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Zhang XX, Zhang HY, Xu WB, *et al*. Leaf epidermal characters and their taxonomic significance in Balsaminaceae, China[J]. Guihaia, 2013, 33(4):528-537

Leaf epidermal characters and their taxonomic significance in Balsaminaceae, China

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Abstract: The leaf epidermal characters of 106 field collections representing 36 species within the family Balsaminaceae, were investigated with light microscopy. Most epidermal characters were constant within species. These characters were valuable in clarifying species circumscriptions and relationships between the two Balsaminaceae genera, *Impatiens* and *Hydrocera*. Although abaxial epidermal characters were found to greatly vary between species, the adaxial epidermis presented much more valuable taxonomic characters. Based on the latter, the species were divided into five groups. Epidermal characters were closely correlated with geographical distribution rather than with gross morphology. Epidermal characters were found useful in demarcating species but had limited value in infrageneric subdivision. Epidermal characters appeared to be heavily modified by different environments.

Key words: Balsaminaceae; *Impatiens*; leaf epidermis; China; taxonomy

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中国凤仙花科植物叶表皮特征及其分类学意义

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摘 要: 利用光学显微镜观察凤仙花科106份样本材料、36种代表植物的叶表皮微形态特征。结果表明:该科植物叶表皮微形态特征种内稳定，对于种间及属间关系的界定具有重要的分类学价值。上表皮细胞的形状及垂周壁的式样种间差异明显，各分类群间有明显的界限，是种间界定的重要依据，因此上表皮微形态特征具有重要的的分类学价值，基于上表皮的微形态特征将研究的36种代表植物划分为5个类型。下表皮的微形态特征虽更为多样，种间差异显著，可用于种间界定，但对于属下划分难以提供有价值的性状。该科植物叶表皮微形态特征与宏观形态特征的相关性较弱，与地理分布格局的相关性较强，关系更为密切。同一地理分布区域内的种类宏观形态特征虽然差别明显，但叶表皮微形态特征却表现出较强的一致性，这似乎也反映了叶表皮微形态这一性状受环境饰变的影响比较明显。综上所述，叶表皮微形态特征可为凤仙花科的系统发育，

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尤其是凤仙花属种间界定提供有价值的分类学佐证。

关键词: 凤仙花科; 凤仙花属; 叶表皮; 中国; 分类学

The Balsaminaceae is comprised of two genera, *Impatiens* and *Hydrocera*. *Impatiens* is a large genus with around 900－1 000 species (Grey-Wilson, 1980; Chen, 2001; Chen *et al*., 2007) distributed all around the world except Australia and south America. *Hydrocera* is a small genus with only 2－3 species (Fischer, 2004) occurring in wet areas in the tropics. The Balsaminaceae is a difficult family in angiosperm taxonomy, because of the large and complex genus, *Impatiens*. The phylogeny of the Balsaminaceae, especially *Impatiens*, and the evolutionary morphologies are not yet well understood. There has been no single acceptable infrageneric taxonomic system in *Impatiens* (Chen, 2001).

Recent studies in molecular systematics indicate that the Balsaminaceae was embedded in the basal clades of the Eriales (Soltis *et al*., 2005). The phylogeny of the Balsaminaceae has been discussed in several papers over recent years. Yuan *et al*. (2004) studied the phylogeny and biogeography of the Balsaminaceae inferred from ITS data; their work showed the monophyly of the Balsaminaceae, and also suggested the monophyly of *Impatiens*, revealing the origin of *Impatiens* in Southeast Asia. Janssens *et al*. (2006) studied the phylogeny of *Impatiens* and *Hydrocera* based on ITS and *atp*B-*rbc*L data. Their results indicated the relationship of these genera, and also found 14 clades within *Impatiens*. There was great progress in *Impatiens* phylogeny based on molecular data rather than morphological characters (Song *et al*., 2003, 2005; Song, 2006; Janssens *et al*., 2006, 2009), but few studies had combined molecular data with morphology to discuss the phylogeny of the Balsaminaceae. Some phylogenies thus lack the support of morphological characters, and progress with an infra-genera taxonomic system for *Impatiens* has thus met a huge barrier.

In order to clarify the relationships and patterns of morphological variation among different groups of *Impatiens*, much work had been done based on morphology, pollen, seed chromosomes, and molecules (Shimizu *et al*., 1985; Song *et al*., 2003, 2005; Song, 2006; Cong *et al*., 2007; Yu, 2008; Gao *et al*., 2011). This work has shown that *Impatiens* is a natural group with strong differentiation. Although there are several obvious phylogenic clades in *Impatiens*, the relationships between the clades are uncertain.

Leaf characters are constant and not influenced by whether the plant is dioecious or monoecious, and leaf epidermal characters have been shown to be important, not only in identifying the fossil remains of angiosperms but also in classification (Stace, 1984). These have been widely used in taxonomic treatments and systematic studies (Chen *et al*., 1991; Luo *et al*., 2001; Ren *et al*., 2003; Yang *et al*., 2005; Liu *et al*., 2011). Only a few species within Balsaminaceae have been examined with the scanning electron microscope (SEM), and the results have had limited taxonomic value (Cong *et al*., 2007). A few species have been examined by light microscope (LM), and as mentioned above, some works have confirmed taxonomical value. However, the value of leaf epidermal characters remains unclear. Our aim was to further study these, in order to search for new and valuable taxonomic characters and to clarify the specific circumscriptions within *Impatiens*.

1　Materials and Methods

We used mature leaves fixed in FAA (formalin-aceto-alcohol) from 106 field collections and representing 36 species with the Balsaminaceae (see Table 1 for details). The taxa were sampled by observing the leaf epidermis of different specimens, including collections from different regions.

