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CYTOTAXONOMIC NOTES ON THE TRIBE HELIEAE (GENTIANACEAE)

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Abstract. A survey of all known chromosome counts of gentian tribe Helieae are presented including new reports for ten species. Amongst the novelties are four genera of the Gentianaceae that are reported for the first time, *Calolisianthus* and *Helia* (both Helieae), and *Schultesia* and *Zygostigma* (both Chironieae). In the Helieae, our results reinforce the hypothesis of Weaver 1969 that two ploidy series occur in the tribe, one polyploid based on n = 20 and one dysploid based on n = 21. The basal chromosome number for the Helieae remains to be determined.

Keywords: Chromosome number, cytology, Gentianaceae, Helieae, Neotropics, Taxonomy

Cytotaxonomy can be a useful complement to morphological or molecular investigations (Dobigny, Ducroz et al. 2004; Guerra 2008). Counting chromosomes has the advantage of being relatively fast and cheap, and yet, it can only be done by removing and fixing developing tissue from living plants. Within the Gentianaceae, many chromosome numbers have been recorded, mostly from tribe Gentianeae (Struwe and Albert 2002; Yuan et al. 1998). Tribe Helieae has many diverse species all occurring in the Neotropics, yet few chromosome observations exist. Weaver and Rudenberg published the only existing Helieae numbers of a few species (Weaver 1969, 1972; Weaver and Rudenberg 1972). The notes presented here are new chromosome counts based on recently collected material in South America.

Materials and methods

Buds were fixed in the field in a variant of Carnoy's solution composed of ¼ pure acetic acid, ¾ ethanol (5% methanol denaturized), a few drops of acetic carmine, and one drop of iron acetate. The fixations were stored in this fixating solution for at least one month, after which the buds were transferred to acetic carmine with one drop of iron acetate, heated gently on a small Bunsen burner flame for two minutes to complete the coloration, and then cooled with 45% acetic acid. The standard squashing technique was used on a portion of stamen or ovules, and preparations were observed with a Leitz Dialux 20 EB microscope. Preparations of interest were permanently fixed using melted resin-lanolin mixture. At least three counts were made for each species.

Results

In addition to the chromosome numbers found in tribe Helieae by Weaver (Weaver 1969, 1975; Weaver and Rudenberg 1972), we obtained new counts for four species of *Macrocarpaea* (*M. glamorosa*, *M. glaziovii*, *M. obtusifolia*, and *M. rubra*,) and four other Helieae (*Calolisianthus pedunculatus*, *Chelonanthus purpurascens*, *Helia brevifolia*, and *Symbolanthus mathewsii*; Fig. 1A–B; Table 1). Additionally we report numbers for two genera of tribe Chironieae: *Schultesia* and *Zygostigma* (Fig. 1C; Table 1). Chromosomes of Helieae are usually short and round, often aggregated, rendering counting difficult.

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TABLE 1: Synthesis of the chromosome numbers in gentian tribe Helieae, and two new in tribe Chironieae.

SPECIES	N	2N	LOCALITY	COLLECTOR (NO.)	REFERENCE
Tribe Helieae					
Genus *Calolisianthus*					
C. pedunculatus Gilg	n = 20		Brazil. Paraná, Mun. Palmeira, Palmeira	Grant, Zeltner & Trunz 4595	This study
Genus *Chelonanthus*					
C. alatus (Aubl.) Pulle	n = 20		Panama. Coclé: 3.5 km S of El Valle de Antón	Weaver 1672	Weaver 1969
C. bifidus (HBK.) Gilg	n = 20		Colombia. Meta: ça. 5 km. N of Villavicencio	Weaver 2639	Weaver and Rudenberg 1975
C. purpurascens (Aubl.) Struwe, S. Nilsson, V.A. Albert		2n = 40	Brazil. Bahia, Chapada Diamantina, Morro do Pai Inacio	Grant & Trunz 4643	This study
C. uliginosus (Griseb.) Gilg	n = 20		Colombia. Cundinamarca: between Bogotá & Villavicencio	Weaver 2640	Weaver and Rudenberg 1975
Genus *Helia*					
Helia brevifolia Cham.		2n = 40	Brazil. Paraná, Mun. Palmeira, Recanto do Papagaio	Grant, Zeltner and Trunz 4594	This study
Genus *Lagenanthus*					
Lagenanthus princeps (Lindl.) Gilg	n = ca. 40		Venezuela. Tachira: base of Paramo de Tamá	Weaver 2612	Weaver and Rudenberg 1975
Genus *Macrocarpaea*					
M. densiflora (Benth.) Ewan	n = 21		Colombia. Tolima: between Fresno & Manizales	Weaver 2644	Weaver and Rudenberg 1975
M. glabra (L.f.) Gilg	n = 21		Colombia. Cundinamarca: above Bogotá on road to Villavicencio	Weaver 2636	Weaver and Rudenberg 1975
M. glamorosa J.R. Grant		2n = 42	Ecuador. Zamora-Chinchipe: between Amaluza and Zumba	Grant 4520	This study
M. glaziovii Gilg		2n = 42	Brazil, Rio de Janeiro, estrada do Sumaré	Grant, Zeltner, Resende Silva & Trunz 4604	This study

TABLE 1 CONT.

