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Aspidistra daqingshanensis (Asparagaceae), a new species from Guangxi, China

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ABSTRACT: Aspidistra daqingshanensis (Asparagaceae) is described and illustrated as a new species from Longzhou County, Guangxi, China. It is morphologically similar to Aspidistra fasciaria G.Z. Li but differs in its leaf blade wider to 4–8 cm, distal margin denticulate, perianth lobes broadly ovate, yellow, stigma white, slightly recurved at apex. A somatic chromosome count of 2n = 38 and a karyotype formula 2n = 22m + 10sm + 6st were determined for A. daqingshanensis. The new species is currently only known from the type locality in southwest Guangxi, near the border with northern Vietnam.

KEY WORDS: Aspidistra fasciari, China-Vietnam border, chromosome number, karyotype, plant diversity, taxonomy.

INTRODUCTION

Aspidistra Ker-Gawl. (Ker Gawler, 1822) (Asparagaceae), currently about 212 species known worldwide, is an Asia endemic genus, mainly distributed in the tropical and subtropical regions of eastern Asia, from eastern India in the west to Kagoshima in Japan in the east, and north to Hubei in China and south to Malaysia (Li, 2004; Tillich, 2023). The China-Vietnam border area is the distribution and differentiation centre of Aspidistra, a large number of new species have been discovered and reported in this region for the past few years (e.g. Xu et al., 2017; Cai et al., 2018; Vislobokov et al., 2019; Pan et al., 2020; Nguyen et al., 2020; Averyanov et al., 2020, 2021; Lin et al., 2023).

In China with more than 110 species which are mainly distributed in southern and southwestern regions, China becomes the richest country in the *Aspidistra* genus. In the spring of 2021, during a field survey of Guangxi economic plants, the first three authors discovered a fruiting *Aspidistra* plant with swordlike leaves in Daqingshan Mountain, Longzhou County, on the China-Vietnam border, and introduced it to the Guilin Botanical Garden for cultivation, it subsequently flowered in winter. After conducting a review of the relevant literature and specimen (Xi *et al.*, 2020; Lu *et al.*, 2020; Trinh *et al.*, 2021; Hsu *et al.*, 2022; Huang *et al.*, 2022; Tillich, 2023; Chinh *et al.*, 2023), and completed the cell chromosome experiment, we identified the *Aspidistra* plant as a new species eventually and reported it in here.

MATERALS AND METHODS

The chromosome preparation method was based on a modification of the wall degradation hypotonic method (Chen *et al.*, 1982; Wu *et al.*, 2013; Ye *et al.*, 2020). The

actively growing root tips were washed and transferred to ice water at 4°C for 24 h in the dark and then fixed in Carnoy's solution (ethanol: glacial acetic acid = 3: 1) at 4°C for 12 h. After washing with distilled water and soaking for 1 h at room temperature, the materials were transferred to enzyme solution (5% cellulase and 1% pectinase) and enzymatically digested at 37°C for 2 h. After removing the enzyme solution, the root tips underwent hypotonic treatment in distilled water for more than 1 h at room temperature, then were placed on slides and stained with 20 µL modified phenol magenta dye solution. The prepared tips were squashed for microscopic examination, the images were captured by an ortho-fluorescence microscope (Nikon ECLIPSE Ni-E, Japan), and then chromosomes were counted and analyzed by ImageJ software.

The best 5 metaphase photographs were taken for chromosome measurements. The data of relative chromosome length (including short arms and long arms) were obtained, the karyotype was analyzed with reference to the criteria of Li and Chen (1985), and the karyotype category was grouped according to the criteria of Levan *et al.* (1964). The average length of the chromosome, the karyotype intrachromosomal (Mean Centromeric Asymmetry, M_{CA}) as well as the interchromosomal (Coefficient of Variation of Chromosome Length, CV_{CL}) asymmetry indexes were calculated (Peruzzi and Eroğlu, 2013, Paszko, 2006).

TAXONOMIC TREATMENT

Aspidistra daqingshanensis Y.L.Pan & C.R.Lin, sp. nov. 大青山蜘蛛抱蛋 Figs. 1 & 2

Type: CHINA. Guangxi Zhuang Autonomous Region, Chongzuo City, Longzhou County, Binqiao town, Daqingshan mountain, near the top of the mountain,





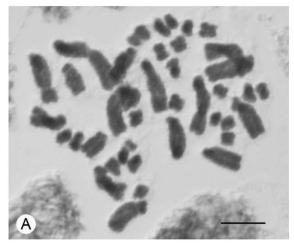
Fig. 1. Aspidistra daqinshanensis sp. nov. A. Plant habit; B. Berry; C-D. Flowers; E. Plants and showing leaf blade margin denticulate; F. Perianth dissected opened to show stamens and pistil; G. Stigma upper view and lower view; H. Flower side view; J. Pistil side view; J. Stamens.

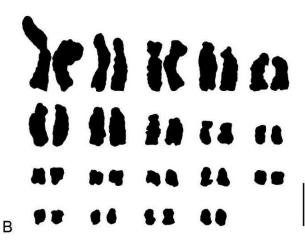




Table 1. Morphological comparison of Aspidistra daqingshanensis with A. fasciaria.

