Kalanchoe ×gunniae Gideon F.Sm & Figueiredo (Crassulaceae), a new South African nothospecies derived from *Kalanchoe paniculata* Harv. × *Kalanchoe sexangularis* N.E.Br.

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Summary: The nothospecies Kalanchoe \times gunniae Gideon F.Sm & Figueiredo (Crassulaceae), a hybrid between Kalanchoe paniculata Harv. and Kalanchoe sexangularis N.E.Br., is described from Pretoria, South Africa.

Zusammenfassung: The nothospecies Kalanchoe ×gunniae Gideon F.Sm & Figueiredo (Crassulaceae), a hybrid between Kalanchoe paniculata Harv. and Kalanchoe sexangularis N.E.Br., is described from Pretoria, South Africa.

Introduction

In 1946 the Pretoria National Botanical Garden (NBG) was established on an isolated 75 hectare portion of the Experimental Farm of the University of Pretoria, after that parcel of land was transferred to what is today the South African National Biodiversity Institute (SANBI) (Smith et al., 1999). The Garden is dominated by an east-west aligned quartzite outcrop with both hot northern and cool south-facing slopes that accommodate about 1000 indigenous flowering plant species. Since its establishment, especially since 1989 when the then National Botanical Institute (NBI) came into being, Garden sections have been developed for public enjoyment, particularly those on the southern side of the outcrop. This has progressed inter alia by establishing expansive lawns and plant beds in which exclusively indigenous southern African flora is displayed.

One species that was introduced for display into the garden several decades ago, the leaf succulent *Kalanchoe sexangularis* N.E.Br. (Crassulaceae) (Figueiredo *et al.*, 2016), has escaped into the comparatively natural, undeveloped, northern parts of the garden. Here it has evidently hybridised with the locally indigenous *K. paniculata* Harv. (Smith *et al.*, 2003; Smith & Figueiredo, 2017a) to produce a genetically stable, self-perpetuating nothospecies that is ecologically successful.

We describe this hybrid derived from Kalanchoe paniculata × Kalanchoe sexangularis as the new nothospecies, Kalanchoe ×gunniae Gideon F.Sm & Figueiredo (Figure 1).

Background

In 1978 the indefatigable plant collector and succulent plant specialist David S. Hardy was then employed as a horticulturalist by the Botanical Research Institute (BRI) (Smith et al., 2016), one of the forerunners of SANBI. He collected material of Kalanchoe sexangularis N.E.Br. (Figure 2) from Wyllie's Poort in the Soutpansberg, Limpopo Province, northern South Africa, for cultivation in the Pretoria NBG. This material was later figured by botanical artist Rosemary Holcroft for The Flowering Plants of Africa (FPA) (Tölken, 1983). The living material collected by Hardy was vouchered by a series of at least three herbarium specimens that each consist of several black-and-white photographs fixed to specimen mounting boards. The photographs were taken by Adela Romanowski, the official photographer at the BRI. The information associated with what appears to be the main Hardy specimen is: SOUTH AFRICA. TRANSVAAL [LIMPOPO



Figure 1. Kalanchoe \times gunniae, the nothospecies derived from *K. sexangularis* and *K. paniculata*, growing in the Pretoria National Botanical Garden.



Figure 2. The red-leaved Kalanchoe sexangularis has become established in the comparatively undisturbed northern part of the Pretoria National Botanical Garden.



Figure 3. Kalanchoe paniculata growing on the Bronberg, Pretoria.



Figure 4. Two forms of *Kalanchoe rotundifolia* are indigenous to the Pretoria National Botanical Garden. The leaves of this one are light glaucous green and oval in outline.



Figure 5. A second form of *Kalanchoe rotundifolia* that occurs naturally in the Pretoria National Botanical Garden has deep green leaves that are round in outline.