Material for light microscopy was macerated in 35% sodium hypochlorite solution. Pieces of leaf epidermis were stained with safranin-alcohol (50%), and then dehydrated in an ethanol series before being mounted in Canada balsam. To check the constancy of epidermal structure, at least five slides were made from different parts of a single leaf for each taxon. Epidermal structures were examined and photographed underan O-

lympus DP70 microscope. The stomatal index

Table 1 Source of materials for observation of leaf epidermal micromorphology

Taxon	Province	Voucher	Upper/lower (epidermis)	Shape of cells	Anticlinal wall	Stomatal type	L/W (Stomata)	Stomatal index	Plate	Group
Hydrocera triflora	Hainan	Z. Huang 35156 (PE)	Upper	polygonal	Straight or arched	Anomocytic	1.20	18.60%	1: A1	I
			Lower	Irregular	Sinuolate	Anomocytic	1.32	28%	1: A2	I
Impatiens apalophylla	Guangxi	S. X. Yu 3748 (PE)	Upper	Irregular	Sinuous	Absent	Absent	Absent	2: I1	III
			Lower	Irregular	Sinuate	Anomocytic	1.13	29.85%	2: I2	III
I. aquatilis	Guangxi	S. X. Yu 3721 (PE)	Upper	polygonal	Straight	Anisocytic	Absent	Absent	1: C1	I
			Lower	Irregular	Sinuous	Anomocytic and anisocytic	1.35	23.23%	1: C2	I
I. arguta	Yunnan	S. X. Yu 4053 (PE)	Upper	Irregular	Sinuolate	Absent	Absent	Absent	3: B1	III
			Lower	Irregular	Sinuate	Most anisocytic and a few anomocytic	1.23	19.35%	3: B2	III
I. clavigera	Guangxi	S. X. Yu 3717 (PE)	Upper	polygonal	Substraight	Absent	Absent	Absent	1: E1	I
			Lower	Irregular	Sinuate	Anomocytic	1.34	21.15%	1: E2	I
I. cornutisepala	Guangxi	S. X. Yu 4023 (PE)	Upper	Irregular	Sinuolate	Anomocytic	1.25	7.45%	4: B1	V
			Lower	Irregular	Sinuate	Most anomocytic and a few anisocytic	1.35	17.94%	4: B2	V
I. dicentra	Guangxi	S. X. Yu 4032 (PE)	Upper	Irregular	Sinuate	Most anomocytic and a few anisocytic	1.45	2.74%	4: F1	V
			Lower	Irregular	Sinuate	Most anomocytic and a few anisocytic	1.27	36.09%	4: F2	V
I. paradoxa	Hubei	S. X. Yu 4093 (PE)	Upper	Irregular	Sinuate	Most anomocytic and a few anisocytic	1.15	3.33%	4: H1	V
			Lower	Irregular	Sinuate	Most anomocytic and a few anisocytic	1.31	27.69%	4: H2	V
I. bodinieri	Guangxi	S. X. Yu 3733 (PE)	Upper	Irregular	Sinuate	Absent	Absent	Absent	3: D1	IV
			Lower	Irregular	Sinuate	Most anomocytic, a few anisocytic and paracytic	1.27	18.92%	3: D2	IV
I. lobulifera	Guangxi	S. X. Yu 3220 (PE)	Upper	Irregular	Sinuolate	Absent	Absent	Absent	2: A1	II
			Lower	Irregular	Sinuate	Most anomocytic and a few anisocytic	1.22	24.49%	2: A2	II
I. hainanensis	Hainan	S. K. Lau 27038 (PE)	Upper	Polygonal	Substraight	Absent	Absent	Absent	1: B1	I
			Lower	Irregular	Sinuous	Most anomocytic and a few anisocytic	1.42	14.63%	1: B2	I
I. lateristachys	Sichuan	S. X. Yu 4084 (PE)	Upper	Irregular	Sinuate	Absent	Absent	Absent	3: I1	IV
			Lower	Irregular	Sinuate	Most anomocytic and a few anisocytic	1.17	17.46%	3: I2	IV
I. laxiflora	Sichuan	S. X. Yu 3987 (PE)	Upper	Irregular	Sinuous	Absent	Absent	Absent	3: F1	IV
			Lower	Irregular	Sinuate	Most anisocytic and a few anomocytic	1.21	14.29%	3: F2	IV
I. lecomtei	Sichuan	S. X. Yu 3873 (PE)	Upper	Irregular	Sinuolate	Most anomocytic and a few anisocytic	1.36	5.63%	4: E1	V
			Lower	Irregular	Sinuate	Anomocytic	1.33	21.98%	4: E2	V
I. leptocaulon	Guangxi	S. X. Yu 3681 (PE)	Upper	Irregular	Sinuate	Anomocytic	1.42	2.86%	4: C1	V
			Lower	Irregular	Sinuate	Most anomocytic and a few anisocytic	1.21	21.43%	4: C2	V
I. longialata	Sichuan	H. N. Qin *et al.* 17027 (PE)	Upper	Irregular	Sinuate	Most anomocytic and few anisocytic	1.33	2.90%	4: D1	V
