

Cytological study on the genus *Syncalathium* (Asteraceae-Lactuceae), an endemic taxon to alpine scree of the Sino-Himalayas

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Abstract Cytological characters of four species in *Syncalathium* (Asteraceae: Lactuceae), a small genus with six identified species endemic to alpine scree of the Sino-Himalayan region, are surveyed in this report. Three species (*Syncalathium pilosum*, *Syncalathium chrysocephalum*, and *Syncalathium disciforme*) are examined for the first time. Combined with our previous counts, five species have been cytologically investigated from the genus and the results indicated that all species are diploid with the basic somatic chromosome number of $x=8$. The karyotype asymmetry of *Syncalathium souliei* is 2A, distinct from the other four species of 1A, and the remaining species are divided into two subgroups with different karyotypes, consistent with their morphological features. The significance of the cytological evolution of *Syncalathium* is briefly discussed.

Key words Asteraceae, chromosome numbers, diploidy, karyotype, *Syncalathium*.

Syncalathium Lipschitz is a small genus with six identified species occurring primarily in the Qinghai-Tibet Plateau of China (Ling, 1965; Bremer, 1994; Shih, 1997; Lack, 2007; Zhang, 2009). It is restricted mainly to the alpine scree or sand areas at altitudes mostly ranging from 3500 to 5400 m in the Himalayan–Hengduan region with relatively restricted and narrow distribution for each species (Wang & Zhang, 1994; Shih, 1997).

The genus was established by Lipschitz (1956), including only *Syncalathium sukaczewii* Lipsch. Ling (1965) expanded the genus with inclusion of the new species of *Syncalathium roseum* Y. Ling and the former *Lactuca* L. sect. *Aggregatae* Franch. (Franchet, 1895; Stebbins, 1940; Kitamura, 1953). The placement of *Syncalathium* within the tribe Lactuceae Cass. has been controversial. It was placed in the subtribe Synclathinae Lipsch. by Lipschitz (1956), and in the subtribe Lactucinae Dumort. by Ling (1965), Bremer (1994) and Lack (2007). Moreover, the genus was put in Synclathia Y.R. Ling of subtribe Lactucinae (Ling, 1997) together with *Soroseris* Stebbins.

Syncalathium resembles *Soroseris* morphologically with similar habitats, but differs mainly in characteristics of its achenes, involucre and setae. *Soroseris* and *Syncalathium* are both rosette herbs, characterized by greatly expanded, apparently hollow stems and closely-crowded heads. They probably represent a convergence resulting from adaptation to extreme habi-

tats (Stebbins, 1940). *Syncalathium chrysocephalum* (C. Shih) S.W. Liu and *Syncalathium qinghaiense* (C. Shih) C. Shih were placed in *Soroseris* as two new species by Shih (1993) for their similar morphology, but were transferred to *Syncalathium* by some taxonomists (Liu, 1996; Shih, 1997).

It is difficult to collect *Syncalathium* because its geographical distribution is mainly in the high elevation of the Himalayan–Hengduan Mountains. Zhang et al. (2007) reported two cytological species in *Syncalathium*, including two populations of *Syncalathium souliei* (Franch.) Y. Ling and one of *Syncalathium kawaguchii* (Kitam.) Y. Ling. The results showed that the two species were diploid with the same chromosome number of $2n=16$, but different karyotype formulae and karyotype asymmetry (KA). Chromosome counts, however, are scanty in contrast with the large amount of cytological data on the Lactuceae as a whole (Stebbins et al., 1953; Turner et al., 1961; Tomb, 1977; Tomb et al., 1978; Jansen & Stuessy, 1980). It is necessary to add new data for the genus for a full understanding of its chromosome evolution.

