VESTURED PITS IN WOOD OF ONAGRACEAE: CORRELATIONS WITH ECOLOGY, HABIT, AND PHYLOGENY

ABSTRACT

All Onagraceae for which data are available have vestured pits on vessel-to-vessel pit pairs. Vestures may also be present in some species on the vessel side of vessel-to-ray pit pairs. Herbaceous Onagraceae do not have fewer vestures, although woods with lower density (Circaea L. and Oenothera L.) have fewer vestures. Some Onagraceae from drier areas tend to have smaller vessel pits, and on that account may have fewer vestures (Epilobium L. and Megacorax S. González & W. L. Wagner). Pit apertures as seen on the lumen side of vessel walls are elliptical, occasionally oval, throughout the family. Vestures are predominantly attached to pit aperture margins. As seen from the outer surfaces of vessels, vestures may extend across the pit cavities. Vestures are usually absent or smaller on the distal portions of pit borders (except for Ludwigia L., which grows consistently in wet areas). Distinctive vesture patterns were observed in the several species of Lopelia Cav. and in Xylonagra Donn. Sm. & Rose. Vestures spread onto the lumen-facing vessel walls of Ludwigia octovalvis (Jacq.) P. H. Raven. Although the genera are presented here in the sequence of a recent molecular phylogeny of Onagraceae, ecology and growth forms are more important than evolutionary relationships with respect to abundance, degree of grouping, and morphology of vestured pits. Designation of vesture types is not warranted based on the distribution of named types in Onagraceae and descriptive adjectives seem more useful, although more data on vesturing in the family are needed before patterns of diversity and their extent can be fully ascertained. Vestures are less common and may have a more complicated genetic basis than helical thickenings in vessels, but may be a more effective form of water column maintenance.

Key words: Ecological wood anatomy, growth form, Onagraceae, pit shape, wood xeromorphy.

Our knowledge of vestured pits is still quite incomplete. Bailey (1933) offered a major advance by not only documenting their systematic distribution, but also by describing and figuring three-dimensional morphology of vestures, using light microscopy with amazing skill. With the availability of SEM, our knowledge of vestured pits has advanced much further. Vestures in vessels and tracheids are in a magnification range (×5000 to ×15,000) at which SEM excels; the depth of focus of SEM facilitates three-dimensional imaging that is needed for understanding these structures. Vestured pits have been surveyed in two groups of Myrtales, Combretaceae (van Vliet, 1978; Carlquist, 2017a) and the Penaeaceae alliance (Carlquist, 2017a). These two groups lie at opposite poles of the Myrtales clade: Combretaceae is sister to all remaining Myrtales; Onagraceae–Lythraceae are sister to all remaining Myrtales except Combretaceae (Berger et al., 2016). Penaeaceae and allied families (Alzateaceae, Oliniaceae, Rhyynchocalycaceae) represent a crown group of Myrtales (Stevens, 2001 onward; Berger et al., 2016). As our knowledge of vestured pits in the families of Myrtales progresses, patterns in the evolution of these structures will become more evident.

The diversity of vestured pits within families of Myrtales and the relationship of these structures to ecology and habit have been largely unexplored because SEM work is time-intensive. Efforts to date have largely featured recording which families and genera have vestured pits and which do not (Jansen et al., 2001). These broad surveys have not permitted the examination of diversity within genera and they are incomplete with respect to the three-dimensional imaging of how vestures are grouped and how they traverse the pit cavities and pit apertures. More detailed studies of this sort have been attempted in the Penaeaceae alliance (Carlquist, 2017a) and in the genus Echium L. of the Boraginaceae (Carlquist, 2017b). Studies of this kind will permit correlations between vesture abundance and morphology to be correlated with diversity in ecology and growth form.

When further studies are carried out, we will understand more fully how these minute structures relate to the evolutionary and ecological radiation of particular
families and genera. For example, the two arboreal (sometimes shrubby) genera that are most species-rich in Australia, *Acacia* Mill. (Fabaceae) and *Eucalyptus* L’Hér. (Myrtaceae), both have vestured pits, leading one to suspect that vestured pits might have played a role in the dominance of these genera on this driest of all continents. Onagraceae offer a variety of growth forms, from annuals to trees to aquatic herbs, and range from moist forests to deserts; thus, they are an ideal group in which to investigate the extent to which vestured pits may be related to ecology and habit. Jansen et al. (2004) concluded that groups with vestured pits have radiated most in warm subtropical climates, but also pointed out that to test this relationship further, one should examine the range of variation in this character in individual families and genera. For example, in Brassicaceae (3710 species), a large family not considered by Jansen et al. (2004), all species apparently have vestured pits (Carlquist, 2016). The distribution of Brassicaceae may be characterized as mostly boreal to cool temperate, and the family is relatively rare in the tropics.