			Lower	Irregular	Sinuate	Most anomocytic and a few anisocytic	1.35	17.07%	4: D2	V
I. macrovexilla	Guangxi	S. X. Yu 3350 (PE)	Upper	Irregular	Microsinuous to Sinuolate	Absent	Absent	Absent	1: H1	II
			Lower	Irregular	Sinuate	Most anomocytic and a few anisocytic	1.27	17.5%	1: H2	II
I. malipoensis	Guangxi	S. X. Yu 4037 (PE)	Upper	Irregular	Sinuous	Absent	Absent	Absent	2: C1	III
			Lower	Irregular	Sinuate	Most anomocytic and a few anisocytic	1.15	13.33%	2: C2	III
I. notolophora	Sichuan	H. N. Qin *et al.* 17140 (PE)	Upper	Irregular	Sinuate	Absent	Absent	Absent	4: A1	IV
			Lower	Irregular	Sinuate	Most anomocytic and a few anisocytic	1.16	12.12%	4: A2	IV
I. obesa	Guangxi	S. X. Yu 3775 (PE)	Upper	Irregular	Sinuolate	Absent	Absent	Absent	1: G1	II
			Lower	Irregular	Sinuate	Anomocytic and anisocytic	1.33	18.33%	1: G2	II
I. omeiana	Sichuan	S. X. Yu 4083 (PE)	Upper	Irregular	Sinuate	Absent	Absent	Absent	3: H1	IV
			Lower	Irregular	Sinuate	Most anomocytic and a few anisocytic	1.46	13.04%	3: H2	IV
I. oxyanthera	Sichuan	H. N. Qin *et al.* 17233(PE)	Upper	Irregular	Sinuolate	Absent	Absent	Absent	2: B1	II
			Lower	Irregular	Sinuolate	Anomocytic	1.31	14.75%	2: B2	II
I. parvisepala	Guangxi	S. X. Yu 3754 (PE)	Upper	polygonal	Straight	Absent	Absent	Absent	1: F1	I
			Lower	Irregular	Sinuate	Most anomocytic and rare anisocytic	1.37	17.78%	1: F2	I
I. platychlaena	Sichuan	S. X. Yu 4081 (PE)	Upper	Irregular	Sinuolate	Anomocytic and anisocytic	1.38	2.08%	4: I1	V
			Lower	Irregular	Sinuate	Most anisocytic and a few anomocytic	1.39	14.04%	4: I2	V
I. potaninii	Sichuan	H. N. Qin *et al.* 17189 (PE)	Upper	Irregular	Sinuous	Absent	Absent	Absent	3: C1	III
			Lower	Irregular	Sinuate	Most Anomocytic, a few anisocytic and rare diacytic	1.42	20.93%	3: C2	III
I. pritzelii	Hubei	S. X. Yu 4092 (PE)	Upper	Irregular	Sinuolate	Absent	Absent	Absent	2: G1	III
			Lower	Irregular	Sinuate	Anomocytic and anisocytic	1.45	17.24%	2: G2	III
I. pterocaulon	Guangxi	S. X. Yu 3719 (PE)	Upper	Irregular	Sinuate	Absent	Absent	Absent	4: G1	
			Lower	Irregular	Sinuate	Most anomocytic and a few anisocytic	1.13	21.57%	4: G2	
I. rostellata	Sichuan	H. N. Qin *et al.* 17192 (PE)	Upper	Irregular	Sinuolate	Absent	Absent	Absent	2: F1	III
			Lower	Irregular	Sinuate	Anomocytic	1.21	16.13%	2: F2	III
I. rubrostriata	Guangxi	S. X. Yu 3724 (PE)	Upper	Irregular	Substraight or microsinuous	Absent	Absent	Absent	1: D1	I
			Lower	Irregular	Sinuous	Anomocytic and anisocytic	1.22	16.67%	1: D2	I
I. procumbens	Guangxi	S. X. Yu 3709 (PE)	Upper	Irregular	Sinuolate	Absent	Absent	Absent	2: H1	III
			Lower	Irregular	Sinuate	Most anomocytic and non-typical anisocytic	1.22	23.26%	2: H2	III
I. spathulata	Guangxi	S. X. Yu 3763 (PE)	Upper	Irregular	Sinuolate	Absent	Absent	Absent	2: E1	III
			Lower	Irregular	Sinuate	Most anomocytic and a few anisocytic	1.67	31.82%	2: E2	III
I. tubulosa	Guangxi	S. X. Yu 3689 (PE)	Upper	Irregular	Sinuolate	Absent	Absent	Absent	3: A1	III
			Lower	Irregular	Sinuate	Most anisocytic and a few anomocytic	1.34	15%	3: A2	III
I. wenshanensis	Guangxi	S. X. Yu 4044A (PE)	Upper	Irregular	sinuolate	Absent	Absent	Absent	2: D1	III
			Lower	Irregular	Sinuolate to sinuous	Most anisocytic and a few anomocytic	1.42	10.11%	2: D2	III
I. wilsonii	Sichuan	S. X. Yu 4071 (PE)	Upper	Irregular	Deep sinuous	Most anomocytic and a few anisocytic	1.40	20.93%	4: G1	V
			Lower	Irregular	Deep sinuous	Most anomocytic and a few anisocytic	1.40	12.5%	4: G2	V