The value of cytological data in the study of plant evolution and diversification has long been established, especially for taxa occurring in extreme habitats such as alpine mountains (Stebbins, 1950, 1971; Hong, 1990; Stace, 2000). Polyploidy is common in plants of cold climates with harsh and stressful environments (Löve & Löve, 1949, 1967; Grant, 1981). In a recent study, however, Nie et al. (2005) reported a relatively low frequency of polyploids in plants from the Hengduan Mountains of southwest China. As part of our effort to accumulate cytological data from this area, we

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Table 1 Localities, geographical positions, altitudes and voucher numbers of four investigated species in *Syncalathium*

Taxon	Locality	Position	Altitude (m)	Voucher (KUN)
<i>Syncalathium kawaguchii</i> (Kitam.) Y. Ling	Zhigela, Riwoqê, Tibet, China	31° 06' N, 96° 30' E	4500	J.W. Zhang & T.Y. Tu 93
<i>Syncalathium pilosum</i> (Y. Ling) C. Shih	Rendui, Namling, Tibet, China	30° 04' N, 89° 06' E	4200	J.W. Zhang & T.Y. Tu 220
<i>Syncalathium chrysocephalum</i> (C. Shih) S.W. Liu	Machala, Riwoqê, Tibet, China	31° 24' N, 96° 40' E	4780	J.W. Zhang & T.Y. Tu 112
<i>Syncalathium disciforme</i> (Mattf.) Y. Ling	Dalijiashan, Xunhua, Qinghai, China	35° 34' N, 102° 45' E	3590	J.W. Zhang & W.D. Zhu 07006

present karyomorphological data for *Syncalathium* and investigate their possible systematic and evolutionary implications.

1 Material and methods

Seeds were collected for four species of *Syncalathium* (Table 1). Voucher specimens are deposited in the herbarium of Kunming Institute of Botany (KUN). All cytological observations were made from root tips. Seeds were stored at 4 °C, soaked overnight in distilled water at room temperature, and germinated on wet filter papers in Petri dishes. Fresh root tips approximately 1 cm long were cut, pretreated in 0.003 mol/L 8-hydroxyquinoline solution at 22–23 °C in the dark for 6–8 h, then fixed with glacial acetic acid : absolute alcohol (1:3) at approximately 4 °C for 2–5 h. They were then placed in 70% ethanol for 10 min and rinsed in distilled water twice for approximately 20 min. Prior to staining, the root tips were hydrolysed in 1 mol/L HCl : 45% acetic acid (1:1) at 60 °C for 30 s, stained with 1% aceto-orcin overnight, and squashed for cytological observation. Permanent slides were made using the standard liquid nitrogen method.

Observations were made of somatic mitotic interphase, prophase and metaphase. Karyomorphological classification of the mitotic interphase nuclei and prophase chromosomes follows Tanaka (1971, 1977, 1987); the designation of the centromeric position as median (m) or submedian (sm) follows Levan et al. (1964); the karyotype asymmetry (KA) is classified according to Stebbins (1971).

2 Results

2.1 Interphase nuclei and prophase chromosomes

The interphase nuclei of *Syncalathium* has an almost homogeneous distribution throughout the nucleus, forming a fibrous network of the chromonemata (Fig. 1: A). According to the system of Tanaka (1971, 1977), they can be categorized as the diffuse type. The chromosomes at mitotic prophase stain darkly and homo-

geneously, showing several small elastic constrictions (Fig. 1: B). There are no conspicuously heterochromatic or euchromatic segments in the four species of the genus. According to the system of Tanaka (1971, 1977, 1987), the prophase chromosomes of the genus are classified as the interstitial type.

2.2 Chromosome counts and karyomorphology

Chromosome number of $2n=16$ is found in all the species. Their KA was categorized as type 1A according to Stebbins (1971). Metaphase chromosomes of the four species of *Syncalathium* are shown in Fig. 1. Their detailed parameters and karyotype formulae are listed in Table 2. Brief descriptions of the cytological features of each species are as follows.

Syncalathium kawaguchii (Kitam.) Y. Ling:

The population was collected from Zhigela (Riwoqê, Tibet, China) with a formula of $2n=16=14m+2sm$, which is the same as that of the *Damala* population (Qamdu, Tibet, China) (Zhang et al., 2007), but no satellite was found in this population. The ratio of the longest to the shortest chromosome is 1.57 and the KA is of type 1A (Fig. 1: C, C').

Syncalathium pilosum (Y. Ling) C. Shih:

The karyotype formula of the Rendui (Namling, Tibet, China) population is $2n=16=14m(2SAT)+2sm$, with pair 3 being sub-metacentric. Two satellites were observed in this population, attached to the short arms of the eighth pair. The ratio of the longest to the shortest chromosome is 1.51 and the KA is of type 1A (Fig. 1: D, D').