If we posit that vestured pits aid in the water economy of dry land Myrtaceae such as *Eucalyptus*, can vestured pits be held to function similarly in dry land Onagraceae such as the members of the annual genus *Clarkia* Pursh? *Clarkia* flowers late in the spring and completes its life span after the places it grows have dried up. The secondary xylem of an annual can be regarded as a single growth ring, with vestured pits clearly functional structures; this relationship is discussed further in the terminal section of this paper. The extremely small size of these vestured pits accounts for the historical lack of study of vestured pits by wood physiologists.

Here our observations are presented according to the evolutionary relationships proposed for Onagraceae (Levin et al., 2004; Wagner et al., 2007), a predominantly herbaceous family that is not well represented in wood collections. Our study includes 23 species (Table 1), a small but representative sample of the 657 species in the family (Wagner et al., 2007); each of the seven tribes is represented, and we have made an attempt to cover the ecological range of the family (Table 1, column 3). As we shall discuss later, the distribution of different kinds of vestured pits in Onagraceae seems clearly to be more closely related to ecology than to their evolutionary relationships. The coverage of species here for the most part matches that in earlier monographs of wood anatomy of the family (Carlquist, 1975, 1977, 1982), with several additions. There is clearly more to be learned about the nature and distribution of vestured pits in Onagraceae, but we consider the results presented here to be a solid start.

**Materials and Methods**

Although presence of vestured pits in Onagraceae was noted and illustrated by means of light microscopy earlier (Carlquist, 1975, 1977, 1982), those illustrations were limited compared to what can be achieved using SEM. As in the study of the Penaceae alliance of Myrtales (Carlquist, 2017a), permanent slides of wood sections mounted in Canada balsam made decades ago proved entirely satisfactory for the present study. For most species studied here, these slides were soaked in xylene until the cover slips loosened and could be removed. The sections were then retrieved. Because vessels in Onagraceae are grouped in radial rows and vessel-to-vessel pits are optimal for study of vesturing, tangential sections were preferred. After retrieval, sections were cleansed of Canada balsam with three changes of xylene at 60°C. The sections were then dried under pressure between pairs of glass slides to assure flatness. Sections were mounted on aluminum stubs, sputter-coated with gold, and examined with a Hitachi S2600N SEM (Tokyo, Japan).

Some wood samples for key taxa not examined in earlier studies (*Epilobium canum* (Greene) P. H. Raven, *Megacorax gracielanus* S. González & W. L. Wagner, and *Oenothera elata* Kunth subsp. *hookeri* (Torr. & A. Gray) W. Dietr. & W. L. Wagner) were taken from dried stem portions and boiled in water. They were then stored in 50% aqueous ethanol and sectioned with single-edged razor blades. The sections were next subjected to three changes of warm 50% aqueous ethanol and dried under pressure between clean glass slides. Tangential sections were studied unless otherwise indicated.

Various methods have been used by particular workers examining woods for vestured pits. There has been concern that minute droplets of substances that are artifacts might be interpreted as vesturing. Sodium hypochlorite (bleach) has been applied to sections in an attempt to minimize or reduce such artifacts (Jansen et al., 2001). We have tried this technique (comparative preparation of the three species listed in the preceding paragraph) and found that some swelling can occur—a minor artifact, perhaps, but one that deserves attention. Artifacts usually are easily detectable because of their patterns: varied and irregular in shape, present in one part of a section but not consistently present in or related to pits. White flecks on the sections of Figure 6 are considered artifacts, but they are easily distinguishable from the vestures. Unfortunately, in studies on plant anatomy and in handbooks of plant microtechnique, results of technique comparisons are rarely offered. Instead, favored recipes are adopted.

Nomenclature for species of Onagraceae has continually been improved as molecular and other evidence has accumulated. Consequently, some names used by Carlquist (1975, 1977, 1982) have been superseded, and the most recent equivalents are offered here (Wagner et al., 2007).
Terminology for morphology of vestured pits or for vestures not closely associated with pits (e.g., Fig. 1B) has been offered by van Vliet (1978) and others. SEM-based data on vestures still do not offer three-dimensional understanding (pits seen from inner surfaces of vessels or pits seen from outer vessel surfaces or sectional views of pits) for very many species of angiosperms, and use of categories rather than descriptive adjectives seems premature at this point in time. The pits shown are vessel-to-vessel pit pairs unless otherwise stated in captions. All magnification bars in Figures 1–12 = 2 μ.

