: *Anthoceros coriaceus* Steph., Sp. Hepat. 5: 991. 1916.

Paraphymatoceros hirticalyx (Steph.) Stotler, **comb. nov.** Basionym: *Aspiromitus hirticalyx* Steph., Sp. Hepat. 5: 966–967. 1916.

Paraphymatoceros skottsbergii (Steph.) Stotler, **comb. nov.** Basionym: *Anthoceros skottsbergii* Steph., Kongl. Svenska Vetenskapsakad. Handl. 46: 90. 1911.

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Appendix

ADDITIONAL SPECIMENS EXAMINED—***Paraphymatoceros coriaceus***. NEW ZEALAND. s.l., (rec'd. 23 March 2006 acc. BJCS 583), *von Konrat s.n.* (ABSH). ***Paraphymatoceros diadematus***. CHILE. R V [Región de Valparaíso], Zapallar, Quebrada El Tigre. Naciente de la quebrada, al Ilegar al Alto del Boldo. 26-Ago-06, Col. *C. Villagrán 1017*, Det. Hässel de Menéndez 2006, (SGO: 153456). ***Phaeoceros hallii***. USA California–EL DORADO COUNTY: Sierra Nevada foothill, Sand Ridge Road ca. 4.7 mi W of jct. with Buck's Bar Road, soil of hillside, elev. ca. 1200 feet, 26 April 1996, *Doyle 8258* (ABSH, UC); Sierra Nevada western slope, Sand Ridge Road at hairpin turn, ca. 1 mi E of jct. with CA Hwy 49, seepage at base of small sloping meadow, with scattered *Quercus* and *Pinus*, metamorphic substrate, shaded by grasses and *Mimulus*, ca. 365 m, N 38°35′78″, W 120°49′98″, 16 May 2005, *Doyle 11363*, 25 May 2005, Doyle *11366* (ABSH, UC); MADERA COUNTY: Sierra Nevada foothill, along CR 274 E of Bass Lake, on thin soil, seepage over granite slab in a small meadow, elev. ca. 3450 feet, 30 April 1996, *Doyle 8276* (ABSH, UC); Sierra Nevada, Oak Grove Road, ca. 4.4 mi W of Madera/Mariposa County line, nr oaks, bases of roadcut, in grass, elev. ca. 700 feet, 9 April 1996, *Doyle 8193*, 30 April 1996, *Doyle 8272*, 20 May 1996, *Doyle 8445* (ABSH); Sierra Nevada, Oak Grove Road ca. 2.3 mi W of Madera/Mariposa County line, soil of roadcut, elev. ca. 800 feet, 30 April 1996, *Doyle 8271* (ABSH); Sierra Nevada, E side of Auberry Road ca. 1.5 mi N of bridge across San Joaquin River, shade of oaks, seepage, depression in hillside, elev. ca. 2400 feet, 30 April 1996, *Doyle 8292* (ABSH); Sierra Nevada, W side of CR 415 ca. 7.2 mi N of Coarsegold on SR 41, scattered oaks, under grass of "mouse runs" nr hillside seepage, elev. ca. 2000 feet, 20 May 1996, *Doyle 8446* (ABSH); MARIPOSA COUNTY: Sierra Nevada foothills, along Oak Grove Road ca. 3.9 mi E of Mariposa/Madera County line, seepage over granite, elev. ca. 1000 feet, 30 April 1996, *Doyle 8269* (ABSH, UC); Sierra Nevada, Oak Grove Road ca. 1.7 mi E of Mariposa/Madera County line, soil of hillside, elev. ca. 900 feet, 30 April 1996, *Doyle 8270* (ABSH); MODOC COUNTY: Modoc National Forest, Yellow Jacket Springs Road ca. 0.8 mi W off National Forest Service Road 84, W off CA 299 nr Canby Bridge, bank of ephemeral creek, shade of pines, 19 July 1995, *Doyle 7486* (ABSH); Modoc National Forest, Yellow Jacket Springs ca. 0.8 mi W off National Forest Service Road 84, volcanic substrate, elev. ca. 4500 feet, 3 August 