

A new interpretation of floral morphology in *Garrya* (Garryaceae)

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Previous descriptions of *Garrya* (Garryaceae) floral morphology are inconsistent. Anatomical studies have long confirmed that the pistillate flowers possess an inferior ovary. Descriptions of the staminate flowers sometimes describe a vestigial superior ovary; other authors report no such structure. New observations of staminate flowers confirm the presence of an intrastaminal structure. I hypothesize that the structure previously interpreted as a vestigial ovary is, in fact, a vestigial nectary disc. Thus the staminate flowers cannot be considered hypogynous and do not contradict the interpretation of the pistillate flowers as epigynous. This observation resolves the paradox of contrasting floral form in the pistillate and staminate flowers.

KEYWORDS: euasterids, floral morphology, *Garrya*, Garryales, ovary position, phylogeny

INTRODUCTION

Garrya Dougl. ex Lindl. (the only genus of Garryaceae) comprises 15 species restricted to western North America, Central America, and the Caribbean. Two subgenera, *Garrya* and *Fadyenia* (Endl.) G.V. Dahling, are recognized. *Garrya* species are dioecious shrubs or small trees with decussate evergreen leaves. The flowers are arranged in catkin-like inflorescences (Figs. 1A–B) and are subtended by connate bracts. The staminate flowers have four free corolla lobes united at their apices by intertwined hairs (Fig. 1A). The pistillate flowers of subgenus *Garrya* have two (rarely four) minute appendages that are adnate to the ovary and alternate with the styles (Fig. 1B); these are interpreted as a reduced perianth (Eyde, 1964). The pistillate flowers of subgenus *Fadyenia* generally lack these appendages, but the subtending bracts are sometimes adnate to the ovary in the terminal flowers of an inflorescence (Eyde, 1964). Inflorescence and floral morphology, abundant pollen production, and winter flowering (in the Pacific coast species) all suggest that *Garrya* species are wind-pollinated (Hallock, 1930; Dahling, 1978).

Molecular phylogenetic studies consistently resolve *Garrya* and *Aucuba* Thunb. (the only genus of Aucubaceae, with three species in East Asia) as sister taxa in the euasterid order Garryales (Soltis & al., 2000; Kårehed, 2001; Bremer & al., 2002). In contrast to *Garrya*, *Aucuba* is entomophilous (Abe, 2001). The staminate and pistillate flowers have dark-colored perianths and prominent nectary discs (Figs. 1C–D).

A relationship with *Aucuba* was first proposed by Baillon (1877) who noted numerous similarities (decussate evergreen leaves, dioecy, tetramerous flowers,

baccate fruit) and reported a successful graft between the two genera. Horne (1914) confirmed the grafting result, and reported unsuccessful grafts with other putative *Garrya* relatives. In an early chemotaxonomic study (Herissey & Lebas, 1910), the iridoid aucubin was described from *Garrya* and *Aucuba*. Other shared chemical traits include abundant petroselinic acid in the endosperm (Kleiman & Spencer, 1982), and the presence of caffeic acid (Bate-Smith, 1962) together with the absence of gallic and ellagic acids (Bate-Smith & al., 1975). Pollen similarities between *Garrya* and *Aucuba* are illustrated by Eramian (1971).

The remaining member of Garryales is the dioecious, East Asian tree *Eucommia* Oliver (forming the monotypic Eucommiaceae). *Eucommia* differs greatly in vegetative (alternate, deciduous leaves) and reproductive morphology (stamens 5–12 or more, perianth absent, fruit a samara). A relationship with *Garrya* had not been proposed prior to the molecular phylogenetic results. However, both genera are dioecious and both synthesize gutta-percha (trans-1, 4-polyisoprene) (Roth & al., 1985). This is a potential synapomorphy for Garryales (although its occurrence in *Aucuba* remains unknown).