续表1

Taxon	Province	Voucher	Upper/lower (epidermis)	Shape of cells	Anticlinal wall	Stomatal type	L/W (Stomata)	Stomatal index	Plate	Group
I. wuchengyii	Yunnan	S. X. Yu 4061 (PE)	Upper	Irregular	Sinuolate	Absent	Absent	Absent	1: I1	II
			Lower	Irregular	Sinuate	Anomocytic	1.41	15.63%	1: I2	II
I. quadriloba	Yunnan	H. N. Qin *et al.* 17027 (PE)	Upper	Irregular	Sinuous	Absent	Absent	Absent	3: E1	IV
			Lower	Irregular	Sinuate	Anomocytic	1.51	15.63%	3: E2	IV

(*SI*) was calculated using the formula: $SI = S/(S+E) \times 100\%$, where S = number of stomata per unit area, and E = number of epidermal cells per same unit area (Dilcher 1974). Stomatal terminology is based on the classification proposed by Baranova (1972, 1987, 1992) and Dilcher (1974).

2 Results and Analysis

Leaf epidermal characters are listed in Table 1. Stomatal and other epidermal features are constant within species and are therefore good characters for taxonomy, especially in clarifying species circumscriptions.

2.1 Epidermal cells

The epidermal cells of the Balsaminaceae as seen under LM are usually polygonal or irregular in form, with anticlinal cell walls sinuous, undulate, and straight to curved. The patterns of anticlinal cell walls vary between species. Furthermore, in the same species the adaxially anticlinal cell walls and the abaxial ones are obviously different from each other. Straight to curved cell walls only occur in adaxial epidermis, namely *Hydrocera triflora* (Fig. 1: A), *Impatiens hainanensis* (Fig. 1: B), *I. aquatilis* (Fig. 1: C), *I. robrustriata* (Fig. 1: D), *I. clavigera* (Fig. 1: E), and *I. parvisepala* (Fig. 1: F). In contrast, abaxial walls are sinuolate to strongly sinuous, such as in *I. macrovexilla* (Fig. 1: H), *I. wenshanensis* (Fig. 2: D), and *I. omeiana* (Fig. 3: H).

2.2 Stomatal apparatus

Stomata are only present on abaxial surfaces in most species. There are also some species with stomata on both surfaces, such as *I. longialata* (Fig. 4: D), *I. paradoxa* (Fig. 4: H), and *I. dicentra* (Fig. 4: F). There are several types of stomata on the same blade; anomocytic, anisocytic, and paracytic. The anomocytic is the most common type in Balsaminaceae. The size of stomata and the length/width (L/W) ratio varies among species. The minimum ratio was 1.13, found in *I. apalophylla* (Fig. 2: I) and *I. pterocaulon* (Fig. 3: G), and the maximum was 1.67, in *I. spathulata* (Fig. 2: E). The stomatal index indicates the density of stomata in per-unit area of epidermis (Dilcher, 1974). The stomatal index value for abaxial epidermis is much larger than for adaxial epidermis if there are stomata on the adaxial side; there are only a few stomata on the adaxial surface if they are present at all. On the adaxial surface the minimum stomatal index value was 2.08%, found in *I. platychlaena* (Fig. 4: I), and the maximum was 18.60%, in *H. triflora* (Fig. 1: A). On the abaxial surface the minimum stomatal index value was 12.12%, found in *I. notolophora* (Fig.: 4: A); the maximum was 36.09%, in *I. dicenra* (Fig. 4: F).

2.3 Guard cells and epidermal hairs

Guard cells in Balsaminaceae usually have elliptical outlines in surface view. The leaves of the Balsaminaceae are usually entirely glabrous, although a small number of species have sparse hairs on the adaxial blade. Occasional epidermal hairs are found, for example, in *I. wuchengyii* (Fig. 1: I).

The following is a key to different groups of species based on leaf epidermal characters:

1. Adaxial epidermis without stomata
  2. Anticlinal cell walls straight or slightly curved ····· 1. Group I
  2. Anticlinal cell walls undulate, sinuolate, or sinuous
    3. Anticlinal cell walls undulate ····· 2. Group II
    3. Anticlinal cell walls sinuolate, or sinuous
      4. Anticlinal cell walls sinuolate ····· 3. Group III
      4. Anticlinal cell walls sinuous ····· 4. Group IV
1. Adaxial epidermis with stomata ····· 5. Group V

3 Conclusion and Disscussion

3.1 Relationships between epidermal features and gross morphology

The taxa studied show great variation in abaxial epidermal characters in terms of cell shape and stomatal index; it is difficult to find criteria for subdivision. We

found very limited correlation between epidermal characters and gross morphological characters. Although species may belong to the same group based on their epidermal characters, their gross morphology allows them to be readily distinguished. In Group I, for example, *Impatiens* species, namely *I. hainanensis* (Fig. 1: B), *I. aquatilis* (Fig. 1: C), *I. rubrostriata* (Fig. 1: D), *I. clavigera* (Fig. 1: E), and *I. parvisepala* (Fig. 1: F). However, there is also great variation of morphology among these *Impatiens* species, except that *I. clavigera* and *I. parvisepala* are close in form. *I. hainanensis* is different from *I. aquatilis* and *I. rubrostriata* in having four carpels and many ovules per carpel.

Except Group I, there was limited correlation between epidermal characters and gross morphology in Group II, Group III, and Group IV. We found a closer correlation between epidermal and gross characters in Group V. In Group V, stomata are present on adaxial surfaces, in spite of different cell shapes and stomatal indexes. In this group, the species studied are similar in morphology, except for *I. wilsonii*, *I. longialata*, and *I. leptocaulon*. The apices of upper and lower petals of lateral united petals have a long hair-like bristle. The species with this character are endemic to China (Hooker, 1908; Chen 2001), and include *I. paradoxa* (Fig. 4: H), *I. dicentra* (Fig. 4: F), and *I. lecomtei* (Fig. 4: E). Some other species in Group V, namely *I. wilsonii* (Fig. 4: G), *I. longialata* (Fig. 4: D), and *I. leptocaulon* (Fig. 4: C), are easily distinguished morphologically. The possession of four carpels and one ovule per carpel is characteristic of *I. wilsonii*. *I. longialata* is easily distinguished by its five carpels and undulate leaf margins. Five carpels and dentate leaf margins are key features of *I. leptocaulon*.