	A. daqingshanensis	A. fasciaria
Leaf	36–80×4–8 cm, distal margin denticulate	50-60×2.5-4.5 cm, entire at margin
Petiole	5–20 cm	10–17 cm
Tube		7–8 mm long, Ø 12–15 mm, white or sometimes with red spots on outside
Lobes	broadly ovate, 5-6×4-5mm, yellow, thickened at base	ovate-triangular, 7-8x3-4 mm, purplish red, verruculose at base
Stigma	Ø 9-10 mm, white, deep 4 lobed at margin, lobes apex	Ø 13 mm, purplish red, 3-5 lobed at margin, lobes apex
	slightly recurved	explanate





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Fig. 2. Somatic chromosomes at metaphase of **Aspidistra daqinshanensis** sp. nov. **A.** Photomicrograph of metaphase chromosome of 2n = 38; **B.** Somatic chromosomes serially arranged by their length and the position of centromeres. Karyotype formula: 2n = 22m + 10sm + 6st. Scale bar = 10 μm.

growing in evergreen broad-leaved forest, 106.7061E, 22.3064N, elev. 985 m, 1 April 2021. *Yan-Lin Pan & Lun-Fa Guo 210401* (holotype: IBK!; isotype: GXMG!).

Diagnosis: Aspidistra daqingshanensis is similar to A. fasciaria G.Z.Li (Lang et al., 1999; Fig. S1) with the leaf blade narrowly lanceolate and perianth shape, but differs by its leaf blade wider to 4–8 cm (vs. 2.5–4.5 cm), distal margin denticulate (vs. entire margin), perianth lobes broadly ovate (vs. ovate-triangular), yellow (vs. purplish red), adaxially thickened at base (vs. verruculose), entirely reddish to dark purplish red on the tube outside (vs. white or sometimes with red spots), stigma upper surface white (vs. purplish red), margin lobes slightly recurved (vs. explanate) at apex. A detailed comparison to distinguish the two species is presented in Table 1.

Description: Herbs perennial, evergreen. Rhizome creeping, epigeous, subterete, 8–10 mm thick, densely covered with nodes, roots numerous. **Sheathing leaves** 4–5, purplish red, 1–6 cm long, enveloping base of petiole, fibrous when withered. **Leaves** solitary or occasionally paired, 1–2.5 cm spaced; **petiole** stiffly upright, 5–20 cm long, 2–3 mm in diameter, adaxially sulcate; **leaf blade** narrowly lanceolate, 36–80 cm long, 4–8 cm wide, dark green, sometimes with pale green spots, apex acuminate, base cuneate, gradually narrowing into a petiole, inequilateral, distal margin denticulate, mid vein strongly prominent on abaxial surface, each half of lamina with 7–8 prominent secondary veins. **Peduncle** 5–10 mm long,

bracts 4-5, two of them adnate to flower base, broadly ovate, 6-8 mm long, 9-12 mm wide, purplish red, apex obtuse. Flower solitary; perianth campanulate, fleshy, 8lobed apically; perianth lobes explanate, usually yellowish, sometimes yellowish green or yellowish-white, broadly ovate to obround, 5–7 mm long, 4–5 mm wide at base, unequal, obtuse or blunt at apex, each with 3-4 parallel keels and thickened at base; **perianth tube** 7–9 mm long, 13-15 mm in diameter, entirely reddish to dark purplish red on both surfaces. Stamens 8, opposite lobes, inserted in base of perianth tube, filaments ca. 1 mm long, anthers oblong, pale yellow, ca. 2 mm long and 1.5 mm wide. Pistil umbrella-like shaped, 4-5 mm long, ovary inconspicuous, style short, white, cylindrical, ca. 2 mm long, 3–4 mm in diameter, **stigma** peltate, glabrous, 9–10 mm in diameter, cream-white and sometimes slightly reddish at margin, upper surface nearly flat, deeply 4lobed at margin, lobes emarginate at apex., lobes margin convoluted, apex slightly recurved. Fruit subglobose, 15-25 mm in diameter, brownish blue to black purple, surface irregular tuberculate.

Phenology: The new species was observed flowering from Oct. to Dec., and fruit to mature in Oct. next year.

Etymology: The specific epithet refers to the type locality, Daqingshan Mountain, Longzhou County in southwestern Guangxi, China.

Conservation status: At present, Aspidistra daqingshanensis is only known from the type locality in



southwestern Guangxi. Due to insufficient field investigation, the natural distribution of this species in the wild is not clear. Further field surveys on the China-Vietnam border may provide more information on its distribution. We here propose a status of 'Data Deficient' (DD) according to the IUCN Red List Categories (IUCN Standards and Petitions Committee, 2022).

Additional specimens examined (paratype): CHINA. Guangxi Zhuang Autonomous Region: Guilin City, Botany Garden of Guilin, taken to cultivation from the type locality, 08 December 2022, Chun-Rui Lin 1475 (IBK).

Karyology: The chromosome number of *Aspidistra daqingshanensis* was determined to be 2n = 38 (Fig. 2), with the karyotype formula of 2n = 38 = 22m + 10sm + 6st. Pairs 2th, 3th, 5th, 6th and 9th are sm-type chromosomes, pairs 4th, 7th and 8th are st-type, others are m-type. The average length of the chromosome is 7.49 μ m, and the karyotype asymmetry indexes M_{CA} and CV_{CL} are 35.57 and 55.93 respectively.

The cytotaxonomy result can provide a basis for the *Aspidistra* genus evolutionary study, to assist the identification and classification of this species. Cytological data in the *Aspidistra* genus have been reported (Bogner and Arnautov, 2004; Li, 2004; Qiao *et al.*, 2008; Hou *et al.*, 2009; Lin *et al.*, 2010, 2013; Liu *et al.*, 2011; He *et al.*, 2013; Meng *et al.*, 2014 Liu *et al.*, 2016), mostly of them have a chromosome number of 2n = 36 or 38. In this study, *Aspidistra daqingshanensis* is no exception with 2n = 38 chromosomes.

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