

A karyotypic and anatomical study of an unidentified liliaceous plant

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ABSTRACT

An unidentified sterile liliaceous plant and three possible relatives were studied karyotypically and anatomically. All these plants have a somatic chromosome number of $2n = 14$. The possibility of the sterile plant having arisen as a result of a mutation appears unlikely, when compared with the possibility of hybrid origin. Chromosome morphology rules out *Bulbine latifolia* (L.f.) R. & S. and *Aloe arborescens* Mill. as possible parents. The sterile plant and *Aloe marlothii* Berger have similar karyotypes and, therefore, *A. marlothii* may be one of the parents. A close relationship between the sterile plant and the genus *Aloe* is further confirmed by their similar epidermal structure.

INTRODUCTION

A curious, liliaceous plant, apparently unknown to science, was found in a neglected garden in Pretoria during 1970. The plant has distichous leaves and a short almost prostrate stem and obviously belongs to the Liliaceae, but flowering material was required for a positive identification. The plant was transplanted to the Pretoria National Botanical Garden but, for the past 12 years, all attempts to induce flowering have proved unsuccessful and it therefore appears to be sterile.

The sterility of the plant may be due to genetic factors, or may indicate a hybrid origin. A search for possible parents revealed only three likely plants in close proximity to the sterile plant, namely *Aloe arborescens* Mill. *A. marlothii* Berger and *Bulbine latifolia* (L.f.) R. & S. This investigation attempted, by karyotypic and anatomical methods, to establish whether hybridization between any of these species could have resulted in this unidentified plant.

MATERIALS AND METHODS

Root tip material of *Aloe arborescens* (Hardy 6045), *A. marlothii* (Henderson 352), *Bulbine latifolia* (Mauve 4995) and the supposed hybrid (Hardy 6055) were collected in the Pretoria National Botanical Garden and pretreated in monobromonaphthalene at room temperature. After four hours, the root tips were washed in running water and fixed in Carnoy fixative (Darlington & La Cour, 1976) for 24-36 hours. After hydrolysis for 10 minutes at 60°C in 1N HCl, the material was stained in leucobasic fuchsin (modified formula after Coleman, 1938; Darlington & La Cour, 1976) for 2 hours. The darkly stained parts were then squashed in aceto-orcein (La Cour, 1941). Slides were made permanent by placing them in a fridge for ± 10 minutes, removing the coverslip with a needle, dehydrating through an alcohol series and mounting in Euparal. Ten cells per plant were studied.

Leaf segments were boiled for 10 minutes in concentrated HNO_3 to prepare epidermal peels. The epidermis was separated from the rest of the leaf during this process and was removed from the debris, stained in Methylene Blue, dehydrated in alcohol and mounted in Euparal.

Voucher specimens of the material studied are lodged in the National Herbarium, Pretoria.

RESULTS

(a) *Bulbine latifolia*

A somatic chromosome number of $2n = 14$ for this species, as reported by Snoad (Darlington & Wylie, 1955) and Jones & Smith (1967), was confirmed. The haploid karyotype (Fig. 1) consists of two large metacentric to submetacentric, two large subtelocentric and three small subtelocentric chromosomes. An idiogram (Fig. 1a) has been compiled from the data given by Jones & Smith (1967). The results of the present study are shown in Fig. 1b.

The two idiograms illustrate small differences in the karyotypes. The present study revealed the second chromosome pair to be submetacentric, whereas the published data show a metacentric chromosome pair. The fourth chromosome pair was found to be of similar length to the third pair, but with the centromeric index slightly lower. Although Jones & Smith also found the fourth pair to be more subtelocentric than the third, the two pairs could also be distinguished by a marked variation in length. The third difference observed in this study lies in the presence of satellites on the long arms of the fifth chromosome pair. No satellites were reported by Jones & Smith (1967).

These karyotype differences between the published and observed data indicate the existence of chromosomal variability in *B. latifolia*. A cytotoxic study of this species should prove valuable.