3.2 Correlation between epidermal characters and geographical distribution

Epidermal characters are more closely correlated with geographical distribution than is the gross morphology. Different groups have different geographical distribution patterns. In Group I, the species studied have either straight or curved anticlinal cell walls, and are confined to the south and southwest of China (Fig. 5: A); *Hydrocera triflora* and *I. hainanensis* are only found in *Hydrocera triflora* (Fig. 1: A) has free petals, berry-shaped fruit, and tricolporate pollen grains. These characters are different from *Impatiens* species, but the species has the same epidermal characters, straight or slightly curved anticlinal cell walls, as some Hainan, *I. clavigera* and *I. parvisepala* are found in southwestern Guangxi. Only *I. rubrostriata* and *I. aquatilis* cover a larger region, but both occur in southwestern China. Of the five groups, Group I have the most southern distribution. Group II includes only five *Impatiens* species in a south-north discontinuous distribution pattern.

In Group III, most species studied are from the south and central areas of China, and no species extends to Hainan, southern Guangxi, or southern Guangdong. This group has the largest distribution (Fig. 5: B), and is indicative of the most common epidermal characters in Balsaminaceae. In Group IV (Fig. 5: C), most species are found in the montane and subalpine zones of China, especially on western part of the second step of Chinese terrain, the eastern slopes of the Qing-Zang plateau. Some species extend to the borders of the second step and third step.

The species belonging to Group V are distributed in the central regions of China with a few species extending to the northwest (Fig. 5: D). Species with hair-like bristles on the apices of lateral united petals are mainly confined to the high altitudes of the central China and western Sichuan. The epidermal characters of these species are closely correlated with the gross morphological characters and geographical distribution. Although epidermal characters are correlated with geographical distribution very well in most species, there are also some geographical 'noises' in Group V. *I. leptocaulon* and *I. wilsonii*, for example, appear to have different morphological characters, and are readily distinguishable from *I. dicentra* and its allies. It seems that epidermal characters may be heavily modified by environmental factors. Some species with obviously different gross morphologies therefore share the same epidermal characters and geographical distribution patterns.

3.3 Relationship between *Hydrocera* and *Impatiens*

Hydrocera forms a sister clade to *Impatiens* within

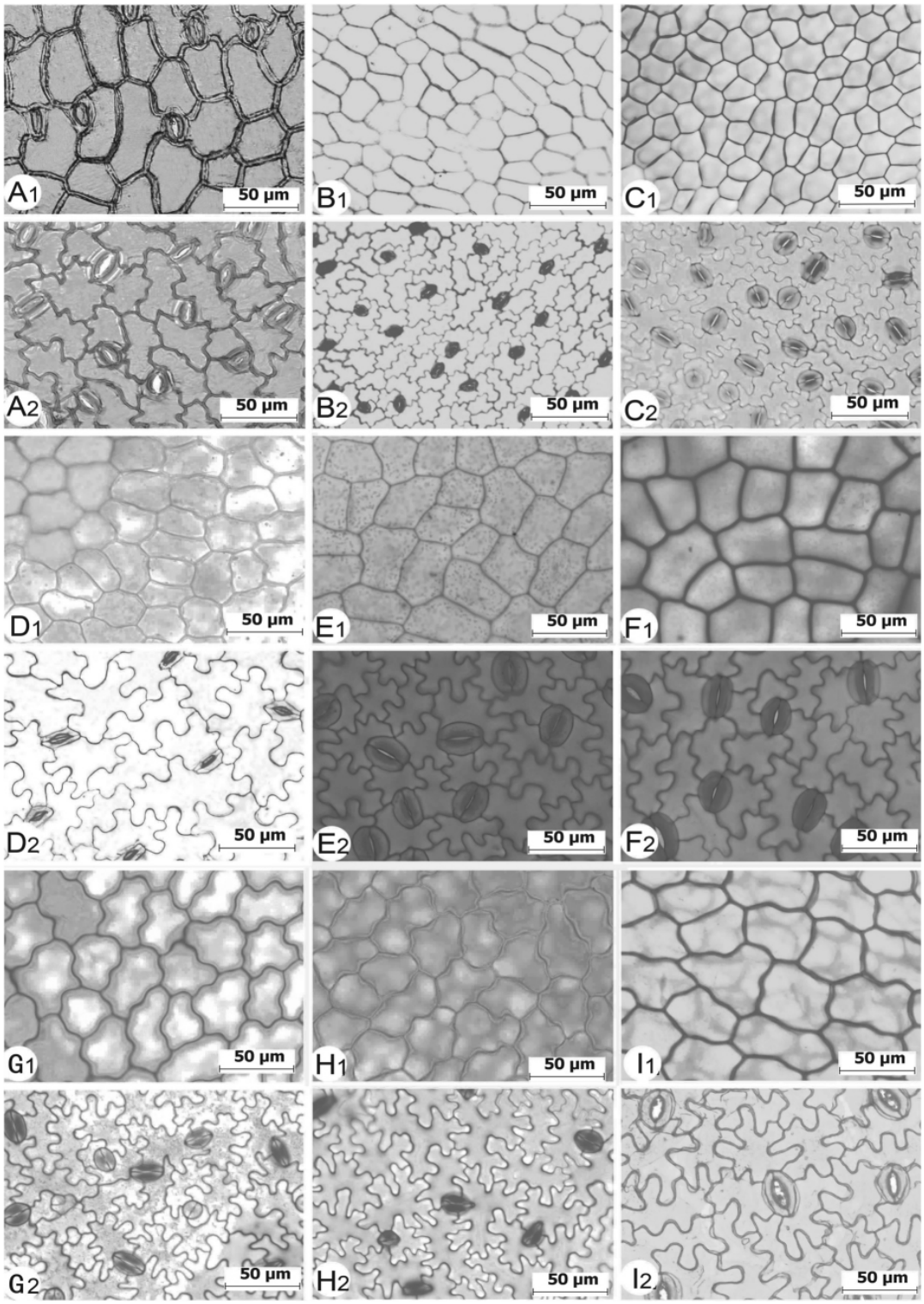

Fig. 1　Micromorphology of the leaf epidermis　**1**. Adaxial epidermis; **2**. Abaxial epidermis; **A.** *Hydrocera triflora*; **B.** *Impatiens hainanensis*; **C.** *I. aquatilis*; **D.** *I. rubrostriata*; **E.** *I. clavigera*; **F.** *I. parvisepala*; **G.** *I. obesa*; **H.** *I. macrovexilla*; **I.** *I. wuchengyii*.