Considering the abundant and diverse supporting evidence, the phylogenetic position of *Garrya* appears well-established. However, the current confidence contrasts with a long history of uncertainty and debate. In his original description of the genus, Lindley (1834) wrote “Although this plant cannot be compared for beauty to the Berberries, *Ribes*, Lupines, *Pentstemons*, *Clarkias*, *Calochorti* [italics and spelling as in the original], and other fine things discovered by Mr. Douglas, it is probable that it is the greatest botanical curiosity in all his collections”. He described the new family Garryaceae and

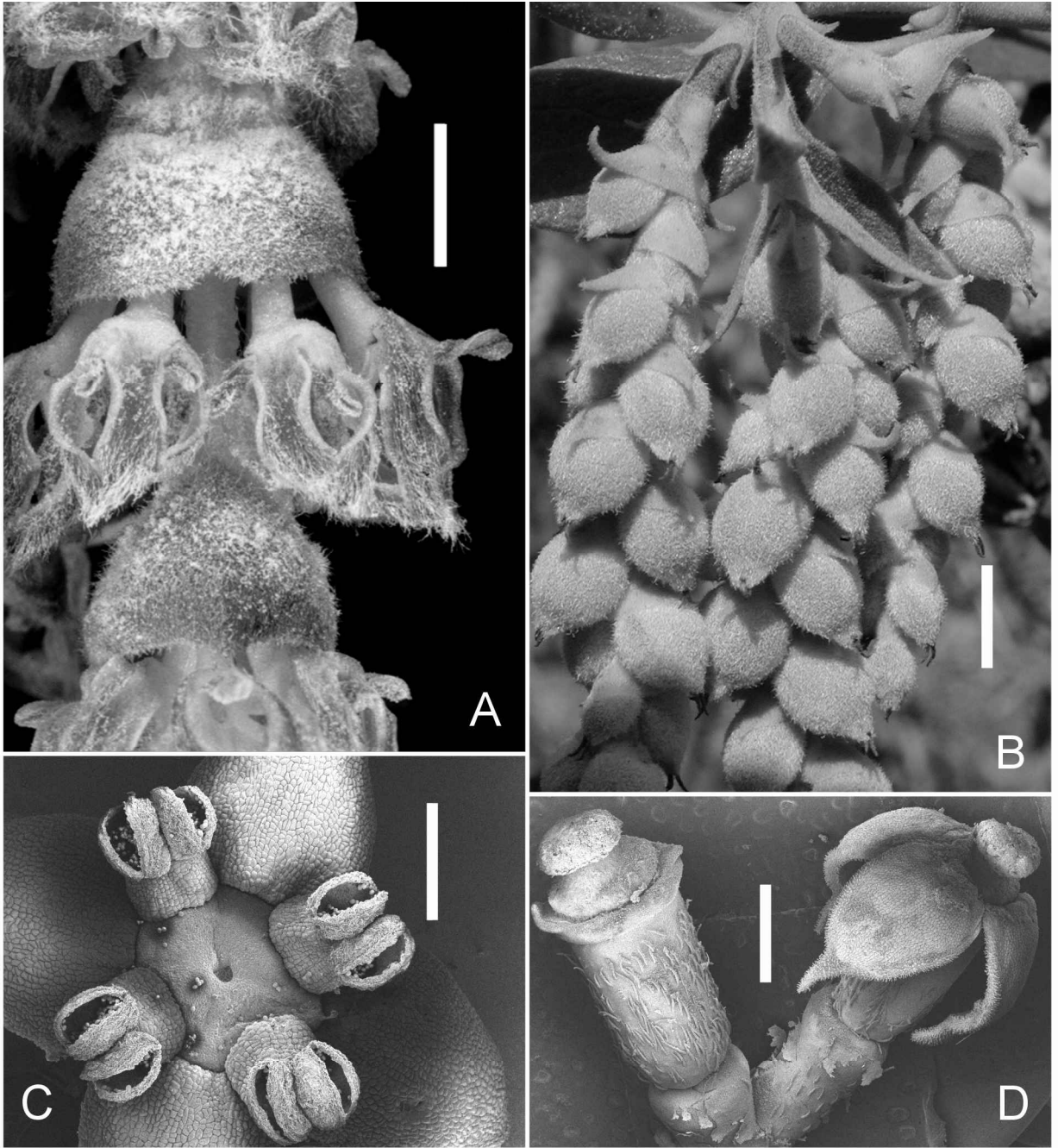


Fig. 1. *Garrya* and *Aucuba* flowers. A, staminate inflorescence of *Garrya elliptica* showing cupulate subtending bracts, corolla lobes united at the apex by intertwined hairs, alternate stamens, and abundant pollen (photograph by Gerald D. Carr); B, pistillate inflorescence of *G. veatchii*, the minute perianth lobes being visible at the base of the dark styles; C, staminate flower of *Aucuba japonica* with prominent nectary disc; D, pistillate flowers of *A. japonica* with prominent nectary discs (the corolla lobes have been removed on one flower to show the calyx lobes and inferior ovary). Scale bars: A–B = 5 mm, C = 1 mm, D = 2 mm.

compared it to the angiosperms Chloranthaceae, Piperaceae, Fagaceae, Urticaceae, Phyllanthaceae (as Stilaginaceae), and Crypteroniaceae (as Hensloviaceae), and the non-flowering seed plant, *Gnetum* L. Lindley (1846) later established the order Garryales for Garryaceae and

Helwingiaceae, and related the order to the above amenitiferous groups.

An affinity with Cornaceae was first proposed by Hooker (1867), who placed *Garrya* in that family. Baillon's (1877) association of *Garrya* and *Aucuba* sup-

