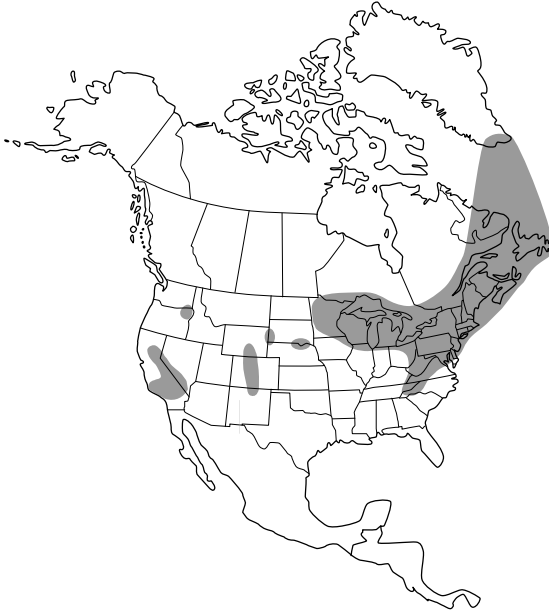


Botrychium simplex

| | |
|---------------------|--|
| Family: | Ophioglossaceae |
| Genus: | <i>Botrychium</i> |
| Species: | <i>Botrychium simplex</i> E. Hitchcock |
| Varieties: | <i>simplex</i> , <i>compositum</i> |
| Common Name: | Least Moonwort |
| Ploidy: | Diploid |



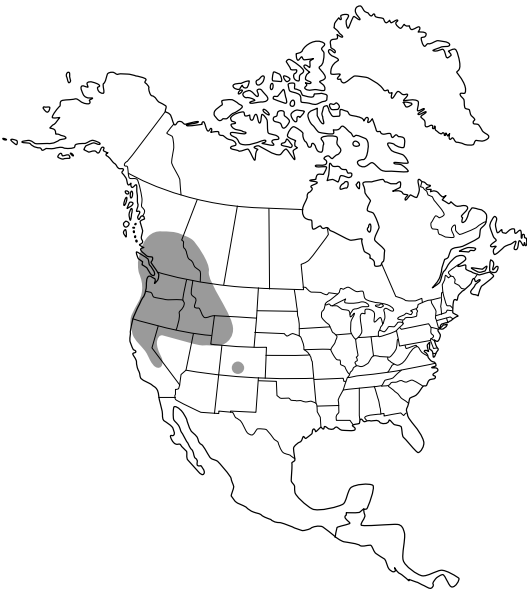
map = var. *simplex*

Technical description: Trophophore stalk 0-3 cm, 0-1.5 times length of trophophore rachis; blade dull to bright green to whitish green, linear to ovate oblong to oblong to fully triangular with pinnae arranged ternately, simple to 2(-3) pinnate, to 7 x 0.2 cm, fleshy to thin, papery or herbaceous. Pinnae or well-developed lobes to 7 pairs, spreading to ascending, approximate to widely separated, distance between 1st and 2nd pinnae frequently greater than between 2nd and 3rd pairs, basal pinna pair commonly much larger and more complex than adjacent pair, cuneate to fan-shaped, strongly asymmetric, undivided to divided to tip, basispic margins \pm perpendicular to rachis, acroscopic margins strongly ascending, basal pinnae often divided into 2 unequal parts, margins usually entire or shallowly sinuate, apex rounded, undivided and boat-shaped to strongly divided and plane, venation pinnate or like ribs of fan, with midrib. Sporophores mainly 1-pinnate, 1-8 times length of trophophores, $2n = 90$. (Wagner and Wagner 1993).



B. simplex var. *simplex*

B. simplex var. *compositum*



Small plant with lower pinnae undivided



Unusually large plant with 1st and 2nd pinna pair divided.

Taxonomy

Botrychium simplex was described by E. Hitchcock in 1823 (Amer. J. Sci. Arts 6:103) based on plants from Massachusetts. Following publication of the species, many botanists called attention to the conspicuous variation within the species. Clausen (1938) treated this variation by recognizing four varieties:

var. *typicum* (= var. *simplex*) for plants with undivided basal pinnae and trophophore stalk equal to or exceeding length of the common stalk; found in open habitats throughout the range of the species in North America, Europe and Japan;

var. *compositum* for plants with secondarily divided basal pinnae and trophophore stalk equal to or exceeding length of the common stalk; found in meadows, primarily in the western US and sparingly in the eastern US and Europe;

var. *laxifolium* for plants with undivided basal pinnae and trophophore stalk shorter than the common stalk; found in dry deciduous woods in New England; and

var. *tenebrosum* for slender plants with undivided basal pinnae and very short trophophore stalk; found in damp heavily shaded woods and edges of swamps from New England to Minnesota with possible occurrences in Washington and Europe (one collection each).

Wagner and Wagner (1993) attributed the variation in *B. simplex* primarily to environmental forms and juvenile stages but recognized possible genetically based differences between plants of eastern and western North America, the western plants differing primarily by a longer sporophore stalk and a very short common stalk, the latter often not extending above ground level. Wagner and Wagner (1993) considered var. *tenebrosum* as possibly a persistent juvenile form.