Balsaminaceae based on molecular data (Janssens *et al*., 2006), and *I. clavigera* and its allies are basal taxa within *Impatiens*. This hypothesis is confirmed by the epidermal characters. In Group I, *H. triflora*, *I. clavigera*, and its ally *I. parvisepala*, including the relatively basal taxon *I. hainanensis*, are all species with straight or curved anticlinal cell walls, except that there are stomata on the adaxial surface in *H. triflora*. Although

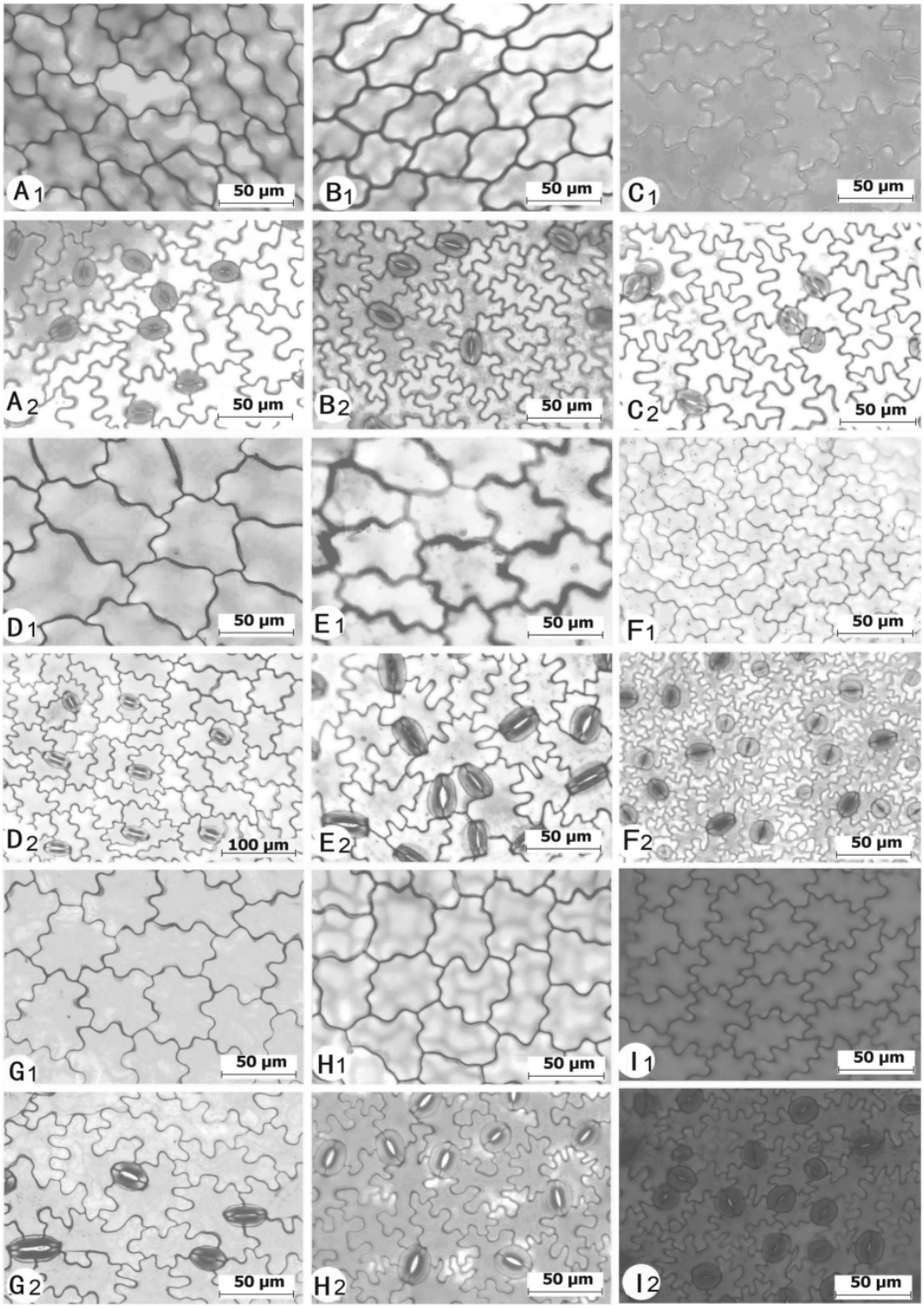

Fig. 2 Micromorphology of the leaf epidermis **1**. Adaxial epidermis; **2**. Abaxial epidermis; **A.** *I. lobulifera*; **B.** *I. oxyanthera*; **C.** *I. malipoensis*; **D.** *I. wenshanensis*; **E.** *I. spathulata*; **F.** *I. rostellata*; **G.** *I. pritzelii*; **H.** *I. procumbens*; **I.** *I. apalophylla*.

there are also some stomata on the adaxial surface in Group V species, the majority of these species are embedded in the upper clade in the phylogenic tree of the family (Yu, 2008). It seems therefore, that in comparison with whether stomata are present on adaxial surfaces or not, the characters of the anticlinal cell walls are very taxonomically valuable.