RESULTS

VESTURED PITs IN ONAGRACEAE: VARIATION AND DISTRIBUTION

Results are presented for taxa in the phylogenetic sequence in Wagner et al. (2007), based in part on Levin et al. (2004). Tribes are recognized only in subfamily Onagroideae.

<table>
<thead>
<tr>
<th>Taxon</th>
<th>Collection (herbarium)</th>
<th>Habit, ecology</th>
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<tr>
<td><strong>Subfamily Ludwigioideae</strong></td>
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<tr>
<td>Ludwigia octovalvis (Jacq.) P. H. Raven</td>
<td>Raven 6571 (DS)</td>
<td>Subtropical woody aquatic herb</td>
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<tr>
<td>L. pedunculosa Michx.</td>
<td>Ekman 13415 (MO)</td>
<td>Subtropical woody aquatic herb</td>
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<td><strong>Subfamily Onagroideae</strong></td>
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<tr>
<td>Tribe Hauyeae</td>
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<td>Hauya elegans DC. subsp. cornuta (Hensl.) P. H. Raven &amp; Breedlove</td>
<td>Breedlove 10589 (DS)</td>
<td>Subtropical moist forest tree</td>
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<tr>
<td>Tribe Circaeae</td>
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<tr>
<td>Fuchsia arborescens Sims</td>
<td>Breedlove 7145 (DS)</td>
<td>Temperate humid forest tree</td>
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<tr>
<td>F. bolissana Carrière</td>
<td>Stork et al. 10395 (UC; cult. UC656965)</td>
<td>Tropical montane shrub</td>
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<td>F. excorticata L. f.</td>
<td>Carlquist 1193 (RSA)</td>
<td>Subtropical moist forest tree</td>
</tr>
<tr>
<td>F. magellanica Lam.</td>
<td>? s. coll. (UC 49:906)</td>
<td>Subtropical moist forest shrub</td>
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<tr>
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<td>A. Christ s.n. in 1975 (MO)</td>
<td>Cool temperate perennial</td>
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<td>Subtropical shrub</td>
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<td>L. seminandra Plitmann, P. H. Raven &amp; Breedlove</td>
<td>Breedlove 4237 (DS)</td>
<td>Subtropical shrub</td>
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<td>S. González 6532 (US)</td>
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<td>Tribe Gongylocarpeae</td>
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<td>Moran 10810 (DS)</td>
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<td>Breedlove 7154 (DS)</td>
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<td>Carlquist s.n. in 2016 (SBBG)</td>
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<td>E. brachycarpum C. Presl</td>
<td>Raven 20668 (RSA)</td>
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<tr>
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<td>Thomas 8215 (DS, RSA)</td>
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<td>Raven 20254 (DS)</td>
<td>Moist to dry annual</td>
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<td>Raven 20172 (DS)</td>
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<td>Chylismia megalantha (Munz) W. L. Wagner &amp; Hoch</td>
<td>Bentley s.n. in 1961 (DS, RSA)</td>
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<td>Raven 19961 (DS)</td>
<td>Temperate biennial</td>
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<td>O. cinerea (Wooton &amp; Standl.) W. L. Wagner &amp; Hoch</td>
<td>Raven 19224 (DS)</td>
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<td>O. elata Kunth subsp. hookeri (Torr. &amp; A. Gray.) W. L. Dietr. &amp; W. L. Wagner</td>
<td>Carlquist s.n. in 2015 (SBBG)</td>
<td>Robust temperate biennial</td>
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<td>Oenothera sect. Pentaphyllum (Pennell) Munz</td>
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<tr>
<td>O. linifolia Nutt.</td>
<td>Raven 19450 (DS)</td>
<td>Temperate perennial</td>
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Subfamily Ludwigioideae (Fig. 1A–D)

Pit apertures as seen on inner vessel surfaces are elliptical (Fig. 1A, D) to oval (Fig. 1B). As seen from outer surfaces of vessels, the pits are circular. Vestures are variously aggregated, either as groups (Fig. 1B), plates, or coralloid-branched fascicles (Fig. 1A, D). In *Ludwigia octovalvis* (Jacq.) P. H. Raven, inner surfaces of vessels may be smooth (Fig. 1A) or vestured (Fig. 1B), the only instance reported here in which inner surfaces of vessels are so vestured. In
this example (Fig. 1B), vestures become sparser distal to the pit apertures. As seen on inner surfaces of vessels, vestures may appear to branch from the pit aperture margins, but in fact, as seen on outer vessel surfaces (Fig. 1C), they branch from pit cavity surfaces as well.