A genetic comparison of the eastern varieties of *B. simplex* using enzyme electrophoresis was conducted by Farrar (1997). This study found no support for recognition of variety *laxifolium*, all plants examined (96 total from Minnesota, Michigan and North Carolina) fell into one of two basic genotypes corresponding to *B. simplex* var. *tenebrosum* and *B. simplex* var. *simplex*. The genetic identity (GI) value between these varieties was 0.67, a GI value similarities to that between other *Botrychium* species such as *B. lunaria* var. *crenulatum* and *B. neolunaria* (GI = 0.70), and near the average value of 0.67 for flowering plant species of the same genus. On this basis we conclude that *B. tenebrosum* warrants recognition as a distinct species as originally described by A. A. Eaton in 1889.

Clausen (1938) recognized *B. simplex* var. *simplex* as the common variety in eastern North America and var. *compositum* as the common variety in western North

America. However, he also recognized var. *simplex* as present in the far western U.S. and annotated a number of California specimens as such. Our studies in California have likewise confirmed the presence of two distinct western varieties of *B. simplex* that differ significantly in morphology, habitat and in allelic composition. The most common, variety *compositum*, is a plant of open, seasonally dry meadows, with morphology as described for the variety. The second variety is a plant of more or less permanently saturated hard water seeps and fens. This variety includes most, if not all, of the plants annotated by Clausen as western plants of variety *simplex*, and these plants do resemble eastern var. *simplex* in morphology more than var. *compositum*. Although they differ somewhat from eastern var. *simplex* in some morphological characters, in habitat, and in allelic composition, pending further study, we are recognizing these plants as *B. simplex* var. *simplex*.

Identification

Because of the substantial differences among and even within the varieties of *B. simplex*, it is difficult to specify morphological traits separating all its forms from all other moonworts. *B. simplex* plants, especially of var. *compositum*, are the only moonworts with a sporophore-trophophore junction near ground level, but eastern plants of var. *simplex* may have the junction midway along the combined length of the sporophore + common stalk, and this may be the case in both varieties in juvenile and in shade-grown plants. The sporophore in all varieties tends to be long and lax with uncrowded branches. The sporophore stalk commonly exceeds the length of the entire trophophore, sometimes greatly so. The trophophore stalk is also prominent, its length usually exceeding the length between the first two pairs of pinnae which, in turn, is much greater than the distance between the second and third pinna pairs. Upper pinnae are strongly ascending with a broad and decurrent attachment to the rachis. Outer pinna margins are rounded as are their junctions with side margins, giving the pinnae a more rounded appearance than those of most other moonwort species. Spores of *B. simplex* are significantly larger than those of most other diploid species.

Differences between the two varieties are subtle. Basal pinnae of var. *simplex* are more broadly attached, more strongly decurrent, more rounded and more glaucous than are those of var. *compositum* (see photos). Enlarged and pinnately divided lower pinnae is not a consistent difference between var. *simplex* and var. *compositum*. Although var. *compositum* expresses this condition more regularly and at smaller sizes than does var. *simplex*, very large plants of var. *simplex* throughout its range produce enlarged, dissected basal pinnae.

Shade grown plants of *B. simplex* without divided or elongated basal pinnae resemble plants of *B. montanum* and its eastern relative *B. mormo* in some characteristics, but these species have a stiffer and stouter, more succulent appearance than *B. simplex* and their pinnae have straight, more or less toothed outer margins. More robust plants of *B. simplex* with undivided, un-elongated basal

pinnae can be distinguished from *B. neolunaria* and similar species by their broad and decurrently attached upper pinnae, non-angular junction of outer and side margins, and strongly stalked trophophore.

Plants of *B. simplex* with divided or elongated basal pinnae can be distinguished from *B. pumicola* by the much shorter trophophore and sporophore stalk of the latter. Such plants can be differentiated from other twice dissected moonworts and from small plants of grapefern species (genus *Sceptridium*) by their un-elongated, undissected pinnae above the first pair.

A major problem in identifying and studying the relationships among varieties of *B. simplex* is their tendency to persist in "juvenile" forms. These are not truly juvenile plants, but especially in drier habitats, plants remain very small (often less than 2 cm) with undivided trophophores or with only a single pair of pinnae below the undivided apical portion. Both varieties, at this size, have more or less the same appearance. Morphologies typical of the variety develop only on larger plants. Thus varietal identification of "juvenile" forms is dependent on presence of some larger plants in the population or use of chemical/molecular methods. However, even "juvenile" forms of *B. simplex* can be distinguished from other species by their relatively undivided, often boat-shaped terminal segments.

Distribution and Abundance

Botrychium simplex is one of the most widely distributed of moonwort species. It occurs across the northern US and southern Canada and southward at high elevations in the Appalachian Mountains to North Carolina and in western mountains south to New Mexico and southern Nevada and California. In the Midwest, it extends further southward (to Iowa and Nebraska) than any other species except *B. campestre*. From southeastern Canada it extends northward to Newfoundland and occurs in southern Greenland, Iceland and Europe. In western North America it occurs in the southern parts of the Canadian provinces and extends up the Pacific coast to southeastern Alaska. It is apparently rare in north central Canada but has been reported from the Northwest Territories.