1996, *Doyle 8649* (ABSH, UC); Modoc National Forest, Yellow Jacket Springs Road, W off National Forest Service Road 84, N off CA 299 nr Canby Bridge, volcanic substrate, on soil bank and rock in bed of ephemeral creek in a pine forest, elev. ca. 1365 m, 26 July 2005, *Doyle 11390* (ABSH, UC); NEVADA COUNTY: Sierra Nevada, Garden Bar Road ca. 2.1 mi SW of jct. with Wolf Road, soil of roadbank, elev. ca. 800 feet, 27 April 1996, *Doyle 8319* (ABSH, UC); TRINITY COUNTY: Klamath Range, NE side of Trinity Lake, Eastside Road ca. 4.8 mi E of jct. with SR 3, drainage through meadow in oak woodland, elev. ca. 3000 feet, 12 June 1996, *Doyle 8545* (ABSH, UC). ***Phaeoceros pearsonii***. USA CALIFORNIA–ALAMEDA COUNTY: Diablo Range, Calaveras Road, S off Interstate Hwy 680, Sunol Regional Wilderness, hillside seepage on N side of creek, ca. 228 m.s.m., 18 May 1983, *Doyle 3139* (UC); AMADOR COUNTY: Western foothills of Sierra Nevada, Clinton Road, ca. 0.8 mi E of jct. with CA Hwy 49, nr Jackson, mixed *Quercus* and *Arbutus*, soil of hillside on S side of road, ca. 380 m.s.m., 21 May 1996, *Doyle 8449* (UC); BUTTE COUNTY: Sierra Nevada, NE of Oroville, North Table Mountain, scattered *Quercus*, volcanic plateau, on soil of shaded drainage, elev. ca. 363 m, 4 May 1999, *Doyle 9706* (UC); CALAVERAS COUNTY: Western foothills of Sierra Nevada, NE

of Altaville, Gogtown Road ca. 0.3 mi W of jct. with Esmaralda Road, soil of hillside, with grass under *Quercus*, 26 April 1996, *Doyle 8248* (UC); CONTRA COSTA COUNTY: Central Coast Region, Morgan Territory Road, ca. 7.9 mi N of jct. with North Livermore Road, mixed *Quercus*, *Lithocarpus*, and *Umbellularia*, soil of roadcut, elev. ca. 150 m, 11 April, *Doyle 8938* (UC); EL DORADO COUNTY: western slope of Sierra Nevada, Sandridge Road, nr 3.76-mi marker, E off CA 49, nr scattered *Quercus* and *Arbutus*, on soil on exposed vertical slope of hillside nr creek, elev. ca. 575 m, 28 May 1996, *Doyle 8463* (UC); HUMBOLDT COUNTY: North Coast Ranges, CA Hwy 36 nr 36.39 mi marker, small creek and road cut on S side of road, mixed *Lithocarpus*, *Pseudotsuga*, *Umbellularia*, and *Acer*, shaded, slow-drying soil of creek bank and road cut, N 40°27′07″, W 123°39′09″, ca. 810 m.s.m., 1 June 2005, *Doyle 11386* (UC); MARIN COUNTY: North Coast Region, Bolinas–Fairfax Road nr Alpine Dam, nr *Sequoia* and *Quercus*, on soil nr base of road cut, ca. 150 m.s.m., 14 May 1996, *Doyle 8423* (UC); North Coast Region, Sir Francis Drake Boulevard, Samuel P. Taylor State Park, *Sequoia*, *Lithocarpus*, *Acer*, and *Alnus*, soil on shaded bank of Lagunitas (Papermill) Creek, elev. ca. 45 m, 3 June 2003, *Doyle 10536* (UC); MENDOCINO COUNTY: North Coast Region, rest area on W side of US 101, nr 101 MEN 58.6 mi marker, canyon W of rest area, mixed *Lithocarpus*, *Quercus*, *Acer*, and *Pseudotsuga*, on soil, steep side of canyon above creek, elev. ca. 350 m, 14 September 1995, *Doyle 7776* (UC); North Coast Region, CA Hwy 1, ca. 1.1 mi W of