PROVINCE]. —2229 (Waterpoort): "Soutpansberg; Wylliespoort [Wyllie's Poort]. Old road. In dense bush." (-DD), 1978. D.S. Hardy 5187 [PNBG No. 24511; PRE No. 57839] (PRE). Two of the photographic specimens note that the pictures of plants in flower were taken on "10.7.1979", while the living specimens themselves date from 1978. The Holcroft FPA plate itself is vouchered by a black-and-white habit sketch of "Kalanc rubra" [sic] that has the same PRE number, 57839, as the Hardy photographic specimens, but additionally dates the Hardy collection to "NOV." 1978, and notes that the FPA plate was prepared in "July 1979". The provisional name "Kalanchoe rubra" written on the Holcroft sketch is likely based on Kalanchoe rubinea Toelken (Tölken, 1978), a synonym of K. sexangularis (Tölken, 1985).

It is possible that other living material of *Kalanchoe sexangularis* was obtained for cultivation in the Pretoria NBG earlier than November 1978, but the Hardy gathering confirms that it has been grown in the Garden for at least the past 40 years.

Two species of *Kalanchoe*, *K. paniculata* (Figure 3) (sight records from the 1980s and 1990s by



Figure 6. The flowers of *Kalanchoe rotundifolia* growing in the Pretoria National Botanical garden vary from deep pink to orange, as here. Note how the corollas become distinctly twisted postanthesis.



Figure 7. *Kalanchoe luciae*, another southern African species known to hybridise with *K. sexangularis*.

one of [GFS]) and K. rotundifolia (Haw.) Haw. (photographic records from 2017 and 2018; Figures 4, 5, & 6) (Figueiredo & Smith, 2017; Smith & Figueiredo, 2017b), are known to be indigenous to the Pretoria NBG. Both species have a wide natural geographical distribution range, with K. rotundifolia extraordinarily so, as it ranges from the Eastern Cape of South Africa, through eastern South Africa and neighbouring Mozambique, northwards through parts of eastern Africa to the island of Socotra. Kalanchoe paniculata is endemic to southern Africa, virtually to South Africa, but with ou ____rs in Mozambique and Zimb = e, and occur in a broad, horseshoe-shaped althon both sides, and further north, of the Lesotho Drakensberg (Smith et al., 2003; Smith & Figueiredo, 2017a). Two further Kalanchoe species, K. luciae Raym.-Hamet and K. thyrsiflora Harv., have also been recorded from the greater

Γable 1. Morphological differe	nces among Kalanchoe sexc	angularis, K. ×gunnio	ae, and K. paniculata.
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#	Character	Kalanchoe sexangularis	Kalanchoe ×gunniae	Kalanchoe paniculata						
	Vegetative characters									
1	Stem colour	Green to deep wine- red Light green to yellowish green, often red-infused		Light green to yellowish green						
2	Stem angles	Often with several lengthwise running ridges, 4-angled at least on sterile shoots		Often with one or more lengthwise running ridges, round to slightly angled						
3	Leaf folding	Longitudinally folded Flat (older leaves) longitudinally folded (young leaves)		Spreading and gracefully recurved						
	Reproductive characters									
4	Colour of inflorescence axis	Reddish green to bright crimson red	Light green to yellowish green	Light green to yellowish green						
5	Pedicel length (mm)	4-8	5-6	4-5						
6	Sepal colour	Reddish green distinctly contrasting against corolla tube	Uniformly shiny light green hardly contrasting against corolla tube	Dull yellowish green hardly contrasting against corolla tube						
7	Sepal fusion	± Separate, basally adnate	Basally fused for ± 1mm	Basally fused for ± 1mm						
8	Corolla length (mm)	14-17	9–11	10-12						
9	Corolla shape (basally)	Somewhat enlarged	Distinctly enlarged	Somewhat enlarged						
10	Corolla tube length (mm)	13–16	8–11	9–10						
11	Corolla lobe shape	Ovate to suborbicular to somewhat pyriform	Orbicular	Deltoid-triangular						
12	Stamen exposure	Slightly exserted	Included	Included						
13	Filament length (mm)	3-5	2-3	3–5						
14	Carpel length (mm)	7–8	5-6	5-6						
15	Style length (mm)	3-7	2-3	2-3						
16	Scale colour	Reddish brown in upper half	Uniformly light green	Uniformly light green						
17	Fruit	Sepals drying early at base of fruit	Sepals remaining conspicuous at base of fruit	Sepals remaining conspicuous at base of fruit						
18	Lifespan	Perennial	Biennial to multi- annual, then monocarpic	Biennial to, more rarely, multi-annual, then monocarpic						