Subfamily Onagroideae (Figs. 1E, F, 2–12)

Tribe Hauyeae (Fig. 1E, F). As seen on inner surfaces of vessels, vested pits of Hauya DC. are elliptical (Fig. 1E, F). Vestures may be coralloid-anastomosing (Fig. 1E) or irregularly aggregated (Fig.
1F). The two species of *Hauya* are trees (a habit rare among Onagraceae), but their vestures have no signature, indicating they are different from those of less woody Onagraceae.

*Tribe Circaeae* (Figs. 2, 3A–E). Although *Fuchsia* L. and *Circea* L. have usually been separated into two tribes in earlier classifications of Onagraceae, they form a strongly monophyletic clade in molecular
analyses (Levin et al., 2004; Berger et al., 2016), and Wagner et al. (2007) found enough morphological synapomorphies that the two genera are best regarded as a single tribe.

The 107 species of Fuchsia, widely distributed in Latin America but with three species in New Zealand and one in Tahiti, are shrubs or small to exceptionally large trees, with one exception: the sprawling woody

Figure 4. SEM images of vested pits in Lopezieae, as seen on inner vessel surfaces (A, B, D–F) and outer vessel surfaces (C). A–D. Lopezia longiflora Decne. —A. Pit notably wide, vestures extend across the pit aperture. —B. Reticulate vesture aggregation of vestures. —C. Vesture tips on the platelike vesture bridges; a narrow pit border portion devoid of vestures. —D. Vessel-to-vessel pits (left) have abundant vestures, whereas vessel-to-ray pits (right) lack them (radial section of wood). E, F. Lopezia lopezoides (Hook. & Am.) Plitmann, P. H. Raven & Breedlove. —E. Pits laterally elongate, wall smooth. —F. Dentate-serrate vesture aggregations, higher magnification. Scale bar = 2 μ.
F. procumbens R. Cunn. of New Zealand. In the three very different species we examined, the lumen surfaces of vessels (Fig. 2A–C) have oval to elliptical pit apertures, vertically wider than those of most other Onagraceae. Vestures do not appear to be restricted to pit aperture margins; they are aggregated in various ways across the extent of the pit aperture, as seen from the lumen side of the vessel. One sees irregular groupings (Fig. 2A, B) or

Figure 5. SEM images of vestured pits in vessels of Lopezieae: Lopezia Cav. (A–D) and Megacorax S. González & W. L. Wagner (E, F); pits seen on inner vessel surface (A–C, top of D, E) and outer surface of vessels (D, bottom; F). A, B. Lopezia miniata Lag. ex DC. —A. Vessel-to-vessel pits broadly oval, with densely aggregated vestures. —B. Vessel-to-ray pits have few vestures. C, D. Lopezia seneiandra Plittmann, P. H. Raven & Breedlove. —C. Oval to circular pits with dense aggregations of vessels covering the pit aperture areas. —D. Section showing inner face of vessel (above) and also exposing outer vessel surface (below); vestures cover entire pit border area. E, F. Megacorax gracielanus S. González & W. L. Wagner. —E. Pits on inner vessel surface are very small, narrow. —F. Outer vessel surface reveals that vestures are relatively few and extend onto the pit border. Scale bar = 2 μ.
coralloid branched fascicles (Fig. 2C, D). A few shallow grooves are present in vessel walls of *F. boliviana* Carrière (Fig. 2B), but walls of the other species studied are smooth (Fig. 2A, C, D). As seen from outer surfaces of vessels, vestures of vessel-to-vessel pits do not cover the entire pit border; the distal portions of the pit borders lack vestures (Fig. 2E, F). Vessel-to-parenchyma pits (Fig. 2E, above) are simple and not vestured.