Leggett, *Sequoia* forest, on soil of road cut, elev. ca. 335 m, 30 April 1997, *Doyle 8974* (UC); North Coast Ranges, Low Gap Road ca. 7.4 miles W of Ukiah, oak, Douglas fir, maple, blackberry, and poison oak, soil of shaded, north-facing roadside drainage, N 39°09′58″, W 123°19′86″, ca. 730 m.s.m., 31 May 2005, *Doyle 11377* (UC); North Coast Ranges, Low Gap Road, ca. 5.2 mi W of Ukiah, mixed *Lithocarpus*, *Pseudotsuga*, and *Acer*, small creek and spring on S side of road, shaded, slow-drying soil, N 39°09′58″, W 123°19′86″, ca. 760 m.s.m., 31 May 2005, *Doyle 11378* (UC); MONTEREY COUNTY: Santa Lucia Range, Los Padres Natl. Forest, Ventana Wilderness, Pine Ridge Trail, Trailhead at Big Sur Station on CA Hwy 1, *Sequoia*, *Acer*, *Lithocarpus*, and *Alnus*, on soil of hillside along trail, R2E T19S S21, ca. 212 m.s.m., 11 May 1994, *Doyle 6418* (UC); PLACER COUNTY: Sierra Nevada, SW of Applegate, Clipper Gap Road, ca. 0.25 mi S of jct. with Pepper Ranch Road, scattered oaks on hillside, shaded drainage along road, on soil, elev. ca. 576 m, 19 May 2002, *Doyle 9963* (UC); North Fork of the Middle Fork of American River at Mosquito Ridge Road, shaded roadbank in *Pinus ponderosa–Quercus chrysolepis–Pseudotsuga* forest, N 39°01.5′, W 120°43′, elev. 1300 feet, 2 May 1992, *A.T.&T. E. Whittemore 3985* (ABSH); SAN BENITO COUNTY: Central Coast Region, Diablo Range, NE of Hollister, Lone Tree Road, ca. 8.3 mi E of jct. with Fairview Road, creek bank on N side of Road, on soil, 10 May 1995, *Doyle 7391* (UC); Gabilan Range, Pinnacles Natl. Monument, Moses Spring Trail, between trail markers 8 and 9, shade of toyon and oak, damp hillside along trail, N 36°28′53″, W 121°11′21″, ca. 500 m.s.m., 28 March 2005, *Doyle 11325*, 18 September 2005, *Doyle 11415*, 27 December 2005 *Doyle 11420*, 9 January 2006, *Doyle 11424*, 20 January 2006, *Doyle 11429* (UC); SAN LUIS OBISPO COUNTY: Santa Lucia Range, Santa Rita Road between Templeton and Cayucus, E side of summit, *Quercus*, *Pinus*, and chaparral, soil of shaded road cut, ca. 273 m.s.m., 19 April 1993, *Doyle 5726* (UC); North Coast Ranges, Montana de Oro State Park, Coon Creek Trail, shade of *Salix*, *Quercus*, and *Umbellularia*, on soil of creek bank, R10E T31S, elev. ca. 45 m, 28 April 1998, *Doyle 9481* (UC); Santa Lucia Range, Santa Rita Road ca. 1.7 miles E of jct. with Old Creek Road, mixed *Quercus*, *Lithocarpus*, *Acer*, *Umbellularia*, and *Salix*, damp, slow-to-dry soil of road cut under *Adiantum*, N 35°28′65″, W 129°50′10″, ca. 120 m.s.m., 12 April 2005, *Doyle 11346* (UC); SAN MATEO COUNTY: Central Coast Region, Santa Cruz Mountains, San Pedro Valley County Park, Hazelnut Trail, sedimentary substrate, scattered chaparral and *Quercus*, on soil, hillside along trail, R6W T4S S24, elev. ca. 105 m, 1 May 2003, *Doyle 10380* (UC); SANTA BARBARA COUNTY: South Coast Region, Transverse Ranges, Santa Ynez Mountains, S side of road between US Hwy 101 and Nojoqui Falls County Park, oaks and shrubs, shaded