Pretoria area. However, we are not aware of records of their having occurred naturally on the grounds of the Pretoria NBG. With its large, soup plate-sized, strongly red-infused leaves, *Kalanchoe luciae* is today cultivated in the Garden to good effect (Figure 7).

The sight records of Kalanchoe paniculata in the Pretoria NBG are corroborated by two historical specimens of this species held in South Africa's National Herbarium: —2528CA, Pretoria, Pretoria National Botanical Garden, University Farm, *L.E.W Codd 3712*, 28 February 1942 (PRE); and —2528CB, Pretoria Distrik: (voorstad) Brummeria, klipperige randjie, suidelike helling [English: Pretoria district: (suburb) Brummeria, rocky ridge, southern slope], *P. Muller 260*, 10 February 1969 (PRE). Brummeria abuts the Pretoria NBG.

Material and Methods

Living material of Kalanchoe sexangularis and the Kalanchoe entity we here describe as K. ×gunniae were both studied in situ. The description of the vegetative and reproductive characters of K. ×gunniae was based on an examination of a broad cross-section of living specimens in the Pretoria NBG.

To estimate the genome size and detect possible major chromosomal mutations, if such exist, material of both Kalanchoe sexangularis and K. ×gunniae were subjected to flow cytometric analyses. Leaf material was tested first, but as the leaves of both K. sexangularis and K. ×gunniae accumulate mucilaginous compounds that affected the quality of the analyses, seeds were used as an alternative. Seed samples were obtained from one specimen of each of K. sexangularis and K. ×gun*niae* and were analysed directly in a flow cytometer. Seeds were co-chopped with leaves from the reference standard (Solanum lycopersicum 'Stupické'; 2C = 1.96 pg; Doležel et al., 1992) with a razor blade in a Petri dish containing 1ml of WPB (Loureiro et al., 2007). The nuclear suspension was then filtered through a 30µm nylon filter and nuclei were stained with 50mg.ml⁻¹ of propidium iodide, and 50mg.ml⁻¹ of RNase was added to prevent staining of double-stranded RNA. Samples were analysed in a Sy _ x CyFlow Space flow cytometer, with at least 1,000 particles per G₁ peak being acquired per sample using the Partec FloMax software v2.4d. The genome size in mass units 2C in pg; sensu Greilhuber et al., 2005) was obtained as follows: K. ×gunniae 2C nuclear DNA content (pg) = (K. × gunniae G_1 peak mean / reference standard G1 peak mean) * 1.96. Descriptive statistics of the genome size results obtained were based on five replicates of each species. Differences in genome size between species were evaluated with a One-Way ANOVA (SigmaPlot).

Results

Two similar-looking, but clearly different kalanchoes grow sympatrically in the undeveloped, comparatively 'natural', northern part of the Pretoria NBG. To the first one the name Kalanchoe sexangularis can be confidently and unambiguously applied as in both vegetative and reproductive characters it coincides exactly with the concept and circumscription of this distinctive, red-leaved species from eastern and northern South Africa, and slightly beyond (Figueiredo et al., 2016). Material of the other entity is in general appearance very similar to that of K. sexangularis, but in all vegetative and inflorescence respects (excluding flower characters) substantially larger than specimens of K. sexangularis (Table 1).

One possibility we considered is that the morphology of the more robust, second kalanchoe could be attributed to a chance chromosomal mutation in material of *Kalanchoe sexangularis*. However, flow cytometric analyses aimed at determining whether there are genome size differences between *K. sexangularis* and the second kalanchoe, indicated that they have the same genome size, i.e. 1.32pg/2C, and therefore no major karyological differences exist between them (Table 2). While such analyses do not produce chromosomal counts, the results obtained indicate that there is no difference between the number of chromosomes of *K. sexangularis* and the entity here described as *K. ×gunniae*.