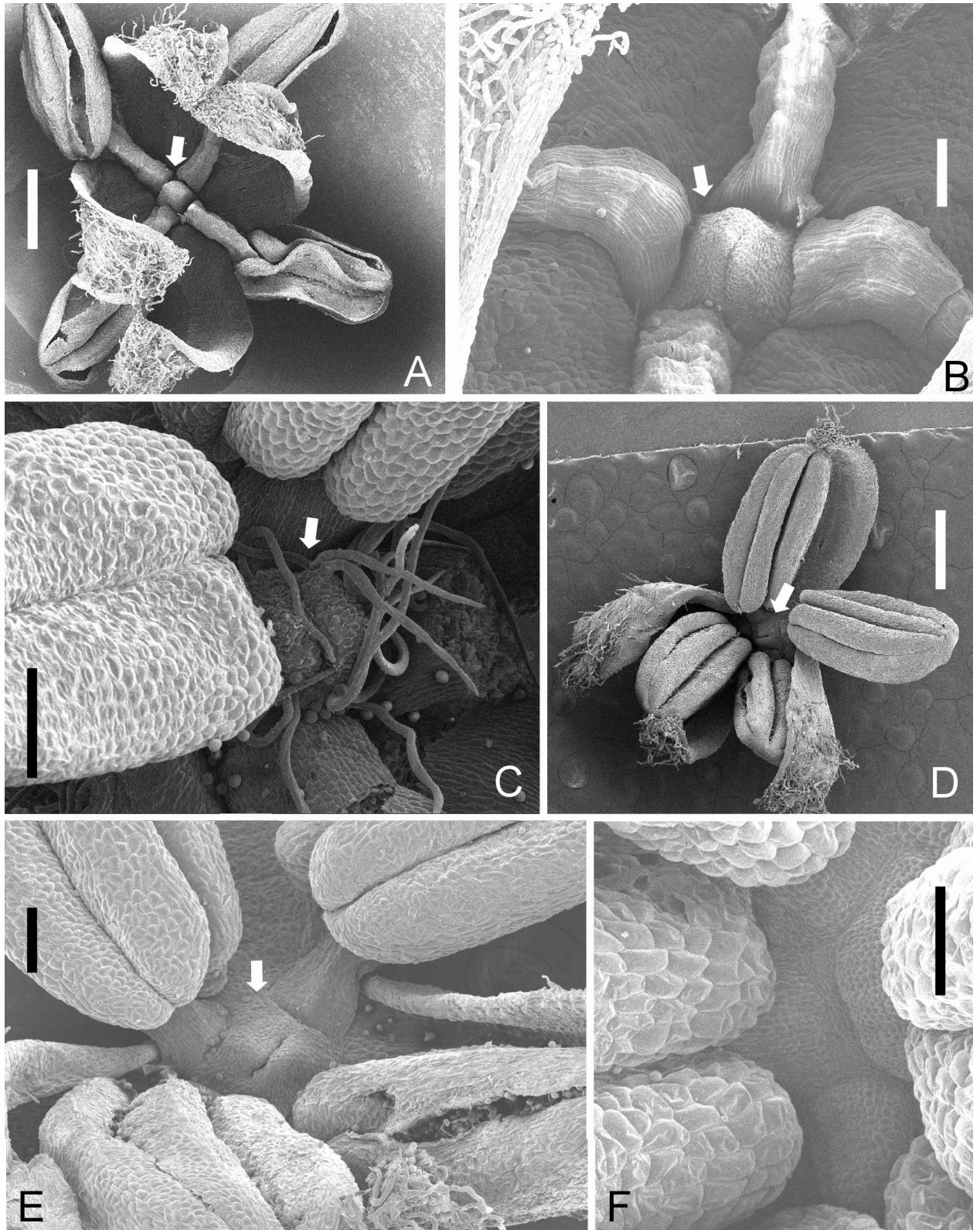


Fig. 2. Staminate flowers of *Garrya*. Arrows point to the vestigial nectary discs. A, *G. veatchii* at late anthesis; B, same flower at higher magnification; C, *G. elliptica*, one stamen removed to show nectary disc, which is covered with long trichomes in this species; D, *G. longifolia* at anthesis; E, same flower at higher magnification; F, another flower in the same inflorescence, lacking a vestigial nectary disc at the intersection of the four filament bases. Scale bars: A, D = 1 cm; B, C, E, F = 250 μ m.

Table 1. Specimens of *Garrya* examined. Vouchers at OSC, unless indicated otherwise.

<i>Garrya elliptica</i> Dougl. ex. Lindl.	U.S.A., California, <i>Elmer 4469, Heller 7220, Randall s.n.</i> (ORE). Oregon, <i>Dean s.n., Gorman s.n.</i> (ORE), <i>Liston 1218, Steiner 1446</i> (ORE), <i>Steward 6006</i> .
<i>Garrya longifolia</i> Rose	Mexico, Morelos, <i>Liston 1216</i> .
<i>Garrya veatchii</i> Kellogg	California, <i>Liston 1146</i> .

ported this viewpoint, since at the time, *Aucuba* was universally placed in Cornaceae. Subsequent authors either accepted a position in or near Cornaceae, or associated *Garrya* with the Englerian “Amentiferae” (summarized in Dahling, 1978). Dahlgren (1975), Cronquist (1981), Thorne (1992), and Takhtajan (1997) all considered Garryaceae a closely related family to Cornaceae.

Uncertainty over the systematic position of Garryaceae prompted several morphological and anatomical studies over the late nineteenth and first half of the 20th century. Much attention focused on the interpretation of the pistillate flower. An inferior ovary would be consistent with Cornaceae and related families, while a superior ovary would not conflict with the heterogeneous “Amentiferae”.