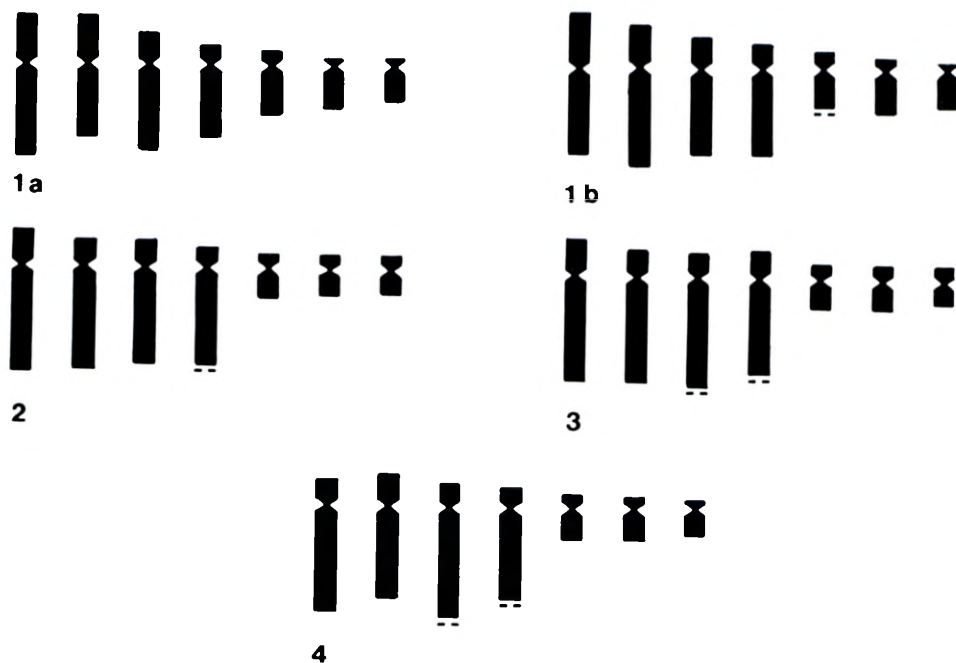
(b) *Aloe arborescens*

A somatic chromosome number of $2n = 14$ was observed and confirms reports in the literature (Taylor, 1925; Ferguson, 1927; Resende, 1937; Muller, 1941; Snoad, 1951; Sharma & Mallick, 1966). The haploid idiogram (Fig. 2) illustrates four long subtelocentric chromosomes and three short subtelocentric ones. Satellites are present on the distal part of the long arms of the fourth chromosome pair. Apart from the SAT-region on the fourth chromosome pair, no secondary constrictions, as described by Sharma & Mallick (1966), were observed.

(c) *Aloe marlothii*

A somatic chromosome number of $2n = 14$ was found and agrees with reported observations (Resende, 1937; Riley, 1959). The haploid idiogram (Fig. 3) shows four long and three short subtelocen-

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FIGS 1-4.—Haploid idiogram. 1, *Bulbine latifolia*: a, compiled from Jones & Smith, 1967; b, this study. 2, *Aloe arborescens*. 3, *A. marlothii*. 4, sterile plant.

tric chromosomes. Satellites were seen on the distal part of the long arms of the third and fourth chromosome pairs. The karyotypes of *A. marlothii* and *A. arborescens* correspond in all respects, except for the satellites on the third chromosome pair.

(d) The unknown liliaceous plant

This plant has 14 chromosomes per somatic cell. All chromosomes are subtelocentric with eight long and six short chromosomes (Fig. 4). The third and fourth chromosome pairs have satellites on the distal parts of the long arms. These satellite-bearing chromosomes have slightly larger arm ratios than the other long chromosomes (Fig. 4). The karyotype suggests that the chromosomes might be arranged in seven homoeologous pairs with no, or very small differences being visible between the chromosomes within a pair.

Morphology and epidermal structure. The small distichous-leaved plant produces suckers. The mother plant is c. 60 mm tall with c. 9 pairs of smooth, linear-acuminate, succulent, unequal leaves, the longest c. 60 mm long and c. 10 mm broad with a few small teeth at the apex. The morphology of the different plants can be seen in Figs 5-8.

The epidermis of *Bulbine latifolia* consists of elongated quadrangular cells (Fig. 9). Both the *Aloe* species, and the sterile plant, have hexagonal epidermal cells. Epidermal cells of *A. arborescens* (Fig. 10) are smaller than the cells of *A. marlothii* (Fig. 11) and the sterile plant (Fig. 12).