soil of hillside, R31W T5N S12, 6 April 1995, *Doyle 7174* (UC); South Coast Region, Purisima Hills, La Purisima Mission State Historic Park, shade of *Salix*, on soil, bank of Purisima Creek, elev. ca. 105 m., 27 April 1998, *Doyle 9473* (UC); SANTA CRUZ COUNTY: Central Coast Region, Santa Cruz Mountains, Branciforte Drive, just E of Goss Avenue, shaded, base of vertical sandstone outcrop on S side of road, on spoil, elev. ca. 42 m, R1W T11S S7, 9 May 1994, *Doyle 6380* (UC);

Central Coast Region, Santa Cruz Mountains, Fall Creek Unit of Henry Cowell Redwoods State Park, *Sequoia*, *Lithocarpus*, *Acer*, and *Umbellularia*, North Fork Trail, nr jct. with South Fork Trail, on soil of N facing cliff face along trail, elev. ca. 180 m, R2W T10S S16, 3 May 1997, *Doyle 8941* (UC); Central Coast Region, Santa Cruz Mountains, Mt. Madonna County Park, CA Hwy 152, nr the 0.87 mi marker, nr small creek on N side of road, *Sequoia*, *Pseudotsuga*, *Lithocarpus*, and *Umbellularia*, on soil, seepage on an old logging road, elev. ca. 303 m, 19 May 2003, *Doyle 10465* (UC); Santa Cruz Mts., Nisene Marks State Park, along trail between park entrance kiosk and Aptos Creek, on soil along trail in grass, ca. 25 m.s.m., 10 January 2006, *Doyle 11426*, 20 January 2006, *Doyle 11428* (UC); SHASTA COUNTY: Klamath Ranges, Trinity Mountains, along Placer Road, SW of Redding, open, drying creek on N side of road, on soil of creek bank with grass and moss, elev. ca. 210 m, 12 May 1997, *Doyle 9117* (UC); SISKIYOU COUNTY: Klamath Ranges, Klamath National Forest, Forest Road 37N24, off Callahan-Cecilville Road, S of South Fork Campground, on soil, seepage into a roadside drainage ditch, elev. ca. 820 m, R11W T37N S10, 9 June 1993, *Doyle 5934* (UC); SONOMA COUNTY: North Coast Ranges, Cazadero Hwy, ca. 1.1 mi NW of Cazadero, mixed *Pseudotsuga*, *Sequoia*, *Acer*, and *Umbellularia*, shaded soil of roadcut, S side of road, elev. ca. 182 m, 3 May 2004, *Doyle 10941* (UC); TRINITY COUNTY: Klamath Ranges, South Fork of Trinity River Road, ca. 3.8 mi S of jct. with CA Hwy 299, shaded soil of road cut, R5E T6N S25, ca. 242 m.s.m., 13 June 1996, *Doyle 8555* (UC); TULARE COUNTY: Sierra Nevada, Sequoia Natl. Forest, Kaweah River drainage, North Fork Road S from Three Rivers, on soil under shrubs at base of road cut, ca. 725 m.s.m., 6 May 1993, *Doyle 5807* (UC); YUBA COUNTY: Sierra Nevada, Tahoe National Forest, Handicap Trail of Independence-Yuba River Trail, off CA Hwy 49, NE of Nevada City, *Pseudotsuga*, *Quercus*, *Calocedrus*, *Pinus*, and *Lithocarpus* forest, on soil, shaded hillside along trail, elev. ca. 425 m, R8E T17N S33, 16 June 1998, *Doyle 9593* (UC); OREGON–LANE COUNTY: Trail along North Fork Middle Fork Willamette River, ca. 5 mi from Westfir Trailhead, ca. 30 mi SE of Eugene, on vertical soil bank that drips in the Spring, ca. 326.8 m.s.m., 19 July 2007, *Wagner m2269* (ABSH); LINN COUNTY, S. Santiam River, Hwy 20, Milepost 50, 29 June 2004, *Wagner m1387* (ABSH).