Garrya was originally described as having an inferior ovary (Lindley, 1834), with the adnate ovary appendages interpreted as sepals or a rudimentary perianth. Baillon (1877), Faure (1924), and Hallock (1930) considered the flowers to be epigynous, whereas Wangerin (1906), Horne (1914), Reeve (1943), and Moseley & Beeks (1955) described the flowers as nude or superior, and interpreted the ovary appendages as bracts. Eyde (1964), in a thorough study of floral vasculature, demonstrated that the ovary appendages observed in subgenus *Garrya* are not homologous with the adnate bracts in terminal flowers of subgenus *Fadyenia*. He concluded that the flowers are epigynous, and thus consistent with a placement of *Garrya* in Cornaceae.

The staminate flowers of *Garrya* have not attracted as much attention. Illustrations of *G. elliptica* Dougl. ex Lindl. in the original publication (Lindley, 1834) and *Die natürlichen Pflanzenfamilien* (Harms, 1898) show no structure internal to the filaments. In contrast, Baillon's illustration of this same species (1877) shows a bilobed structure. As noted by Wangerin (1906), Baillon does not describe this structure in the text. Wangerin (1906) also illustrated this structure in *G. elliptica* and interpreted it as a rudimentary ovary (Ovarrudiment). He further argued that this was evidence of the hypogynous nature of *Garrya* flowers. A bilobed structure is also evident in the *G. elliptica* illustration of Horne (1914). Horne (1914) provides the only apical view of open staminate flowers in other species. In his illustration of a putative hybrid between a male *G. elliptica* and female *G. fadyena* Hook., a bilobed structure is evident. His illustration of *G. laurifolia* Benth. (subg. *Fadyenia*) shows no such

structure. Illustrations in subsequent publications either copy the above, or do not show an apical view of an open staminate flower.

