

## Trichome Morphology of *Cimicifuga* L. (Ranunculaceae) and Its Taxonomic Significance

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Trichomes found in the genus *Cimicifuga* were examined with scanning electron microscopy. Four basic types of trichomes were found in the genus; (1) filiform unicellular trichomes, (2) saccate unicellular trichomes, (3) pyriform unicellular trichomes, and (4) uniseriate multicellular trichomes. All the taxa examined develop at least two different types of trichomes, and many taxa are distinguished by differences in type, microornamentation, density, and position of trichomes. In addition, trichome features appear to be very useful in recognizing species relationships in the genus.

**Keywords :** Ranunculaceae, *Cimicifuga*, trichome morphology, microstructure, species relationships

*Cimicifuga* L. (Ranunculaceae) is a taxonomically difficult and variable genus of ca. 18 species (Tamura, 1966, 1990). Plants of *Cimicifuga* are perennial, erect herbs which usually occupy shady, moist habitats in rich mountain woods. They are readily distinguished by their large ternately compound leaves that are long-petioled, racemose or paniculate inflorescences bearing many small flowers, and follicular fruits with four to 15 seeds (Ramsey, 1965; Tamura, 1966, 1990; Lee and Park, 1994). The species of *Cimicifuga* are distributed in temperate to cold temperate regions of the Northern Hemisphere including Asia, Europe, and North America; with the greatest concentration of species in eastern Asia. In Korea, six taxa are found usually in the shade of moist, deciduous forests, in ravines, and on mountain slopes. In addition, many of the species have long been cultivated for medicinal and ornamental purposes in Europe and Asia (Bailey, 1949; Ramsey, 1965; Hsiao, 1979; Bensky *et al.*, 1986).

Although *Cimicifuga* was monographed by Huth (1892) and more recently by Ramsey (1965), taxonomic relationships and delimitations of many species in the genus are still uncertain due to the extreme

morphological variation and overlapping of major distinguishing characters (Lee and Park, 1994). In addition, many new taxa were recently described from China (Wang and Hsiao, 1965, 1980), but their taxonomic identities have not been critically examined.

As part of a comprehensive systematic study on the genus *Cimicifuga*, trichomes found in 18 taxa of the genus were examined using a scanning electron microscope (SEM) in an attempt to provide more insight into the systematics of the genus. The main purpose of this paper is to describe the microstructure of trichomes found in the genus *Cimicifuga* and to discuss their taxonomic usefulness.

### MATERIALS AND METHODS

Eighty-five individuals representing 18 taxa of *Cimicifuga* (Table 1) were examined for trichome microstructure with scanning electron microscopy. For *C. foetida* L. and its closely related taxa, we follow the delimitation previously proposed by the authors on the basis of numerical analyses of major morphological characters (Lee and Park, 1994). For SEM, trichomes on stems, leaves, peduncles, and follicles were examined using fresh samples fixed in FAA or segments removed from herbarium speci-

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**Table 1.** Taxonomic distribution of trichome types in the genus *Cimicifuga*. 1a, filiform unicellular trichome with micropapillae on the surface; 1b, filiform unicellular trichome with smooth surface; 2a, saccate unicellular trichome with micropapillae on the surface; 2b, saccate unicellular trichome with smooth surface; 3, pyriform unicellular trichome; 4, uniseriate multicellular trichome