Discussion

Results obtained from the flow cytometry analyses indicated that one or more chromosomal aberrations can be ruled out as the reason for the large size of Kalanchoe \times gunniae, as opposed to the similar-looking, but consistently much smaller K. sexangularis. Indeed, both taxa present the same average genome size, with values spanning the same range. Contrary to what has been observed in other taxonomic groups (see for example Morgan-Richards et al., 2004 on Hieracium subg. Pilosella [Asteraceae]; and Jeschke et al., 2003 on Amaranthus spp. [Amaranthaceae]) flow cytometric analyses of genome size in the current Kalanchoe study did not inform the putative detection of homoploid hybrids.

In addition, we have not encountered a species with the characters of *Kalanchoe* ×*gunniae* in the wild, nor in herbarium collections as preserved specimens. We considered the possibility that ma-

Table 2. Genome size results obtained from flow cytometric analyses* of *Kalanchoe sexangularis* and *Kalanchoe ×gunniae*.

#	Taxon	Genome size (pg/2C)					
		Mean	SD	CV (%)	Minimum	Maximum	n
1	Kalanchoe sexangularis	1.32	0.021	1.6	1.29	1.35	5
2	Kalanchoe ×gunniae	1.32	0.018	1.4	1.3	1.35	5

*The values are given as mean and standard deviation (SD) of the genome size (2C in picograms) of the individuals. Information of the CV value of the genome size estimations as well as of the minimum and maximum values is also provided. The number of analysed individuals (n) is also given. No statistically significant values at P < 0.05 were observed between the two entities.

terial described here as $K. \times gunniae$ might represent K. sexangularis var. intermedia (R.Fern.) R.Fern. (Fernandes, 1982: 99, 1983: 66), but this variety, which occurs naturally in south-tropical Africa and is absent from the five-country *Flora* of Southern Africa region (Namibia, Botswana, Swaziland, Lesotho, South Africa), has a much longer corolla (to 17mm long) and corolla tube (± 16mm long), leaf margins that usually lack crenations, and petioles that are short or absent.

We therefore conclude that the only realistic, alternative interpretation is that the second kalanchoe from the Pretoria NBG is of hybrid origin, here described as the nothospecies Kalanchoe \times gunniae. In such a case, K. sexangularis is an obvious candidate as one parent, and we argue that K. paniculata is the other. The Pretoria NBG is a known locality of Kalanchoe paniculata, even though a search did not reveal any specimens of this species in the Garden at present. The hybrid zone that one might anticipate (Harrison & Larson, 2014), with parental types, F_1 hybrids, and multiple generation hybrids and backcrosses present in varying proportions is not in evidence at the Pretoria site. Rather, introgression over at least 40 years seems to have progressed to the extent that genetic material of K. paniculata has been subsumed into that of the much more robust and clearly ecologically successful hybrid between it and *K. sexangularis*. This hybrid is, as far as is known, restricted to the Pretoria NBG and not known from places in eastern and northern South Africa where the two species, K. sexangularis and K. paniculata, occur sympatrically. It has seemingly not migrated from the Garden, as suitable habitats in the vicinity have been largely transformed into residential suburbia. Kalanchoe sexangularis is known to hybridise with other Kalanchoe species in cultivation and is one parent of *K*. 'Vivien', a cultivar described from a spontaneous horticultural hybrid between *K*. sexangularis and *K*. luciae, which was reported from Port Elizabeth, Eastern Cape, South Africa, a location very much distant from the natural geographical distribution ranges of both species (Bischofberger, 2015).