The other genus of the tribe, *Circaea*, consists of nine species of rhizomatous perennial herbs...
distributed in cooler regions across the Northern Hemisphere. The species we examined, *C. canadensis* (L.) Hill, like all members of the genus, is scarcely woody, and its less extensive vesturing may be related to its habit. Some pits are oval and moderately

vestured (Fig. 3A), whereas others are horizontally elongate and sparsely vested (Fig. 3B–D). Vestures are restricted to the edges of pit apertures, whether seen from the inner sides of vessels (Fig. 3A–D) or from outer surfaces (Fig. 3D). In other Onagraceae,
the vestures extend distally from the pit apertures onto the pit border.

*Tribe Lopezieae (Figs. 4, 5).* Lopezieae are distinctive in having relatively large pits with numerous and densely placed vestures; each of the four species (of 22 total) that we examined belongs to a different section of this diverse genus, which occurs in Mexico and Central America. *Lopezia longiflora* Decne. (section Jehlia (Rose) Plitmann, P. H. Raven & Breedlove) is notable for its vertically wide pits (Fig. 4A–D), oval in shape. Vestures...
are present across the pit, as seen from the lumen side of the vessel. Vestures are aggregated into plates (Fig. 4B) from which vestures branch outwardly. Although vessel-to-vessel pits are densely vestured in L. longiflora, vessel-to-ray pits (Fig. 4D, right) are not vestured.

*Lopezia lopezioides* (Hook. & Arn.) Plitmann, P. H. Raven & Breedlove (section *Diplandra* (Hook. & Arn.) Plitmann, P. H. Raven & Breedlove) has distinctive horizontally elongate vessel-to-vessel pits (Fig. 4E, F). The vestures are aggregated into interdigitating...
irregular shapes (Fig. 4E). At higher magnification, one sees that these vesture groupings terminate in round and sometimes serrate edges (Fig. 4F).

*Lopezia miniata* Lag. ex DC. (section *Lopezia*) (Fig. 5A, B) has large vessel-to-vessel pits that are broadly oval in shape. The vesture aggregations are dense, with numerous vesture tips evident (Fig. 5A). Vessel-to-ray pits (Fig. 5B) are vertically wider than those of vessel-to-vessel pits and may either lack vesture or have a few.

Lopezia semeiandra Plitmann, P. H. Raven & Breedlove (section Riesenbachia (C. Presl) Plitmann, P. H. Raven & Breedlove) has oval pits as seen on the inner vessel surface. The vestures are globular and densely aggregated (Fig. 5C, D, top). As seen from the outer surface of a vessel (Fig. 5D, below), vestures cover most of a pit border. The density of vestures in L. miniata and L. semeiandra is notable.

The dry land subshrub Megacorax S. González & W. L. Wagner (Fig. 5E, F) has, by contrast, very small pits as seen from the outer surface.
vessel-to-vessel pits that are narrowly elliptical in shape as seen from the lumen side of a vessel wall (Fig. 5E). Vestures are mostly located on the margins of the inner pit aperture, and as seen from the outside of the vessel, they decrease in size toward the edge of the pit border (Fig. 5F). The vestures show little aggregation compared to those of *Lopezia* Cav. The monotypic, northern Mexican *Megacorax*, which was first
collected in 2001, has a number of features that are ancestral to those exhibited by the closely related Lopezia.

**Tribe Gongylocarpeae (Figs. 6, 7A, B).** This tribe consists of a single genus, Gongylocarpus Schltdl. & Cham., with two species, both endemic to Mexico. The pits shown for *G. rubricaulis* Schltdl. & Cham. (Fig. 6A–F), a robust annual, are all from radial sections; a similar range of pit shapes occurs on tangential walls of vessels as well. Some pits are markedly elongate horizontally (Fig. 6A, B). These elongate pits may be evenly vestured (Fig. 6A) or vestures may be lacking in some places (Fig. 6B). In contrast, the majority of vessel-to-vessel pits is elliptical to oval (Fig. 6C–F). Where most abundant, the vestures form coraloid branching patterns (Fig. 6C) or serratoid edgings of the pit apertures (Fig. 6D, E). Smaller pits (Fig. 6E) have fewer vestures than larger pits. As seen from the outer surface of a vessel, the pits are vestured adjacent to the pit apertures, but the border portions are not vestured (Fig. 6F).

The other species of Gongylocarpus, *G. fraticulosus* (Benth.) Brandegee, is a subshrub that occurs on very dry sites around Bahía de Magdalena on the Pacific Coast of Baja California. It has small pits (Fig. 7A, B) compared to those of most Onagraceae. The vessel-to-vessel pits illustrated (Fig. 7A) are on tangential walls of vessels, which are lightly grooved on the lumen side. The vestures, viewed from the outer surfaces of a tangential vessel wall (Fig. 7B), are relatively few and variously aggregated. The vestures cover the entire border surface (Fig. 7B).