Most descriptions of *Garrya* flowers do not mention an intrastaminal structure. Exceptions are Moseley & Beeks (1955), Kapil & Mohana Rao (1966), and Takhtajan (1997), where it is described as a “rudimentary” ovary, pistil, or gynoeceum, respectively. Kapil & Mohana Rao (1966) and Takhtajan (1997) accept Eyde's (1964) conclusion that the pistillate flower is epigynous, but fail to note a condition that would be otherwise unknown in angiosperms: unisexual flowers that are epigynous in one sex, and hypogynous in the other. In order to resolve this paradox, I examined staminate flowers of three *Garrya* species representing both subgenera.

MATERIALS AND METHODS

Staminate inflorescences of *Garrya elliptica* and *G. veatchii* Kellogg (subgenus *Garrya*) and *G. longifolia* Rose (subgenus *Fadyenia*) were collected and preserved in FAA. Eight herbarium specimens of *G. elliptica* were also examined. Voucher specimens are deposited in the Oregon State University Herbarium (Table 1). For scanning electron microscopy (SEM), specimens were prepared in a critical point dryer (Balzers CPD 020), mounted on aluminum stubs, and coated with a gold/palladium alloy applied using an Edwards S150B sputter coater. Specimens were observed and photographed with an AmRay 3300FE.

RESULTS AND DISCUSSION

The staminate flowers of two species of *Garrya* subgenus *Garrya* possess a small, bilobed structure internal to the four filaments (Figs. 2A–C). This intrastaminal structure is consistently present in multiple flowers of an inflorescence (Table 2). The structure is also visible in all examined herbarium specimens of *G. elliptica*, representing the entire geographic range of this species (Table 1), and in several cultivated individuals (vouchers not preserved). Considering the sister group relationship of *Aucuba* and *Garrya*, I have hypothesized that the intrastaminal structure is in fact a vestigial nectary disc. Thus

Table 2. Nectary disc variation in staminate flowers of *Garrya* species.

Species	Flowers observed	Nectary disc present	length (μm ; mean \pm s.d.)	width (μm ; mean \pm s.d.)
<i>G. elliptica</i>	18	18	274 \pm 33	279 \pm 48
<i>G. longifolia</i>	98	50	248 \pm 178	223 \pm 138
<i>G. veitchii</i>	18	18	371 \pm 62	357 \pm 51

the staminate flowers cannot be considered hypogynous (Wangerin, 1906), and do not contradict the interpretation of the pistillate flowers as epigynous (Eyde, 1964).

There is no evidence that the nectary disc plays a role in *Garrya* pollination. The disc is small, absent in pistillate flowers, and inaccessible to insects due to the united perianth lobes (Fig. 1A). Further evidence for the vestigial nature of the disc is its inconsistent development in male flowers of *G. longifolia* (Fig. 2D–F, Table 2).

The consistent presence of an intrastaminal structure in *G. elliptica* suggests that the original illustration (Lindley, 1834) was inaccurate. Subsequent authors who have illustrated the structure have either misinterpreted it (Wangerin, 1906), or not commented on its significance (Baillon, 1877; Horne, 1914). The discrepancies in illustrations of subgenus *Fadyenia* species (Wangerin, 1906; Horne, 1914) apparently reflect the inconsistent development of the intrastaminal structure in this subgenus.

In his revision of *Garrya*, Dahling (1978) wrote “The presence of a degenerate or obsolete ovary within the male flowers has been periodically reported, but my own observations have not verified this”. Although best observed in preserved material, the intrastaminal structure is visible in rehydrated flowers from herbarium specimens. Perhaps the implausibility of an hypogynous staminate flower led Dahling to discount evidence for the presence of an “obsolete ovary”.