SPECIES	N	2N	LOCALITY	COLLECTOR (NO.)	REFERENCE
M. harlingii J.S. Pringle		2n = 42	Ecuador. Zamora-Chinchipe: between Amaluza and Zumba	Grant 4518	This study
M. obtusifolia (Griseb.) Gilg		2n = 42	Brazil, Rio de Janeiro, estrada do Sumaré	Grant, Zeltner, Resende Silva & Trunz 4605	This study
		2n = 42	Brazil, Sao Paulo, Mun. Ubatuba, Serra do Mar,	Grant, Zeltner, Calió & Trunz 4597	This study
M. rubra Malme		2n = 42	Brazil, Paraná, Mun. Quatro Barras, Morro Sete	Grant, Zeltner & Trunz 4593	This study
M. thamnoides (Griseb.) Gilg	n = 21		Jamaica. St. Andrew: Fairy Glade on Mt. Horeb	Weaver 952	Weaver 1969
M. valerii Standl.	n = 21		Costa Rica. Heredia: 1.5 miles N of Vara Blanca	Weaver 1405	Weaver 1972
Genus *Symbolanthus*					
S. frigidus (Sw.) Struwe & K. Gould	n = 40		St. Vincent : Soufrière	Howard, Cooley & Weaver, 17712	Weaver 1969
S. mathewsii (Griseb.) Ewan	n = 40		Ecuador. Zamora-Chinchipe, Yangana	Grant 4502	This study
S. pulcherrimus Gilg	n = 40		Costa Rica. Cartago: 13 mi. SW of El Empalme	Weaver 1406	Weaver 1969
S. tricolor Gilg	n = 40		Colombia. Cundimarca: near San Miguel	Weaver 2635	Weaver and Rudenberg 1975
Tribe Chironieae					
Genus *Schultesia*					
Schultesia bahiensis E.F. Guim & Fontella.		2n = 28	Brazil, Bahia, Chapada Diamantina, Morro do Pai Inacio	Grant & Trunz 4642	This study
		2n = 28	Brazil, Bahia, Rio de Contas, Pico das Almas	Grant & Trunz 4646	This study
Genus *Zygostigma*					
Zygostigma australe		2n = ca. 72	Brazil. Paraná: Palmeira	Grant, Zeltner & Trunz 4596	This study

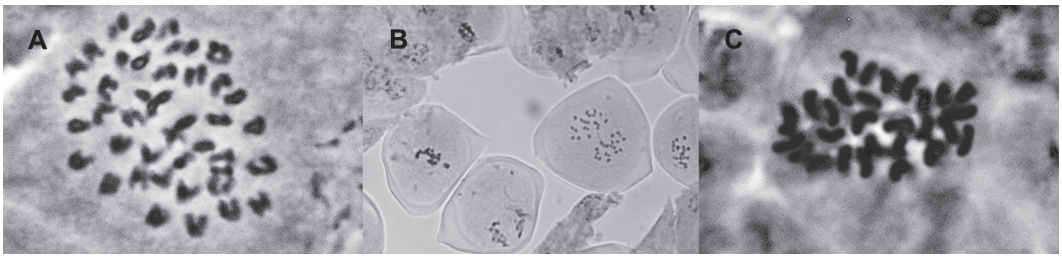

FIGURE 1. Chromosomes of three species of Gentianaceae. **A**, *Chelonanthus purpurascens* (Aubl.) Struwe, S. Nilsson & V. A. Albert (from *Grant & Trunz 4643*); **B**, *Helia brevifolia* Cham. (from *Grant 4594*); **C**, *Schultesia bahiensis* E. F. Guim. & Fontella (from *Grant & Trunz 4642*).

DISCUSSION

The new chromosome numbers reported in this study slightly reinforce the hypothesis made by Weaver (Weaver, 1969) of two ploidy series (one polyploid and one dysploid), but several numbers are still missing and important to confidently resolve the ploidy patterns and basal number (x) among Helieae. The current hypothesis we join is that, n = 20, obtained in every *Chelonanthus* species and *Helia brevifolia* may be a polyploid series base of which *Lagenanthus* (n = 40) and *Symbolanthus* (n = 40) belong. *Chelonanthus* and *Helia* may be the base for a dysploid series too, and *Macrocarpaea* (n = 21) would belong to that one.

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