**Tribe Epilobieae (Fig. 7C–F).** This tribe includes two genera, the very robust rhizomatous *Chamaenerion* Ség., with eight widespread boreal species, not examined for this study, and the cosmopolitan genus *Epilobium* L., largest in the family, with the great majority of its 165 species herbaceous perennials, mainly growing in moist habitats. The species of the evolutionarily divergent smaller sections include a number of xeric and some woody species, of which we examined two for this study: the robust annual *E. brachycarpum* C. Presl (section *Xerolobium* P. H. Raven) and the subshrub *E. canum* (section *Zauschneria* (C. Presl) P. H. Raven).

The vessel-to-vessel pits are small, with relatively few vestures, in both of these species, *Epilobium brachycarpum* (Fig. 7C–E) and *E. canum* (Fig. 7F). The pits are narrowly elliptical, with vestures restricted to margins of the pit apertures (Fig. 7C–F). As seen from the lumen side of a vessel, the vestures appear as rounded lumps (Fig. 7D) or serratoid edgings of the pit apertures (Fig. 7E, F).

**Tribe Onagreae (Figs. 8–12).** This tribe includes the remaining 13 genera in the family, including many familiar species, all restricted as native plants to the Western Hemisphere.

*Xylonagra* Donn. Sm. & Rose (Fig. 8) is comprised of a single distinctive species of red-flowered shrubs restricted to desert areas in the central Baja California Peninsula of Mexico. Phylogenetically, it is sister to the remaining genera of Onagreae (Levin et al., 2004). Walls of vessels are either striate with helical thickenings (Fig. 8A–C, E) or smooth (Fig. 8D, F), with *Xylonagra* the only member of Onagreae in which such helices have been observed. All vessel-to-vessel pits are horizontally elongate and elliptical in shape (Fig. 8A–F). The vesturing can be sparse (Fig. 8B, E), but more commonly vesturing is extensive. Vestures in some are fused together into perforated plates (Fig. 8A, C, D, F). This is sufficiently distinct from other examples of vesturing that we tested portions of *Xylonagra* wood sections to determine whether organic solvents (benzene, alcohol) or oxidizing agents (sodium hypochlorite) would dissolve these bridges of apparent secondary wall material. The bridges did not dissolve in these reagents, so the structures illustrated in Figure 8 are judged to be valid expressions of vesturing.

*Clarkia* (Fig. 9), which consists of 42 species of annual herbs centered in California, grows in places that dry out as the plants mature. Its vessels present well-defined vesturing. This might not be expected for an annual, but vesturing is well represented in other annual Onagraceae. The vestures in *Clarkia* may reflect its occurrence in dry habitats. Vessel-to-vessel pit apertures, as seen on inner surfaces of vessels, are narrowly elliptical (Fig. 9A, B) to oval (Fig. 9C). Pit cavities are elongate (Fig. 9D) to circular (Fig. 9E). Vessel walls are thick; vestures may be seen in pit canals at various levels (Fig. 9F). Vesturing is primarily distributed along the pit apertures (Fig. 9A–E) and portions of pit borders distal to the apertures (Fig. 9D, E). Degree of vesture abundance differs: wider pits (Fig. 9A–C) have more numerous vestures than narrower ones (Fig. 9B). Vestures along the pit apertures are fused into short fascicles (Fig. 9A–E).

*Chylismia* (Torr. & A. Gray) Nutt. ex Raim. (Fig. 10C), *Eulobus* Nutt. ex Torr. & A. Gray (Fig. 10D–F), and *Oenothera* sect. *Calylophus* (Spach) W. L. Wagner & Hoch (Fig. 10A, B), *Oenothera* sect. *Gaura* (L.) W. L.Wagner & Hoch (Fig. 11), *Oenothera* sect. *Oenothera* (Fig. 12A–C), and *Oenothera* sect. *Peniophyllum* (Pennell) Munz (Fig. 12D–F) are members of the most species-rich clade in Onagraceae. Paraphyly in the group formerly treated as *Camissonia* Link led to the recognition of several new genera, including *Chylismia* and *Eulobus*, and *Oenothera* proved to be polyphyletic, leading to inclusion of formerly segregate genera *Calylophus* Spach and *Gaura* L. (Levin et al., 2004; Wagner et al., 2007).
Vestured Pits in Wood of Onagraceae