Taxa	Stem	Leaf		Inflorescence	Pistil	Follicle
		Adaxial surface	Abaxial surface			
Eurasian taxa						
<i>C. biternata</i> (Siebold & Zucc.) Miquel	(1a)	1a	1a	2a	—	—
<i>C. brachycarpa</i> Hsiao	—	1a	1a	2a	2a	2a, 3
<i>C. dahurica</i> (Turcz. ex Fischer & Meyer) Maxim.	(1a)	—	1a	2b	—	—
<i>C. foetida</i> L.						
var. <i>foetida</i>	3	—	1a, 3	2a, 3	2a, 3	2a, 3
var. <i>micrantha</i> Zapalowicz	(1a)	—	1a	2a	2a	2a, 3
var. <i>velutina</i> Franchet	1a	1a	1a	2a	2a	2a, 3
<i>C. frigida</i> Royle	(1b)	(1b)	1b, (3)	2b	2b	2b, 3
<i>C. heracleifolia</i> Komarov						
var. <i>heracleifolia</i>	(1a)	—	1a	2a	—	—
var. <i>bifida</i> Nakai	—	—	(1b), 3	2b, (3)	(3)	3
<i>C. japonica</i> (Thunb.) Spreng.	(1a)	1a	1a	2a	—	—
<i>C. simplex</i> (DC.) Turcz.	(1a)	(1a)	1a	2a	2a	2a
<i>C. yunnanensis</i> Hsiao	1b, (3)	1b	1b, 3	2b	2b	2b, 3
North American taxa						
<i>C. americana</i> Michx.	—	—	1b	2b	—	—
<i>C. arizonica</i> Watson	—	—	3, 4	4	(3)	3
<i>C. elata</i> Nutt.	4	—	4	3, 4	3	3
<i>C. laciniata</i> Watson	—	—	1b	2b	2b	2b
<i>C. racemosa</i> (L.) Nutt.	—	1a	1a	2a	—	—
<i>C. rubifolia</i> Kearney	—	—	3, 4	4	—	—

mens. In the case of segments taken from herbarium specimens, they were first rehydrated by soaking them in warm water for 12 hours. All samples were then dehydrated in a graded series of acetone concentrations, followed by critical-point-drying in a Polaroid Model E3000 with liquid carbon dioxide. Dried samples mounted on aluminum stubs were coated with gold using an Eiko Model IB-3 ion coater at 6 mA for three minutes, and examined and photographed using an Akashi Model SX-40 SEM. In addition, over 1,700 specimens of *Cimicifuga* from the major herbaria of the world were examined with light microscopy for inter- and intra-specific variation in trichome morphology and distribution. A list of specimens examined for this study is available from the authors.