Phaeoceros proskaueri. USA CALIFORNIA–EL DORADO COUNTY: Western foothills of Sierra Nevada, Rattlesnake Bar Road, ca. 1.6 mi W of jct. with CA Hwy 49 nr Pilot Hill, easily disturbed soil of hillside nr chaparral and oak, ca. 305 m. s.m., 28 May 1996, *Doyle 8466* (UC); Sandridge Road ca. 0.2 mi E of jct. with CA Hwy 49, open north-facing, easily disturbed soil of hillside, ca. 260 m.s.m., 28 May 1996, *Doyle 8454* (UC); Sandridge Road, ca. 7.8 miles W of jct. with Buck's Bar Road, easily disturbed soil of road cut, nr oak, ca. 305 m.s.m., 26 April 1996. *Doyle 8262* (UC); Western foothills of Sierra Nevada, Sandridge Road nr mile marker 3.67, soil, NW facing road cut, partial shade of *Umbellularia*, *Pinus*, and *Quercus*, N 38°35′95″, W 120°47′50″, ca. 440 m.s.m., 25 May 2005, *Doyle 11368* (ABSH, UC); MARIPOSA COUNTY: Sierra Nevada, western slope, Old Toll Road, ca. 0.65 mi W of CA Hwy 49 jct., chaparral-covered hillslope on N side of road, metamorphic substrate, on soil in shade of shrubs, on remnants of abandoned dirt road, ca. 600 m.s.m., N 37°30′58″, W 120°03′41″, 21 April 2005, *Doyle 11357* (ABSH, UC); Sierra Nevada western slope**,** W of Mt. Bullion, N side of Old Toll Road, ca. 0.65 mi. W of CA Hwy 49 jct., partial shade of chaparral, remnant of old dirt road, soil of roadbed, ca. 600 m.s.m., N 37°30′58.8″, W 120°03′46″, 10 January 2006, *Doyle 11425* (ABSH, UC); Western slope of Sierra Nevada, Old Toll Road, ca. 0.4 mi W of jct. with CA Hwy 49 at Mt. Bullion, S side of road, open areas in chaparral, easily disturbed soil of hillside, ca. 605 m.s.m., 30 April 1996, *Doyle 8267* (UC); MONTEREY COUNTY: Santa Lucia Range, nr gate to old Graniterock Quarry, 22 May 1995, Doyle 7402 (ABSH); Fort Ord Public Lands, Bureau of Land Management, small canyon that crosses Crescent Bluff Road, ca. 0.3 mi S of junction with Barloy Canyon Road, open areas in chaparral and oaks, sandy soil, elev. ca. 225 ft., 10 April 2005, *Doyle 11339* (ABSH); Fort Ord Public Lands-Bureau of Land Management, Barloy Canyon Road, ca. 1.1 mi W of Crescent Bluff Road jct., nr Fort Ord sign, exposed NE facing slope, on fine-grained soil, N 36°37′99″, W 121°44′14″, ca. 125 m.s.m., 18 April 2005, *Doyle 11355* (ABSH, UC), 29 December 2006, *Doyle 11421* (ABSH), 5 January 2006, *Doyle 11422* (ABSH), 19 January 2006, *Doyle 11430* (ABSH); Fort Ord Public Lands-Bureau of Land Management, Crescent Bluff Road, ca. 0.5 mi south of jct. with Barloy Canyon Road, hillside with chaparral and scattered oak, soil, openings in chaparral, N 36°38′30″, W 120°38′30″,

ca. 95 m.s.m., 10 April 1996, *Doyle 8195* (UC); Fort Ord Public Lands–Bureau of Land Management, Eucalyptus Road, ca. 1.8 mi E of jct. with Parker Flat Road, oak woodland, soil of open areas on hillside, ca. 115 m.s.m., 15 April 1998, *Doyle 9453* (UC); Fort Ord Public Lands-Bureau of Land Management, Barloy Canyon Road, ca. 1.1 miles W of jct. with Crescent Bluff Road, chaparral covered hillside, soil of open, sloping drainage on N side of road, N36°38′1″,W 121°44′16″, ca. 