**Oenothera sect. Calylophus, Chylismia (as Camissonia megalantha) (Munz) P. H. Raven, and Eulobus (as Camissonia californica) (Nutt. ex Torr. & A. Gray) P. H. Raven** were reported to lack vestures on vessel-to-vessel pits (Carlquist, 1975) on the basis of light microscopy. Using SEM, we found vestures in all three groups, although the vestures are not abundant (Fig. 10). The same collections were examined with both light microscopy and SEM. *Oenothera hartwegii* Benth. (section *Calylophus*), a slightly woody perennial, has irregular vestures evident on margins of pit apertures (Fig. 10A). However, as seen on the outer surfaces of vessels (Fig. 10B), vestures spread to various extents onto the pit border. *Chylismia megalantha* (Munz) W. L. Wagner & Hoch (Fig. 10C) and *Eulobus californicus* Nutt. ex Torr. & A. Gray (Fig. 10D–F) are annual herbs and similar in having vestures on vessel-to-vessel pits. Vessel-to-ray pits are wide and nearly devoid of vestures (Fig. 10E). As seen from the outside of vessels, vessel-to-vessel pits of *E. californicus* have vestures on the pit apertures, but vestures are absent or nearly so on the pit borders (Fig. 10F).

*Oenothera sect. Gaura* includes *O. gaura* W. L. Wagner & Hoch (formerly *Gaura biennis* L.; Fig. 11A–C), a biennial with relatively small, elliptical pits (Fig. 11A, B). Variation in grouping of vestures can be visualized by comparing Figure 11A and B. As seen from the outer surface of vessels, vessel-to-vessel pits show irregularly aggregated vestures; vestures are absent from the pit borders (Fig. 11C). *Oenothera cinerea* (Wooton & Standl.) W. L. Wagner & Hoch (*G. villosa* Torr.; Fig. 11D–F), a woody perennial, has pits a little wider and vestures more aggregated than those of *O. gaura*. These aggregations in *O. cinerea* can take the form of interdigitating ridges (Fig. 11D) or coralloid fascicles (Fig. 11E). The numerous globular tips of these fascicles can be seen in Fig. 11F; the pit borders lack vestures.

*Oenothera sect. Oenothera* (Fig. 12A–C). *Oenothera elata* subsp. hookeri is a rosette-forming biennial. The rosettes and inflorescence bases provide sufficient secondary xylem for study, although this wood is less dense (manoxylic) than that of most other Onagraceae. Pits are distinctively large and circular to oval (Fig. 12A–C) and thereby unusual for Onagraceae. The vestures on vessel-to-vessel pits are slender and branched (Fig. 12C), rather unlike those of other Onagraceae illustrated. Vessel-to-ray pits are sparsely vestured or non-vestured (Fig. 12B).

*Oenothera sect. Peniophyllum* (Fig. 12D–F). Vessel-to-vessel pits of the annual *O. linifolia* Nutt. have narrowly elliptical pit apertures, as seen from the insides of vessels (Fig. 12D, E). The stems of *O. linifolia* are slender and pycnoxylic (high density) compared to those of the robust biennial *O. elata*. These differences appear to be correlated with the difference between the two species in pit shape. As seen from the lumen sides of vessels, the vestures are small and irregular in shape. As seen from the outer surfaces of vessels (Fig. 12F), vestures are moderately aggregated and absent from pit borders.

**IMPERFORATE TRACHEARY ELEMENTS**

As far as is known, all Onagraceae have living fibers as imperforate tracheary elements (Carlquist, 1975, 1977, 1982). In this respect, Onagraceae contrast with Penaeaceae, which have bordered pits, often vestured, on imperforate tracheary elements (Carlquist, 2017a) as well, but the imperforate tracheary elements of Penaeaceae are tracheids (Carlquist & DeBuhr, 1977). Living fibers of Onagraceae are illustrated here for *Lopezia longiflora* (Fig. 3F). The fibers have gelatinous walls (as do those of other Onagraceae: Carlquist, 1975). These walls shrink into layers in anhydrous preparations. Pits are extremely small and may be visible only occasionally on fiber walls (Fig. 3F, arrow). Ovoid starch grains, held in place by cytoplasm, are abundant. Fiber-tracheids are uncommon in Myrtales; tracheids and vasicentric tracheids or libriform fibers (including living fibers) are more common.

**Conclusions**

1. All vessel-to-vessel pit pairs in Onagraceae have at least some vesturing as seen either from the lumen side or the outer surface of the vessels. Vesturing is sparse to absent on the vessel side of vessel-to-ray pit pairs.