Syncalathium chrysocephalum (C. Shih) S.W. Liu:

The sample from Machala (Riwoqê, Tibet, China) has a formula of $2n=16=16m(4SAT)$. Four satellites were observed in this population, attached to the short arms of the sixth and eighth pairs. The ratio of the longest to the shortest chromosome is 1.42, and the KA is of type 1A (Fig. 1: E, E').

Syncalathium disciforme (Mattf.) Y. Ling:

The population from Dalijiashan (Xunhua, Qinghai, China) has a formula of $2n=16=16m(2SAT)$. Two satellites were observed in this population, attached to the short arms of the seventh pairs. The ratio of the

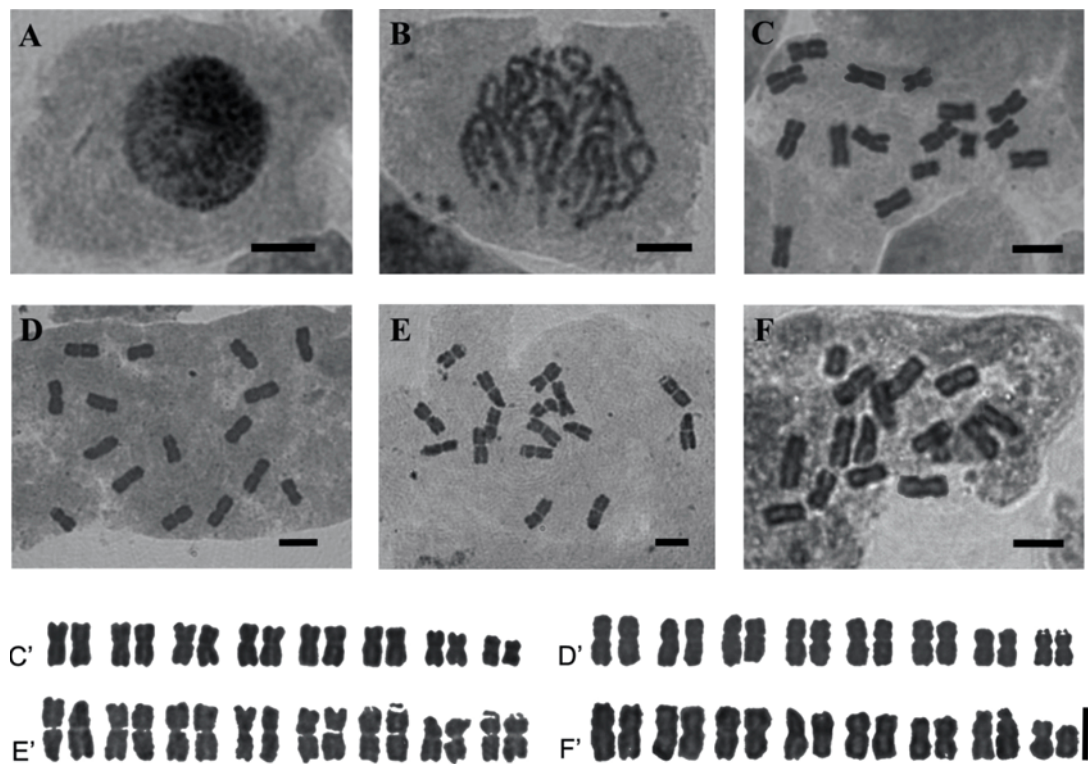


Fig. 1. Mitotic nuclei, metaphase chromosomes and karyotypes of four species in *Syncalathium*. **A**, Mitotic interphase nuclei. **B**, Mitotic prophase. **C–F**, Mitotic metaphase. **C'–F'**, Karyotypes. **C, C'**, *Syncalathium kawaguchii* (Zhigela, Riwoqê), $2n=16$. **D, D'**, *Syncalathium pilosum* (Rendui, Namling), $2n=16$. **E, E'**, *Syncalathium chrysocephalum* (Machala, Riwoqê), $2n=16$. **F, F'**, *Syncalathium disciforme* (Dalijashan, Xunhua), $2n=16$. Scale bars = 5 μ m.

longest to the shortest chromosome is 1.51, and the KA is of type 1A (Fig. 1: F, F').

3 Discussion

All species of *Syncalathium* examined had the diffuse type for the interphase nuclei and the interstitial type for a prophase chromosome condensation pattern, the same as our recent report (Zhang et al., 2007). The same chromosome number of $2n=16$ and the basic number of $x=8$ are found in all species, consistent with the results of the previous study (Zhang et al., 2007). As shown in Table 2, all the species showed a KA of type 1A

in this report, but *S. souliei* has a distinct 2A type with different karyotype, as observed in our previous study (Zhang et al., 2007).