125 m.s.m., 6 February 2006, *Doyle 11431* (ABSH, UC); Fort Ord Public Lands–Bureau of Land Management, Barloy Canyon Road, nr Native Plant Reserve and Ft. Ord sign, Hills primarily with coastal scrub chaparral, on north-facing slope on silty soil of roadcut, N 36°37′99″, W 121°44′14″, ca. 133 m.s.m., 24 February 2006, *Doyle 11433*, *11434* (ABSH, UC); RIVERSIDE COUNTY: Peninsular Ranges, Santa Ana Mountains, Santa Rosa Plateau Ecological Reserve, soil of drainage on S side of Clinton Keith Road, with grass nr coastal sage scrub, ca. 545 m.s.m., 16 April 1995, *Doyle 7273* (UC); Peninsular Ranges, Santa Ana Mountains, Santa Rosa Plateau Ecological Reserve, Sylvan Meadow Ranch Unit, soil, open areas in coastal sage scrub, ca. 545 m.s.m., 25 April 1995, *Doyle 7268* (ABSH, UC); Peninsular Ranges, Santa Ana Mountains, Santa Rosa Plateau Ecological Reserve, W of old parking lot at main entrance to Reserve, soil, open grassy areas in coastal sage scrub, ca. 540 m.s.m., 26 April 1995, *Doyle 7275* (UC); Santa Ana Mts., Santa Rosa Plateau Ecol. Reserve, Sylvan Meadow Ranch Unit, N side of Clinton Keith Road, open areas nr and under coastal sage scrub, N33°32′40″, W117°16′32″, ca. 545 m.s.m., 4 May 2006, *Doyle 11450* (ABSH, UC); SAN DIEGO COUNTY: Forester Creek, nr jct. of La Cresta Road and Old Bend road W of La Cresta, nr chaparral and oak, soil of open areas of hillside just above creek, ca. 240 m.s.m., 4 April 1995, *Doyle 7156* (UC); Kearney Mesa E off Eastgate Road, nr top of ravine overlooking Interstate Hwy 805, soil, open areas nr chaparral, R3W T15S S8, ca. 95 m.s.m., 3 April 1995, *Doyle 7194A* (UC); SAN LUIS OBISPO COUNTY: Eastern side of Santa Lucia Ranges, Chimney Rock Road W of Paso Robles, ca. 4.4 mi NE of jct. with Klaus Mine Road, soil, open areas on hillside with scattered oak, ca. 260 m.s.m., 23 April 1996, *Doyle 8210* (UC); SAN MATEO COUNTY: Eastern side of Santa Cruz Mts., Edgewood County Park, Sylvan Trail, soil nr top of trail bank at switchbacks, ca. 160 m.s.m., 7 April 1998, *Doyle 9442* (UC); SANTA BARBARA COUNTY: Transverse Ranges, Santa Ynez Mts., Los Padres National Forest, Snyder Trail S off Paradise Road, nr chaparral and oak, soil high on nr vertical bank of small ephemeral creek, ca. 335 m.s.m., 19 May 1998, *Doyle 9551* (UC); SANTA CLARA COUNTY: Diablo Range E of Gilroy, E side of Cañada Road, ca. 3.4 mi SW of jct. with Jamison Road, nr. oak and chaparral, ca. 115 m.s.m., 18 April 1978, *Doyle 2360* (UC); SANTA CRUZ COUNTY: Santa Cruz Mts., Santa Cruz campus of the University of California, Fuel Break Road nr jct. with Red Hill Road, chaparral and scattered oak, soil in openings of chaparral on hillside, ca. 290 m.s.m., 10 April 1997, *Doyle 8932* (UC); TUOLUMNE COUNTY: Western foothills of Sierra Nevada, Reynolds Ferry Road N of Tuttle Town, ca. 0.7 mi W of jct. with CA Hwy 49, ca. 260 m.s.m., 21 May 1996, *Doyle 8448* (UC); TULARE COUNTY: Sequoia National Park, on soil under weeds on a sloping hillside meadow (swale), 5 April 2005, *Doyle 11328* (ABSH, UC).