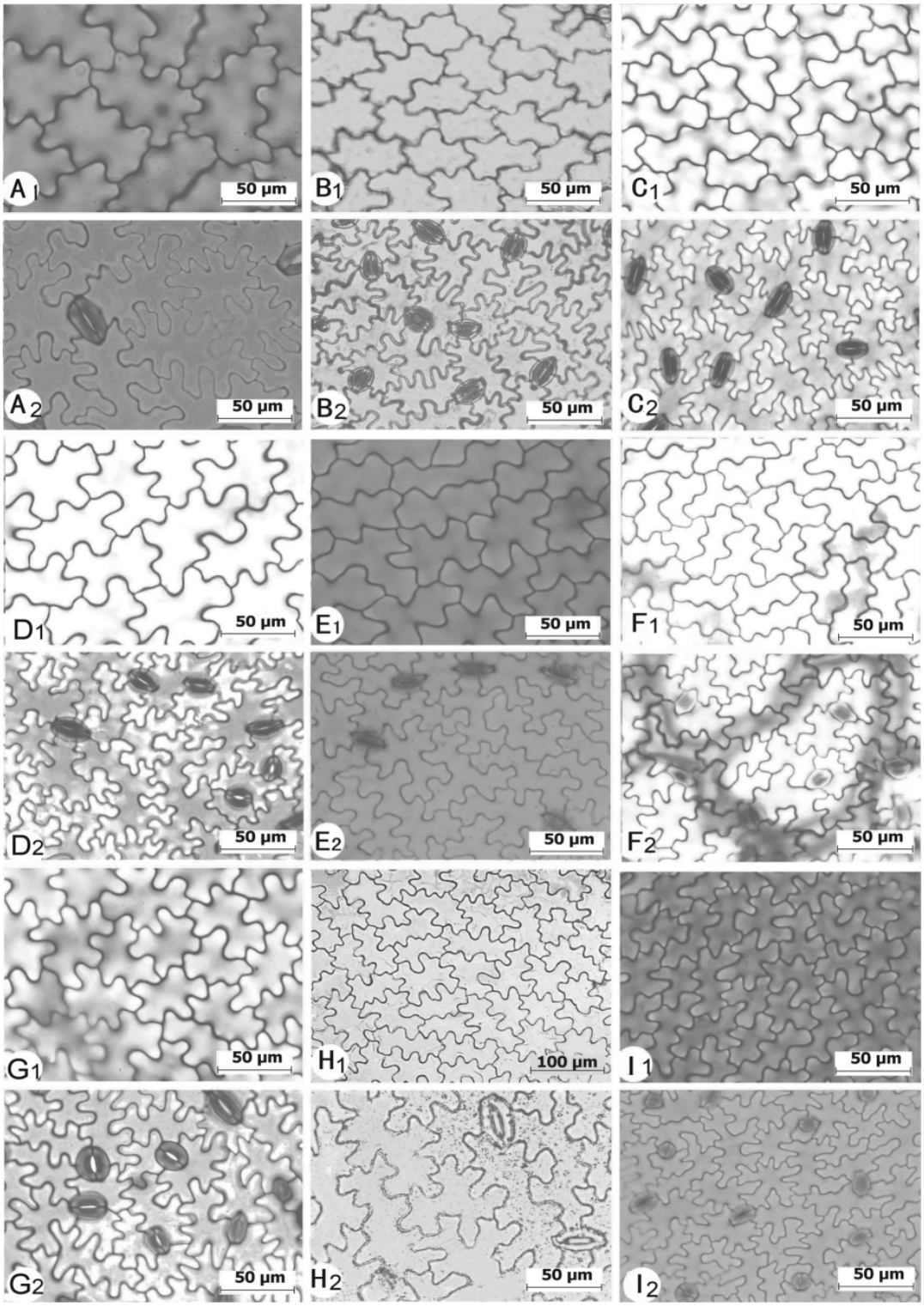

Fig. 3　Micromorphology of the leaf epidermis　**1**. Adaxial epidermis; **2**. Abaxial epidermis; **A.** *I. tubulosa*; **B.** *I. arguta*; **C.** *I. potaninii*; **D.** *I. bodinieri*; **E.** *I. quadriloba*; **F.** *I. laxiflora*; **G.** *I. pterocaulon*; **H.** *I. omeiana*; **I.** *I. lateristachys*.

3.4 Evidence for species circumscriptions

Variation in epidermal characters on adaxial and abaxial surfaces is easy to recognise. There is obvious variation on abaxial surfaces in cell shape and stomatal index, but it is difficult to summarize these variation patterns. The abaxial epidermal characters are much more valuable for clarification of species circumscriptions than for subdivision of the genus *Impatiens*. The epidermal