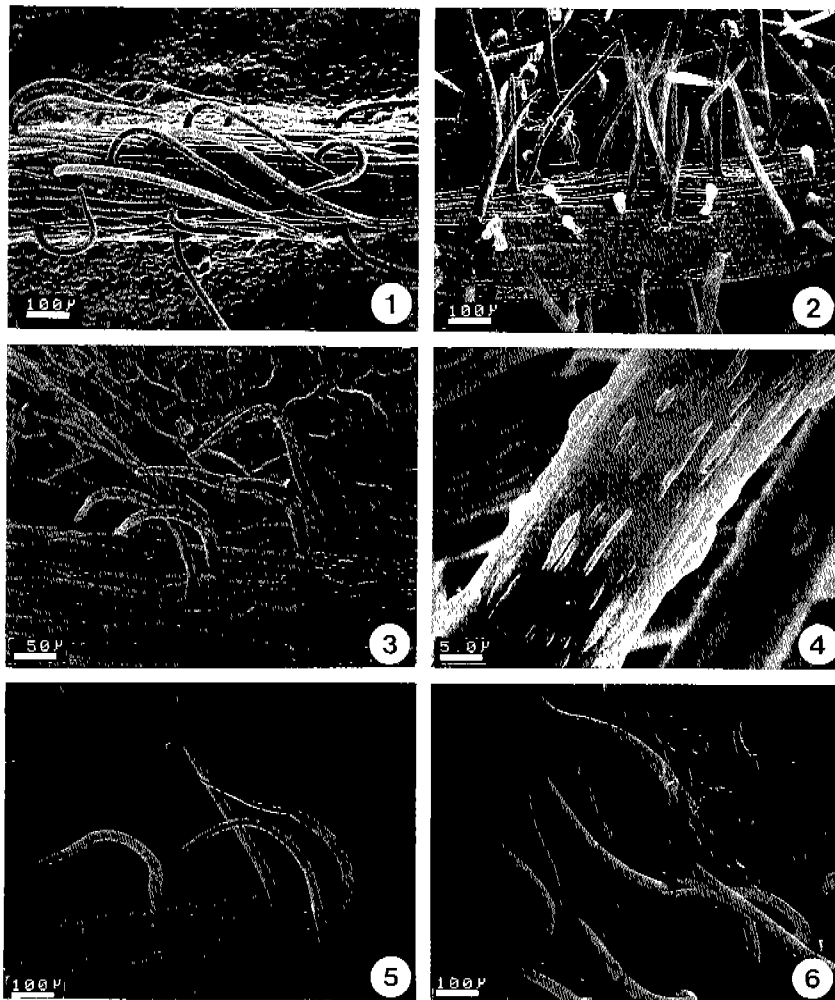
## RESULTS AND DISCUSSION

Representative trichomes encountered in *Cimici-*

*fuga* are shown in Figs. 1-18. Four basic types of trichomes occur in the genus: Type 1-filiform unicellular trichomes (Figs. 1-6); Type 2-saccate unicellular trichomes (Figs. 7-12); Type 3-pyriform unicellular trichomes (Fig. 13); and Type 4-uniseriate multicellular trichomes (Figs. 14-18). All trichomes are simple and unbranched. In some taxa, micropapillae with varying length (Figs. 4, 9) are present on the surface of filiform and saccate unicellular trichomes. All taxa develop at least two different types of trichomes, and Table 1 summarizes the taxonomic distribution of trichome types in the genus *Cimicifuga*.

Trichome type is considered to be one of the most informative characters in assessing phylogeny in the Ranunculaceae (Hoot, 1991). In *Cimicifuga*, trichome morphology and distribution of trichome types on stems, leaves, inflorescences, pistils, and follicles appear to be taxonomically useful (Table 1).

Filiform unicellular trichomes (trichome type 1; Figs. 1-6) are found in all taxa in the genus except