B. simplex var. *simplex* is widespread in eastern North America, extending from North Carolina to Canada, Greenland and Iceland. This is also the varietal designation given to Scandinavian, European and Japanese plants, but recent investigations indicate that many of the European plants (especially in Iceland and Greenland) designated as *B. simplex* are *B. tenebrosum*. *B. simplex* extends westward across the northern Great Plains to the eastern Rocky Mountains from New Mexico to Wyoming. Most plants of western Montana, Washington, Oregon and California are var. *compositum*, but var. *simplex* also occurs in southeast Washington and northeast Oregon, and is the most common variety in southern California.

B. simplex var. *simplex*
In the far west



Large plant with pinnately divided lower pinnae.



Smaller plant with enlarged but not divided lower pinnae. Sporophore wilted.

B. simplex var. *simplex* is the common variety in the San Gabriel and San Bernadino Mountains and in the White Mountains. Habitat descriptions indicate that all of these sites are under the influence of hardwater seeps and springs. Such habitats also support variety *simplex* in Alpine, Eldorado, Fresno, Mariposa, Mono, Nevada and Tuolumne Counties, all within the range of the more common variety *compositum*. The nature of variety *simplex* habitats in Butte and Tehama Counties is not clear, but the presence of many other moonwort species in this area suggests presence of soils different from those supporting variety *compositum* in the central and southern Sierra Mountains.

In western Montana, Idaho, Washington, Oregon and California, *Botrychium simplex* var. *compositum* is often the most common and frequently the only species of moonwort present at a site, particularly in the southwestern parts of its range in alluvial meadows of the high Sierra Mts. Variety *simplex* and other moonwort species are absent in these meadows derived from granitic bedrock and present only locally in sites influenced by hardwater seeps.

B. simplex var. *compositum* is the most widespread *Botrychium* species in California, occurring in 18 counties containing the principal Sierra Nevada Mountain Range. Plants likely of this variety but needing genetic confirmation have also been collected in Modoc and Colusa Counties.

Habitat

Botrychium simplex is primarily a plant of open habitats, occurring in pastures, meadows, orchards, prairies, wetlands, fens, sand dunes and in lake and stream edge vegetation. Most of these habitats are at least temporarily wet and some (fens) are permanently saturated. Most plants in permanently wet fen and hardwater seep habitats in western North America are var. *simplex*. Within these habitats, plants may be among sparse vegetation and fully exposed to the sun, or they may grow among tall dense herbaceous vegetation, receiving only very low light. Plants in the latter can be detected only by parting the vegetation in diligent search on hands and knees. *B. simplex* is a frequent companion of *B. multifidum*.

Population Genetics

The genetic differentiation within *Botrychium simplex* is discussed above with regard to recognition of varieties. It is important to recognize that genetic variability within each of the varieties is lower than in the species as a whole. Within populations of var. *simplex*, and in Washington and Oregon populations of var. *compositum*, genetic variability is low and similar to that of other moonwort species. Within California, variability among and within populations of var. *compositum* is higher than that found in nearly all other moonwort species. A plausible

interpretation of these patterns of genetic variability is that California was the site of origin of var. *compositum*, and continues to be an area of differentiation. Why and how this species, especially var. *compositum*, has remained evolutionarily more dynamic than other moonworts is not clear. *Botrychium simplex* does, however, provide a window of insight into how differentiation of ancestral forms may have produced the current suite of diploid moonwort species.

Gene Flow and Migration

The genetic variability within western *B. simplex* permits deeper investigation of population interaction than is possible with other moonwort species. In particular, allelic differences among populations allow study of the efficiency of spore dispersal and species migration.

In populations in the Sierra Nevada range of California, populations of *B. simplex* var. *compositum* display great variability in allelic composition, with several alleles restricted to one or a few populations. Such differences would not likely be maintained among populations with unrestricted inter-population migration. Limited migration is further demonstrated in an intensive sampling of a large metapopulation within a 2 x 0.5 mile meadow south of Lake Tahoe. Among 14 subpopulations separated by a minimum of 50 meters, extreme differentiation among populations is present. This differentiation includes unique alleles and sharply different frequencies in common alleles between populations less than 100 M apart, demonstrating restriction of spore dispersal even though the plants grow in short meadow vegetation with their sporophores elevated above the vegetation. Such restriction of migration indicates that populations more than a few miles apart may be effectively isolated and that suitable uncolonized habitats at these distances have a low probability of receiving sufficient spore rain to assure colonization.

Phylogenetic Relationships

Botrychium simplex is most closely related to *B. montanum*, *B. mormo*, and *B. pumicola*. Among genetic differences between these species are alleles unique to each species and overall genetic differentiation expected between species (but see discussion of *B. montanum* vs *B. mormo*).

Botrychium simplex has not participated widely in hybridization with other moonworts or production of allotetraploid species. Putatively, *Botrychium simplex* has combined with *B. pallidum* in Montana to produce *B. "adnatum"* (currently under investigation), known only from the western side of Glacier National Park in Montana.