2. Vessel-to-vessel pit apertures are mostly narrowly elliptical as seen on the lumen side, somewhat wider as seen on vessel-to-ray contacts. Pit cavities (pit borders) are circular.

3. Vestures are most commonly aggregated into fascicles, which are reminiscent of coralloid branching as seen from the lumen side. The branching tips, as seen from the outer surface of the vessel (if pit membranes are removed by sectioning), are knoblike.

4. Diversity is present within genera and even within species (e.g., *Gongylocarpus rubricaulis*) with respect to modes of grouping of vestures. In addition to coralloid fascicles, ridgelike groupings along the pit aperture and irregular groupings of various kinds occur. The presence of intermediate conditions and of variation within a sample suggests that recognition of numbered types (e.g., van Vliet, 1978) is probably not feasible, at least in Onagraceae and in a number of other Myrtales. Individual species may have species-specific modes of vesture occurrence.
(5) Smaller pits, with fewer vestures, characterize some dry land species with other xeromorphic wood features: Epilobium spp. (both species reported here grow in xeric habitats), Megacorax, Oenothera gaura, and O. elata. Relatively large pits with large numbers of vestures characterize Onagraceae of relatively mesic habitats, such as Fuchsia and, to a degree, Lopezia. Woods with low density may have fewer vestures per pit (e.g., Circaea canadensis and O. elata). Stems that are filiform with higher density (e.g., Clarkia xanti-ana A. Gray and O. linifolia, both annuals) tend to have narrower pit apertures and more numerous vestures per pit. All of these observations suggest that pit shape and vesture configurations are related to ecology, growth form, and wood density rather than to phylogeny per se. One should keep in mind that some species that grow during a moist season may flower and fruit under arid conditions, and wood features representing the latter extreme may govern xeromorphic wood configurations.

(6) Vestured pits are universally present in vessels of Onagraceae despite their wide diversity in ecological preferences and habits; the same is true for some unrelated families, including Brassicaceae (Carlquist, 2016). This suggests that vesture presence may have evolved in a limited number of angiosperm families (e.g., Jansen et al., 2001), and that the genetic nature of vesturing may be complex, neither attained nor lost readily.

(7) Distinctive types of vesturing were observed in Xylonagra (plates of fused vestures) and the various species of Lopezia (large numbers of vestures per pit, crowded but not fused into fascicles). More instances of distinctive vesture types may be expected to be found as more families of Myrtales are examined closely. Because of technical considerations, we are only beginning to survey families in detail, so that any assessment of patterns of diversity should be considered preliminary.

(8) Ludwigia octovalvis has vestures on the lumen surfaces of vessels, extending away from the pit apertures, in some areas of vessels. Other Onagraceae examined here lack such extended vesture distribution, although this phenomenon has been reported and illustrated in other Myrtales, such as Metrosideros Banks ex Gaertn. of the Myrtaceae (Meylan & Butterfield, 1974, 1978) or Alzatea Ruiz & Pav. of the Alzateaceae (Carlquist, 2017a). Since Ludwigia L. is phylogenetically sister to all other Onagraceae, the distribution of its vestures may, like many of its other features, represent a retained archaic characteristic.

(9) Pit borders, as seen on outer vessel surfaces where pit membranes have been removed by sectioning, may be entirely covered with vestures (Ludwigia and Lopezia), or vestures may be absent from distal portions of the border and concentrated closer to the pit canal (Onagraceae). Where vestures on pit apertures are few (Circaea and Euobus californicus), vestures may be entirely restricted to the edges of the pit apertures (vessel lumen side) and absent elsewhere on the pit borders.

(10) Narrow pit apertures and presence of vestures both seem related to wood xeromorphy (Carlquist, 1982, 2017a). Prominent helical thickenings, indicative of xeromorphy in wood of angiosperms at large (Carlquist, 1966, 1975), are present in Onagraceae studied only in Xylonagra. A related phenomenon, grooves interconnecting pit apertures (coalescent pit apertures) was observed in a few Onagraceae, such as Gongylocarpus fruticulosus, where the grooves are relatively inconspicuous. Helical sculpture of vessel walls may be less effective at wetting or moisture reten-tion than pit vestures, although both have been hypothesized to serve in this regard (Carlquist, 1982, 2017a; Kohonen, 2006; Kohonen & Helland, 2009).

Literature Cited