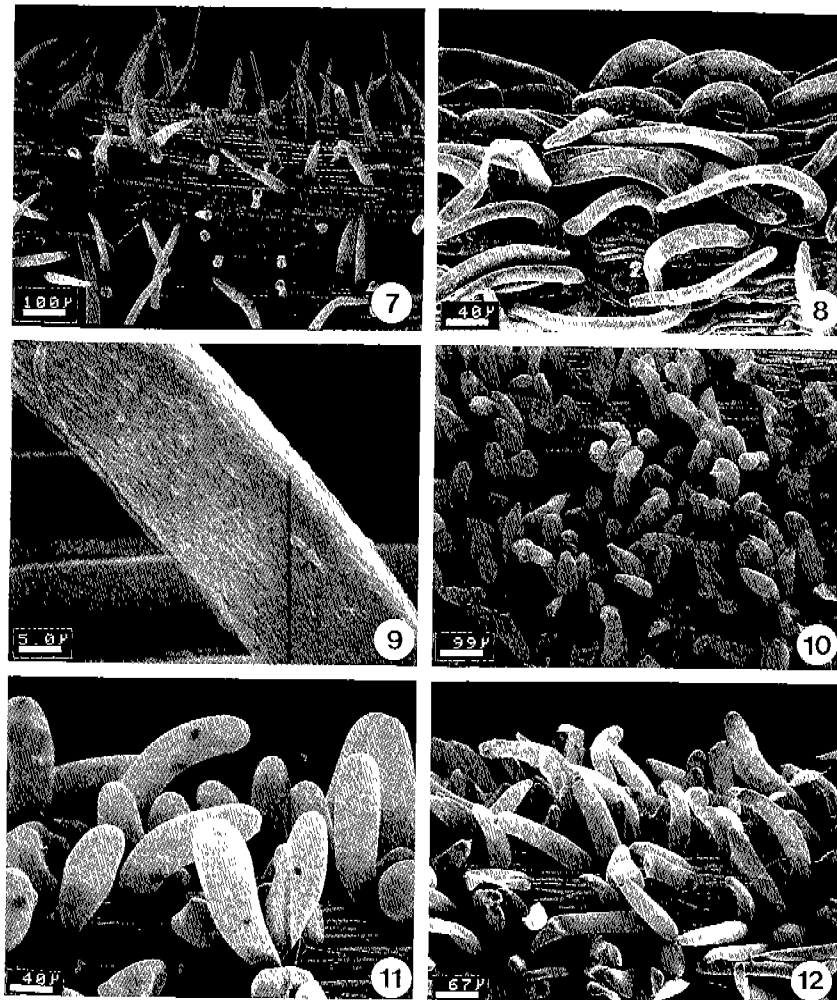


**Figs. 1-6.** Filiform unicellular trichomes (type 1) on leaf veins (abaxial surface) of representative taxa of *Cimicifuga*. 1. Type 1a, *C. dahurica*. 2. Type 1a, *C. foetida* var. *foetida*. 3. Type 1a, *C. racemosa*. 4. Micropapillae on type 1a trichome, *C. dahurica*. 5. Type 1b, *C. americana*. 6. Type 1b, *C. laciniata*. Trichome type numbers correspond to those in Table 1.

*C. arizonica* Watson, *C. elata* Nutt., and *C. rubifolia* Kearney. They are mainly distributed on the surface of major leaf veins and petioles, but found on stems and leaf blades also (Table 1). The length ranges from 0.1 to 0.8 mm, and is somewhat variable depending on the position. The longest ones are generally found along major leaf veins. In most taxa, micropapillae (Fig. 4) are present on the surface of filiform unicellular trichomes (Figs. 1-3), but they are absent in *C. americana* Michx. (Fig. 5), *C. frigida* Royle, *C. heracleifolia* Komarov var. *bifida* Nakai, *C. laciniata* Watson (Fig. 6), and most *C. yunnanensis* Hsiao (Table 1).

Saccate unicellular trichomes (trichome type 2; Figs. 7-12) are found in most taxa, and their occurrence is mainly restricted to the surface of peduncles,

pedicels, pistils, and follicles (Table 1). They are usually 0.1-0.4 mm long, and show intraspecific variation in length to some extent. The shape and the surface feature of saccate unicellular trichomes show variation among the taxa, and are useful in distinguishing many taxa in the genus. In majority of the taxa, the saccate unicellular trichomes are somewhat lanceolate with acute to acuminate tips (Figs. 7, 8), but in *C. americana* (Fig. 10), *C. dahurica* (Turcz. ex Fischer & Meyer) Maxim. (Fig. 11), *C. frigida*, and *C. laciniata* (Fig. 12), they are narrowly elliptic to oblong with obtuse to round tips. In addition, they are almost straight and spreading in *C. foetida* var. *foetida* (Fig. 7), whereas they are somewhat appressed and curved toward the apex in the rest of the taxa (Figs. 8, 10-12). As in filiform unicellular trichomes,