It is difficult to understand why previous investigators had not homologized the intrastaminal structures of *Aucuba* and *Garrya*, when a relationship between the two genera was long suspected. One explanation is that floral nectaries have been generally neglected in plant systematics (Chesselet & al., 2002). Lorch (1972) recounts the troubled history of the nectary, and the reluctance of plant morphologists to accept it as a legitimate organ. This attitude may be responsible for its absence from many plant systematic studies.

In this context, it is noteworthy that Wangerin (1906), working in the typological tradition of plant morphology, is the only previous author to have considered the possibility that the intrastaminal structure in *Garrya* represents a vestigial nectary disc. However, he rejected this interpretation for two reasons. First, there is no evidence of a disc in the pistillate flowers of *Garrya*. In *Aucuba* the nectary disc is prominent in the flowers of both sexes, as expected since the nectary is functional in

this entomophilous genus (Abe, 2001). But in the wind-pollinated *Garrya*, the nectary is nonfunctional, and there is nothing to keep it from being lost (in the pistillate flowers) or much reduced (in the staminate flowers). Wangerin’s second argument is based on his observation of a mutation in some staminate flowers, where the “rudimentary ovary” is transformed into one or two extra stamens. He states (translated from the German): “Undoubtedly these occurrences are clear proof of the phylome-nature of the structure, and speak absolutely against their interpretation as a rudimentary disc”.

The idea that sterile and fertile floral structures are fundamentally different has been long discredited and is clearly rejected by molecular developmental findings (Coen & Meyerowitz, 1991).

Conclusions. — The interpretation of the intrastaminal structure as a vestigial nectary resolves a paradox that has existed in the taxonomic literature for over a century. These results were obtained from mature flowers of *Garrya*. Comparative studies focused on the earliest stages of floral development are still required to resolve the homology of the perianth parts in *Garrya*. Additional species of *Garrya* should also be examined for variation in the presence/absence of the intrastaminal structure.

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LITERATURE CITED

- Abe, T. 2001. Flowering phenology, display size, and fruit set in an understory dioecious shrub, *Aucuba japonica* (Cornaceae). *Amer. J. Bot.* 88: 455–461.
- Baillon, H. E. 1877. Organogénie florale des *Garrya*. *Compt. Rend. Assoc. Franç. Avancem. Sci.* 1877: 561–566. [Reprinted in *Adansonia* 12: 262–269. 1879.]
- Bate-Smith, E. C. 1962. The phenolic constituents of plants and their taxonomic significance. *J. Linn. Soc., Bot.* 58: 95–173.
- Bate-Smith, E. C., Ferguson, I. K., Hutson, K., Jensen, S.