Previous examples of natural hybridisation in the genus *Kalanchoe* in southern African are not known to us, and were not even postulated by Tölken (1985) in his *Flora* treatment of the subcontinent. Further north, in the Flora zambesiaca region. Fernandes (1983: 51) speculated that the Mozambican K. fernandesii Raym.-Hamet might be a hybrid between K. lateritia Engl. and K. lanceolata (Forrsk.) Pers., by proceed that available material was too scanty to facilitate a firm conclusion. In the Flora of Tropical East Africa, Wickens (1987: 40, 51) included reference to two natural hybrids: between K. glaucescens Britten and K. nyikae Engl. subsp. auriculata Raadts, and between K. densiflora Rolfe and K. lanceolata, respectively. Researchers also have successfully managed to artificially hybridise genus members (Hamet, 1908: 38-39; Shaw, 2008), even across section boundaries (see for example Kuligowska et al., 2015). Although these authors identified hybridisation barriers in Kalanchoe at different time points during the hybridisation process, at both pre- and post-fertilisation levels, they were nonetheless able to generate intra- and inter-ploidy crosses. The production of seeds and subsequently plants was however, more successful within sections rather than across them; in the case of K. \times gunniae the postulated cross is within K. subg. Kalanchoe. Note that we prefer to treat the sections referenced by Kuligowska et al. (2015) as subgenera (Smith & Figueiredo, 2018).



Figure 8. *Kalanchoe* ×*gunniae* (foreground) is in all vegetative characters more robust than K. sexangularis (background).

Taxonomy and nomenclature of *Kalanchoe* ×gunniae

Kalanchoe ×gunniae Gideon F.Sm. & Figueiredo nothospec. nov. [=Kalanchoe paniculata Harv. × Kalanchoe sexangularis N.E.Br.]. Type: SOUTH AFRICA. GAUTENG PROVINCE. —2528 (Pretoria): Pretoria National Botanica Garden, 2 Cussonia Avenue, Brummeria, Pretoria, northern slope of quartzite koppie, (-CA), 20 June 2017, G.F. Smith & E. Figueiredo 57, (PRU; holo-).

Diagnosis: A robust, biennial to multi-annual, ultimately monocarpic, nothospecies that is intermediate between *Kalanchoe sexangularis* var. *sexangularis* and *K. paniculata*. It differs from *K. sexangularis* by being in all vegetative characters more robust (Figures 8 and 9); by having light green to yellowish green, red-infused, but not bright red, leaves when exposed to full irradiation (Figure 10); corollas that only reach a length of 11mm, not 17mm (Figure 11); and by ultimately being monocarpic. *Kalanchoe* ×gunniae differs from *K. paniculata* by having crenate, not entire, leaf margins (Figure 12), but the inflorescence structure of *K. ×gunniae* is similar to that of *K. paniculata* (Figure 13). In both *K. ×gunniae* and *K. paniculata* the fruit is subtended by persistent sepals (Figure 14); in the case of *K. sexangularis* the sepals shrivel soon after fruit has started to develop (Figure 15).

Description: Perennial, few-leaved, unbranched or very sparsely branched, glabrous, robust succulent, to 2.2m tall. *Stems* light green to yellowish green, few, unbranched, arising from a brittle, corky base, erect to leaning to creeping. *Leaves* opposite-decussate, petiolate, green to yellowish green to copper-coloured, infused with red when under environmental stress, succulent, spreading or recurved, coriaceous and papery on drying; *petiole* 5–50mm long, channelled above, leaves not clasping the stem; *blade* 6–14 × 4–10cm, broadly elliptic or obovate to oblong, often somewhat folded lengthwise, recurved in upper half; *apex* rounded-obtuse; *base* narrowly triangular to



Figure 9. A clump of *Kalanchoe* \times *gunniae* with plants averaging a height of 2m; *K. sexangularis* does not achieve such dimensions.