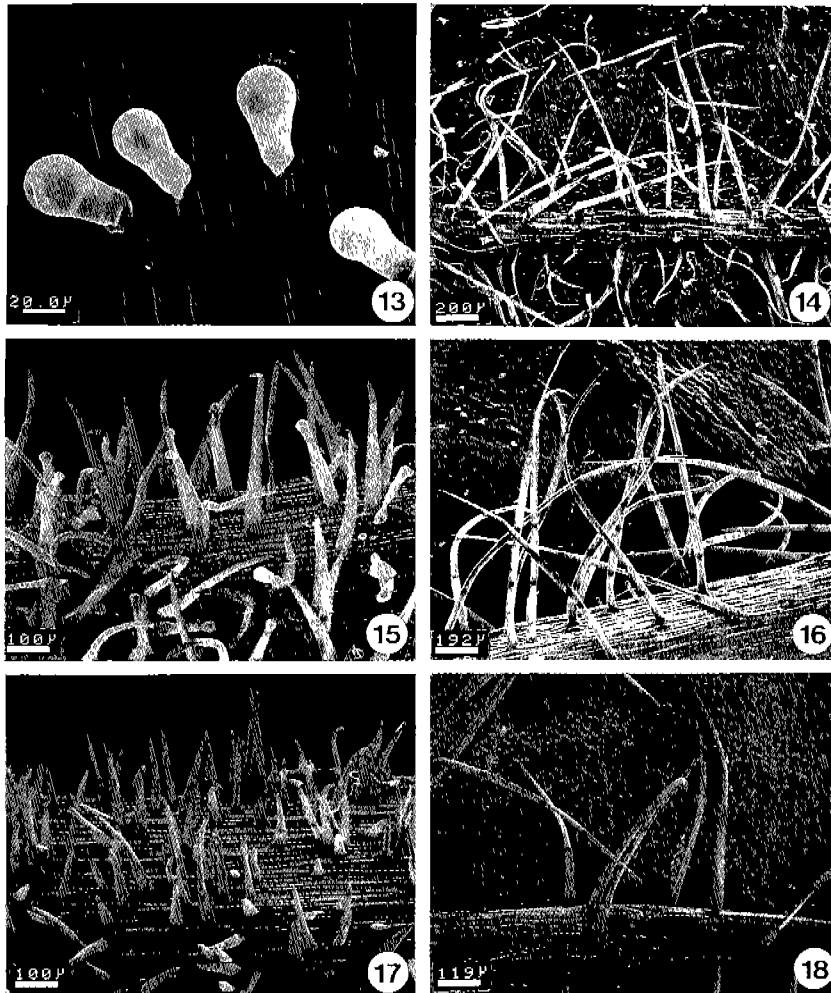
**Figs. 7-12.** Saccate unicellular trichomes (type 2) on inflorescences of representative taxa of *Cimicifuga*. 7. Type 2a, *C. foetida* var. *foetida*. 8. Type 2a, *C. racemosa*. 9. Micropapillae on type 2a trichome, *C. simplex*. 10. Type 2b, *C. americana*. 11. Type 2b, *C. dahurica*. 12. Type 2b, *C. laciniata*. Trichome type numbers correspond to those in Table 1.

micropapillae (Fig. 9) are present on the surface of saccate unicellular trichomes in majority of the taxa. In general, the taxa with smooth filiform unicellular trichomes also tend to have smooth saccate unicellular ones (Table 1). The only exception is *C. dahurica*; it has micropapillate filiform unicellular trichomes (Figs. 1, 4), but has smooth saccate unicellular ones (Fig. 11).

Pyriform unicellular trichomes (trichome type 3) have conspicuously swollen bulbous tips, and are smooth on the surface (Fig. 13). They are found in 10 taxa in the genus, and usually distributed on the surface of pistils and follicles. However, in some taxa, they also occur on the surface of stems, major leaf veins, and inflorescences (Table 1). In particular, they are moderately to densely distributed on stems,

major leaf veins, petioles, bracteoles, and follicles of *C. foetida* var. *foetida* (Figs. 2, 13; Table 1). In most taxa, pyriform unicellular trichomes are relatively short and seldom exceed 0.1 mm in length, but in *C. elata*, they are much longer and usually 0.1-0.25 mm long (Fig. 15).