- R., Nielsen, B. J. & Swain, T. 1975. Phytochemical inter-relationships in the Cornaceae. *Biochem. Syst. Ecol.* 3: 79–89.
- Bremer, B., Bremer, K., Heidari, N., Erixon, P., Olmstead, R. G., Anderberg, A. A., Källersjö, M. & Barkhordarian, E. 2002. Phylogenetics of asterids based on 3 coding and 3 non-coding chloroplast DNA markers and the utility of non-coding DNA at higher taxonomic levels. *Molec. Phylog. Evol.* 24: 274–301.
- Chesselet, P., Smith, G. F. & van Wyk, A. E. 2002. A new tribal classification of Mesembryanthemaceae: evidence from floral nectaries. *Taxon* 51: 295–308.
- Coen, E. S. & Meyerowitz, E. M. 1991. The war of the whorls: genetic interactions controlling flower development. *Nature* 353: 31–37.
- Cronquist, A. 1981. *An Integrated System of Classification of Flowering Plants*. Columbia Univ. Press, New York.
- Dahlgren, R. M. T. 1975. A system of classification of the angiosperms to be used to demonstrate the distribution of characters. *Bot. Notiser* 128: 119–147.
- Dahling, G. V. 1978. Systematics and evolution of *Garrya*. *Contrib. Gray Herb. Harvard Univ.* 209: 1–104.
- Eramian, E. N. 1971. Palynological data on the systematics and phylogeny of Cornaceae Dumort. and related families. Pp. 235–273 in: Kuprianova, L. A. & Yakovlev, M. S. (eds.), *Pollen Morphology of Cucurbitaceae, Thymelaeaceae, Cornaceae*. Nauka, Leningrad.
- Eyde, R. H. 1964. Inferior ovary and generic affinities of *Garrya*. *Amer. J. Bot.* 51: 1083–1092.
- Faure, A.-L. 1924. *Étude Organographique Anatomique et Pharmacologique de la Famille de Cornacées*. Ph.D. Thesis, Fac. Méd. Pharm., Lille.
- Hallock, F. A. 1930. The relationship of *Garrya*. The development of the flowers and seeds of *Garrya* and its bearing on the phylogenetic position of the genus. *Ann. Bot.* 176: 771–812.
- Harms, H. 1898. Cornaceae. Pp. 256–257 in: Engler, A. & Prantl, K. (eds.), *Die natürlichen Pflanzenfamilien*, vol. 3 (8). Wilhelm Engelmann, Leipzig.
- Herisse, H. & Lebas, C. 1910. Présence de l'aucubine dans plusieurs espèces du genre *Garrya*. *Beih. Bot. Centralbl.* 117–176.
- Hooker, J. D. 1867. Cornaceae. P. 951 in: Bentham, G. & Hooker, J. D. (eds.), *Genera Plantarum*, vol. 1(3). Reeve, London.
- Horne, A. S. 1914. A contribution to the study of evolution of the flower with special reference to the Hamamelidaceae, Caprifoliaceae, and Cornaceae. *Trans. Linn. Soc. London, Bot.* 8: 239–309.
- Kapil, R. N. & Mohana Rao, P. R. 1966. Studies of the Garryaceae II. Embryology and systematic position of *Garrya Douglas ex Lindley*. *Phytomorphology* 16: 564–578.
- Kårehed, J. 2001. Multiple origin of the tropical forest tree family Icacinaceae. *Amer. J. Bot.* 88: 2259–2274.
- Kleiman, R. & Spencer, G. F. 1982. Search for new industrial oils. XVI. Umbelliflorae-seed oils rich in petroselinic acid. *J. Amer. Oil Chem. Soc.* 59: 29–38.
- Lindley, J. 1834. *Garrya elliptica*. *Edward's Bot. Reg.* 20: 1686.
- Lindley, J. 1846. *Vegetable Kingdom*. Bradbury & Evans, London.
- Lorch, J. 1978. The discovery of nectar and nectaries and its relation to views on flowers and insects. *Isis* 69: 514–533.
- Moseley, M. F., Jr. & Beeks, R. M. 1955. Studies of the Garryaceae I. The comparative morphology and phylogeny. *Phytomorphology* 5: 314–346.
- Reeve, R. M. 1943. Comparative ontogeny of the inflorescence and the axillary vegetative shoot in *Garrya elliptica*. *Amer. J. Bot.* 30: 608–619.
- Roth, W. B., Carr, M. E., Davis, E. A. & Bagby, M. O. 1985. New sources of gutta-percha in *Garrya flavescens* and *Garrya wrightii*. *Phytochemistry* 24: 183–184.
- Soltis, D. E., Soltis P. S., Chase, M. W., Mort, M. E., Albach, D. C., Zanis, M., Savolainen, V., Hahn, W. H., Hoot, S. B., Fay, M. F., Axtell, M., Swensen, S. M., Price, L. M., Kress, W. J., Nixon, K. C. & Farris, J. S. 2000. Angiosperm phylogeny inferred from 18S rDNA, *rbcL*, and *atpB* sequences. *Bot. J. Linn. Soc.* 133: 381–461.
- Takhtajan, A. 1997. *Diversity and Classification of Flowering Plants*. Columbia Univ. Press, New York.
- Thorne, R. F. 1992. Classification and geography of the flowering plants. *Bot. Rev.* 58: 225–348.
- Wangerin, W. 1906. Die Umgrenzung und Gliederung der Familie der Cornaceae. *Bot. Jahrb. Syst.* 18: 1–88.