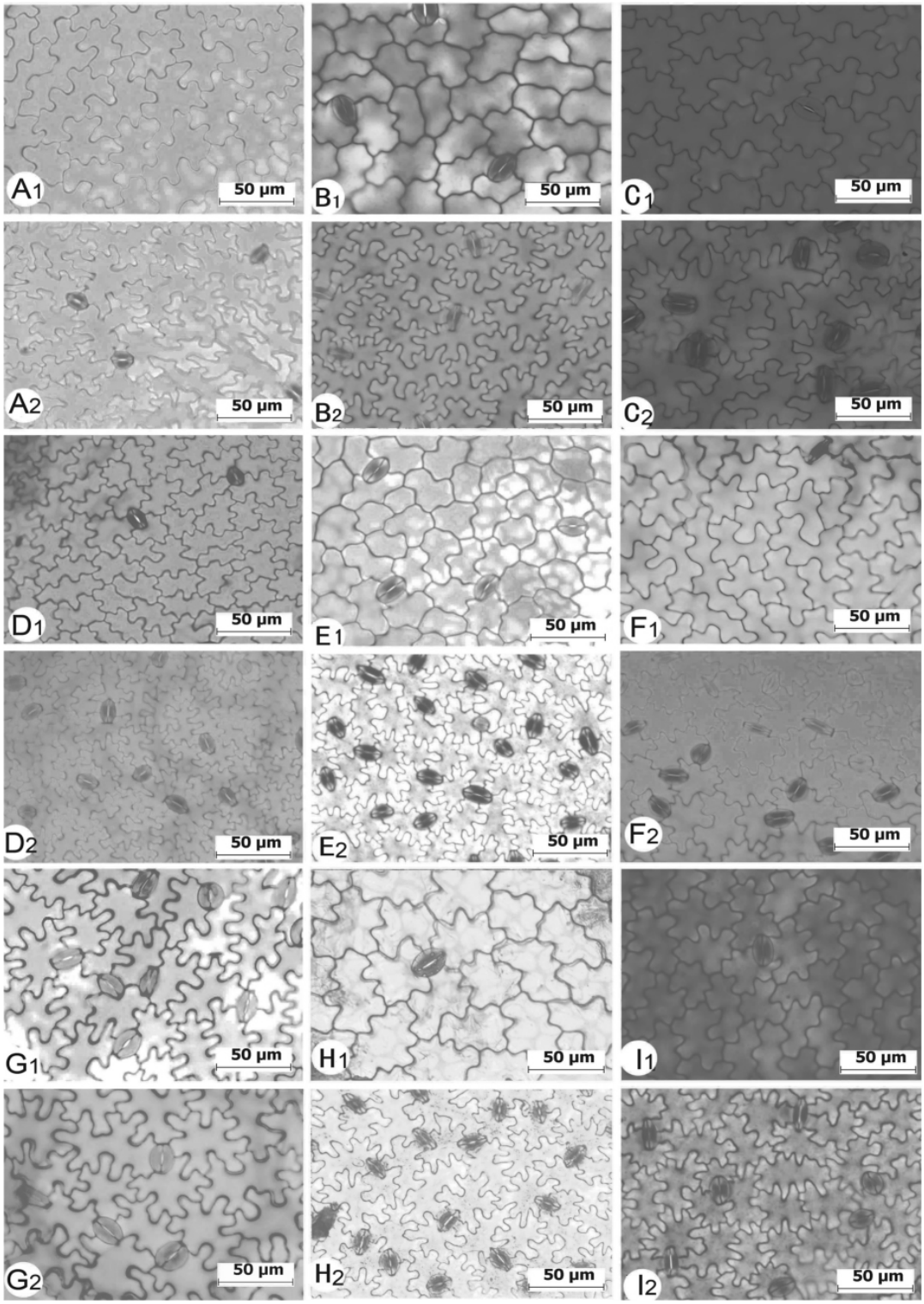

Fig. 4 Micromorphology of the leaf epidermis **1**. Adaxial epidermis; **2**. Abaxial epidermis; **A.** *I. notolophora*; **B.** *I. cornutisepala*; **C.** *I. leptocaulon*; **D.** *I. longialata*; **E.** *I. lecomtei*; **F.** *I. dicentra*; **G.** *I. wilsonii*; **H.** *I. paradoxa*; **I.** *I. platychlaena*.

characters on adaxial surfaces provide a much more valuable message in species demarcation than those on the abaxial surface. In addition, the adaxial epidermal characters are correlated with geographical patterns and gross morphologies.

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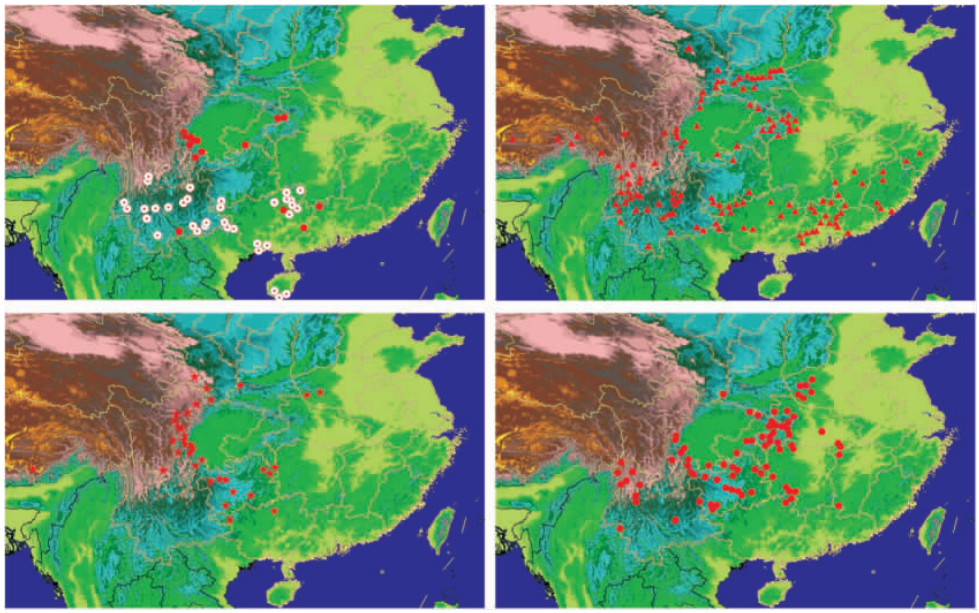

Fig. 5　Geographical distribution patterns with the Balsaminaceae　**A.** Group Ⅰ(⊙) and Group Ⅱ(⬟); **B.** Group Ⅲ(▲); **C.** Group Ⅳ(★); **D.** Group Ⅴ(●).

its preparation.

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