Figure 11. The corollas of Kalanchoe \times gunniae reach a length of only 11mm, not 17mm as in K. sexangularis.

cuneate; margins coarsely crenate or undulatecrenate into rounded, harmless, crenations. Inflorescence 50-150cm tall, erect to leaning, apically dense, many-flowered, flat-topped thyrse with several dichasia, round to somewhat ellipsoid in outline when viewed from above, branches opposite, slanted away from main axis at about 45°, subtended by leaf-like bracts, without, or more rarely with, leafy branchlets in axils, axis light green to yellowish green; pedicels slender, 5-6mm long. Flowers erect, greenish-yellow (tube) to bright yellow (lobes); *calyx* uniformly shiny light green, not strongly infused with small red spots; sepals 4, basally fused for $\pm 1 \text{ mm}, \pm 2 \times 1 \text{mm}, \text{tri-}$ angular-lanceolate, acute-tipped, hardly contrasting against corolla tube; corolla 9-11mm long, distinctly enlarged lower down, not twisted apically after anthesis, yellowish green and yellow; corolla tube 8-11mm long, distinctly 4-angled box-shaped-square when viewed from below, lon



Figure 10. Leaves of *Kalanchoe* \times *gunniae* are light green to yellowish green and red-infused, but not bright red as in K. sexangularis.



Figure 12. *Kalanchoe* ×*gunniae* differs from *K. paniculata* in presenting crenate rather than entire leaf margins.

gitudinally fluted above, greenish-yellow; *lobes* 2×2 mm, \pm circular, rounded at apex, apiculate, bright yellow, faintly brown-tipped. Stamens inserted well above the middle of the corolla tube, included; *filaments* 2–3mm long, thin, light green; anthers 0.3-0.5mm long, greenish-yellow. Pistil consisting of 4 carpels; *carpels* 5–6mm long, light green; *styles* 2–3mm long; *stigmas* very slightly capitate, whitish yellow; $scales \pm 1.0-1.5$ mm long, linear, light green throughout. Follicles basally box lantern-shaped, margins reddish infused, brittle, grass spikelet-like when dry, initially light green to dull whitish green, enveloped in light brownish white dry remains of corolla, later drying light to dark brown, 7-10mm long. Seeds 0.5-1.0mm long, reddish brown to dark brown, cylindrical to club-shaped to slightly bananashape curved. Chromosome number: unknown. Genome size: 1.32 pg/2C.



Figure 13. The inflorescences of Kalanchoe ×gunniae are reminiscent of those of K. paniculata.

Flowering time: *Kalanchoe* ×*gunniae* flowers from late-May to September, from early-winter to spring in the southern hemisphere, peaking from mid- to late-June to July.

Eponymy: *Kalanchoe* ×*gunniae* is named for Miss Mary Davidson Gunn [Kerriemuir, Forfarshire, Scotland, 16 May 1899-31 August 1989, Pretoria(?), South Africa] who developed the botanical library of SANBI in Pretoria into a worldrenowned facility. She worked for more than 55 years in the library, which was dedicated to her in 1969 and officially opened in 1970. Mary Gunn was an extraordinary librarian in every respect and through her commitment _____rengthen the Institute's publications holdings, for example, arranged for Field Marshal Jan Christiaan Smuts, South African Prime Minister, international statesman, and amateur botanist, to carry a copy of the rare Les Liliaceés by Pierre-Joseph Redouté back to the library in Pretoria in his personal luggage when he returned to South Africa from Europe in 1919 following the death of General Louis Botha (Beukes, 1996: 100). SANBI's scholarly journal Bothalia was named after General Botha.



Figure 14. The fruit of *Kalanchoe* \times *gunniae* is subtended by persistent sepals.



Figure 15. Once *muchoe sexangularis* reaches fruiting stage the state are shrivelled.

Mary Gunn was also an outstanding biographer who co-authored Gunn & Codd (1981), which is widely cited as a model for documenting historical and modern botanical exploration of any region or country. As far as we know Mary Gunn has never before been commemorated in the name of a plant.

Horticulture: *Kalanchoe* ×*gunniae* is a vigorous grower that requires virtually no aftercare once planted and established. It grows easily from its very fine seed, which can be broadcast in the place where the plants are intended to grow. Plants also do well in containers. During its vegetative phase, before it reaches flowering maturity, plants, both he large and small, transplant well.

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