Morphological data also support the separation of *S. souliei* from the genus. The achene of *S. souliei* differs largely from the remaining species in having four prominent ridges and short beak (vs. one and two nerves on each face and lack of beak in all other species in the genus). The achene characters have been considered as important traits in classification in the Compositae (Singh et al., 1972; Mukherjee & Sarkar, 1995; Blanca & Guardia, 1997; Kilian, 1997; Dasgupta & Mukherjee, 2007; Das & Mukherjee, 2008). *S. souliei* might be close to *Lactuca* s.l., because the species was

Table 2 Somatic chromosome number ($2n$), basic chromosome number and ploidy level, karyotype formula ($2n$), karyotype asymmetry (KA), locality and data source of *Syncalathium* in this and previous reports

Taxon	$2n$ /basic no./ploidy level	Karyotype formula ($2n$)	KA	Locality	Data source
<i>Syncalathium souliei</i>	16/8/2x	6m+10sm	2A	Dêqên, Yunnan, China	Zhang et al. (2007)
	16/8/2x	6m+10sm	2A	Zhongdian, Yunnan, China	Zhang et al. (2007)
<i>Syncalathium kawaguchii</i>	16/8/2x	14m(2SAT)+2sm	1A	Qamdu, Tibet, China	Zhang et al. (2007)
	16/8/2x	14m+2sm	1A	Riwoqê, Tibet, China	This study
<i>Syncalathium pilosum</i>	16/8/2x	14m(2SAT)+2sm	1A	Namling, Tibet, China	This study
<i>Syncalathium chrysocephalum</i>	16/8/2x	16m(4SAT)	1A	Riwoqê, Tibet, China	This study
<i>Syncalathium disciforme</i>	16/8/2x	16m(2SAT)	1A	Xunhua, Qinghai, China	This study

m, median region; SAT, satellite; sm, submedian region.

traditionally treated as a member of the section *Aggregatae* in *Lactuca* (Stebbins, 1940). However, *S. souliei* is also different from the *Lactuca* s.l. group, in that the achene of the former has only one ridge on each face and a very short beak, but the latter has more ridges and a longer beak. *Syncalathium souliei* might be recognized as a distinct taxon at the generic level. Molecular studies on *Soroseris* and *Syncalathium* also suggested that *S. souliei* should be excluded from *Syncalathium*, in that *Syncalathium* (excluding *S. souliei*) has a close relationship with *Soroseris*, and the two taxa are in the subtribe Crepidinae, whereas *S. souliei* is still in the subtribe Lactucinae (Zhang, 2009).

The remaining four species show the same asymmetry type of 1A and similar karyotypes (Table 2), indicating a close relationship among them. Cytological data also showed two kinds of karyotypes among the four species. The chromosomes of *S. disciforme* and *S. chrysocephalum* are both of the median type (16m), whereas those of *S. kawaguchii* and *S. pilosum* are of the submedian type (14m and 2sm). These are highly consistent with their morphological features, such as the former two having yellow florets and the latter two having purple florets. Levitzky (1931) and Stebbins (1971) pointed out that plants with more symmetrical karyotypes were primitive and those with more asymmetrical karyotypes were advanced. Therefore, cytological data suggested that the two species of *Syncalathium* with purple flowers were derived from the yellow flower species, also consistent with the molecular results (Zhang, 2009).

Because habitats of *Syncalathium* are restricted to the very harsh alpine scree, we expected to find high levels of polyploidy among them. However, all are diploid with a basic number of $x=8$. This pattern is largely congruent with the results in *Solms-laubachia* Muschler (Brassicaceae) (Yue et al., 2004), *Tibetia* (Ali) H.P. Tsui (Fabaceae) (Nie et al., 2002), *Delphinium* L. (Ranunculaceae) (Yuan & Yang, 2008), and so on. The results support the viewpoint proposed by Nie et al. (2005) that polyploidy, especially infrageneric polyploidy, has not played an important role in the evolution of the plants in the Hengduan Mountains region. However, further observations in the various taxa are needed to reveal the role of polyploidy in this biodiversity hotspot.

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