Especially noteworthy is the occurrence of uniseriate multicellular trichomes (trichome type 4; Figs. 14-18) in three North American species, *C. arizonica*, *C. elata*, and *C. rubifolia* (Table 1). Hoot (1991), in her attempt to elucidate phylogeny of the Ranunculaceae, examined trichomes of 107 species, including six *Cimicifuga* species (the list of species examined not given), of the Ranunculaceae with scanning electron or light microscopy. However, she did not report any multicellular trichomes for the family, and



**Figs. 13-18.** Pyriform unicellular (type 3) and uniseriate multicellular trichomes (type 4) on representative taxa of *Cimicifuga*. 13. Type 3, stem, *C. foetida* var. *foetida*. 14. Type 4, abaxial surface of leaf vein, *C. elata*. 15. Type 4, peduncle, *C. elata*. 16. Type 4, abaxial surface of leaf vein, *C. rubifolia*. 17. Type 4, peduncle, *C. rubifolia*. 18. Type 4, abaxial surface of leaf vein, *C. arizonica*. Trichome type numbers correspond to those in Table 1.

for *Cimicifuga*, she reported only filiform unicellular trichomes (type 1 trichome by Hoot, 1991) and saccate unicellular trichomes (type 3 trichome by Hoot, 1991).

The uniseriate multicellular trichomes on these species are usually composed of 2-6 cells, and smooth on the surface (Figs. 14-18). They are distributed on the surface of stems, leaves, petioles, and inflorescences (Table 1). Those on stems, leaves, and petioles are relatively long (0.3-1.2 mm long), linear and somewhat curved in shape, and usually composed of 3-6 cells (Figs. 14, 16, 18), whereas those on inflorescences are much shorter (0.1-0.4 mm long), linear lanceolate to lanceolate in shape, straight, spreading, and usually composed of 2-3 cells (Figs.

15, 17). In addition, the uniseriate multicellular trichomes on inflorescences of these species are always intermixed with simple unicellular ones similar in shape (Figs. 15, 17), suggesting that they are probably derived from simple unicellular ones. The length of uniseriate multicellular trichomes shows interspecific variation; the uniseriate multicellular trichomes on major leaf veins of *C. elata* (Fig. 14) and *C. rubifolia* (Fig. 16) are conspicuously longer than those of *C. arizonica* (Fig. 18).

The trichome types and their distribution are also informative in recognizing certain species relationships in the genus. For example, of six species found in North America, *C. arizonica*, *C. elata*, and *C. rubifolia* are readily distinguished from all other species

in the genus in having uniseriate multicellular trichomes on leaves and inflorescences (Figs. 14-18; Table 1). In gross morphology, *C. rubifolia* is similar to *C. elata*, and the close relationship between the two has been suggested by Ramsey (1965); sharing of uniseriate multicellular trichomes in these two species (Figs. 14-17) provides additional evidence of close relationship. *Cimicifuga arizonica* resembles *C. racemosa* (L.) Nutt. in having triternate leaves with deeply trifid leaflets and relatively long pedicels. In contrast, the two species show fundamental differences in trichome type (Table 1). Uniseriate multicellular and pyriform unicellular trichomes found in *C. arizonica* are entirely absent in *C. racemosa*; the latter species possesses micropapillate filiform and saccate unicellular trichomes instead (Figs. 3, 8). Considering the trichome types, in conjunction with other morphological characters including number of bracteoles and sepals, follicle shape, and seed surface features, *C. arizonica* seems to have closer relationship with *C. elata* than with *C. racemosa*.

In trichome morphology, *C. racemosa* (Figs. 3, 8) rather approaches its sympatric species *C. americana* (Figs. 5, 10) and the western *C. laciniata* (Figs. 6, 12) in having filiform and saccate unicellular trichomes. However, it is quite distinct from the latter two species by its small, long-stipitate, narrowly elliptic petals with no nectariferous glands, four sepals and a single sessile pistil per flower, capitate stigmas, and lunate seeds with thin scales along the three edges. It also shows a difference in trichome surface feature; micropapillae present on the trichomes of *C. racemosa* are absent in those of the latter two species (Figs. 5, 6, 10, 12).

*Cimicifuga americana* is similar to *C. laciniata* in most morphological characters (Ramsey, 1965), and these two species have polyforate pollen which is unique in the genus (Lee and Park, unpubl.). They also have identical trichome types represented by smooth filiform and saccate unicellular trichomes except for a slight difference in shape of saccate unicellular ones (Figs. 5, 6, 10, 12; Table 1); the distribution of same trichome types supports the close relationship between the two species.

The trichome types found in the Eurasian taxa are represented by filiform, saccate, and pyriform unicellular trichomes. As in the North American species, many taxa can be distinguished by differen-

ces in type, microornamentation, density, and position of trichomes (Table 1). For example, *C. dahurica* differs from all other species in the genus by its unique combination of trichome types. In this species, the filiform unicellular trichomes on stems and leaves are conspicuously micropapillate, but the saccate unicellular ones on inflorescences are completely smooth on the surface. The species is also very distinctive in having dioecy, a condition not observed in any other taxa in the genus.

*Cimicifuga japonica* (Thunb.) Spreng., centered in southern Korea and Japan, closely resembles its sympatric relative *C. biternata* (Siebold & Zucc.) Miquel in most morphological characters, and the two species have often been confused. However, as noted by Ramsey (1965), *C. japonica* can be reliably distinguished from *C. biternata* by the distribution pattern of filiform unicellular trichomes on adaxial surfaces of leaves. In the former, the filiform unicellular trichomes are distributed uniquely in a narrow zone along the leaf margin, whereas in the latter, they are found on the surface of major veins.

*Cimicifuga simplex* (DC.) Turcz. is a widespread and variable species. Extending from southwestern China through Manchuria, eastern Siberia, and Korea to Japan, the species shows extreme variation in leaf pubescence. In plants from China, Manchuria, and eastern Siberia, adaxial surfaces of leaves are glabrous or very sparsely pubescent with filiform unicellular trichomes along the major veins. However, at the eastern part of its range, which comprises Japan, leaf pubescence becomes far more variable, and shows geographical partitioning to some extent. As noted by Emura (1970), plants from Hokkaido and northern Honshu tend to have leaves with glabrous adaxial surfaces, but in central and southern Honshu, plants densely pubescent on adaxial surfaces of major leaf veins are commonly found. In addition, individuals with sparsely pubescent leaf veins are sporadically distributed throughout the central and southern Honshu. Emura (1970) found a slight difference in karyotype among these forms, and suggested a possible correlation between the karyotype and the leaf pubescence.

In *C. foetida* and related species including *C. brachycarpa* Hsiao, *C. frigida*, *C. heracleifolia*, and *C. yunnanensis*, there is also much variation among species in type and distribution of trichomes (Table 1), and

the usefulness of trichome features in delimiting these closely related species was demonstrated by the authors (Lee and Park, 1994).

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## 승마屬(미나리아재비과) 植物의 털의 形態와 分類

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### 적 요

승마속 식물에 분포하는 털의 미세구조를 주사전자현미경을 사용하여 조사하였다. 그 결과, 본 속 분류군들에는 크게 (1) 비교적 길게 신장되는 선형의 단세포 털, (2) 짧은 주머니 형태의 단세포 털, (3) 구형의 선단부가 존재하는 배모양 단세포 털, (4) 2-6개의 세포가 한줄로 연결된 선형의 다세포 털 등 네 가지 형태의 털이 분포하는 것으로 밝혀졌다. 속내 각 분류군에는 두 가지 이상의 털이 분포하는 것으로 나타났으며, 대부분의 분류군들은 털의 종류, 분포 위치, 밀도 및 털표면의 미세구조 등의 차이에 의해 구분되었다. 또한 털의 형태 및 미세구조는 승마속내 종간 유연관계를 인식하는데 매우 유용한 것으로 밝혀졌다.

주요어: 미나리아재비과, 승마속, 털의 형태, 미세구조, 종간 유연관계

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