DISCUSSIONS AND CONCLUSIONS

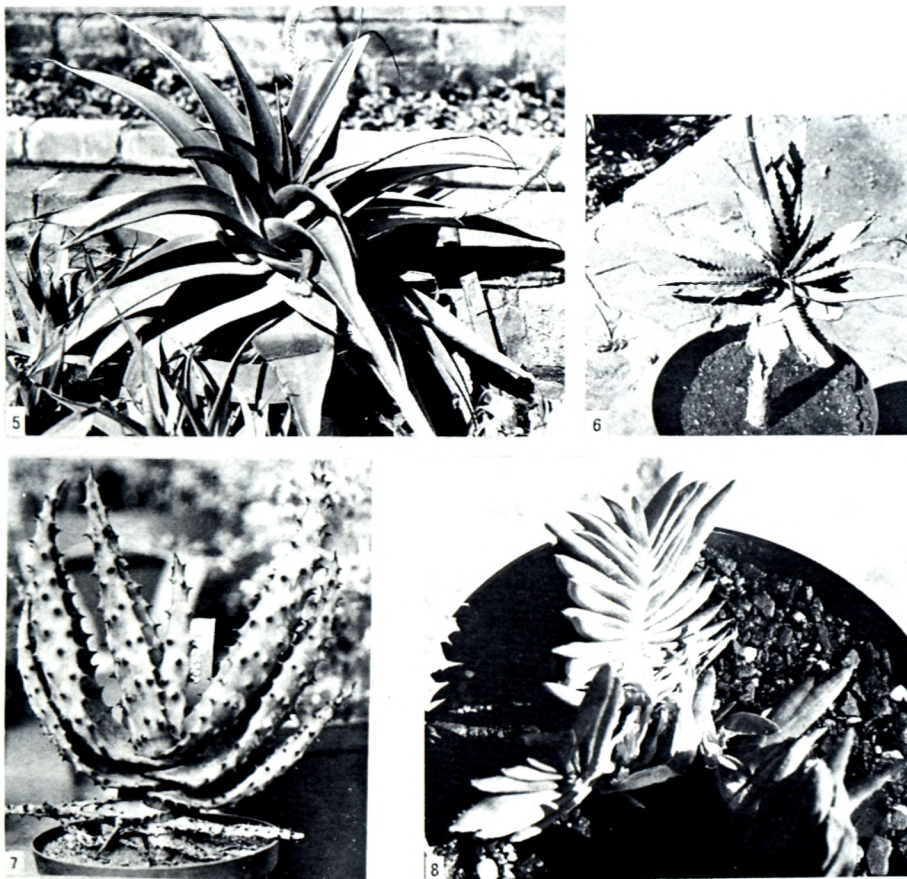
The sterile plant might represent an undescribed species or may have originated by hybridization or by mutation. If it is a hybrid, the parents must have occurred in the vicinity of the hybrid and the hybrid will have one genome from each parent. Since all possible parental species in close proximity to the sterile plant have been investigated, the similarity between genomes might have an indication of relationships.

A comparison of the haploid idiogram of the possible hybrid with that of *Bulbine latifolia* reveals large differences. The presumed hybrid has no chromosomes that are nearly metacentric as in *B. latifolia* and the short-satellited chromosomes of *B. latifolia* are absent in the hybrid. If *B. latifolia* was one of the parental species, the resulting hybrid must have received at least two large metacentric chromosomes and a short chromosome with satellites from this source. This observation indicates that *B. latifolia* could not have been one of the parents. Further evidence for this is illustrated by the epidermal structure, where *B. latifolia* has a totally different epidermal structure from the hybrid which has a typical *Aloe* pattern (Figs 9-12).

The epidermal structure of the hybrid corresponds closely with the patterns found in *Aloe arborescens* and *A. marlothii* (Figs 10-12). A closer relationship between the hybrid and the Aloes studied is further confirmed by the similarity in their karyotypes. However, as the hybrid has two satellited chromosome pairs and *A. arborescens* only one, *A. arborescens* is an unlikely parent. The karyotypes of *A. marlothii* and the hybrid agree in almost all respects. The only observed differences were in the arm ratios of the first two chromosome pairs. These small differences might result from a statistically inadequate sample, or from the fact that the first two pairs are of almost similar length and the first and second pair might be interchanged in the presumed hybrid.

If the sterile plant is the product of mutation, the parents must have a karyotype similar to *A. marlothii*. The chances of a mutation transforming a fertile plant, a few metres tall, to a 6 cm sterile plant and hard thorny leaves to smooth fleshy leaves at the same time seems very unlikely. The suggestion of hybridization seems more probable.

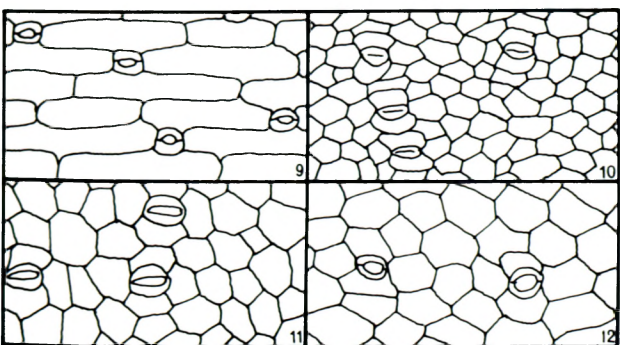
The restricted growth habit and absence of flowers in the sterile plant suggest hybridization rather than a new species. The similarity between the sterile plant and *A. marlothii* with regard to



FIGS 5-8.—Photographs of studied plants. 5, *Bulbine latifolia*; 6, *Aloe arborescens*; 7, *A. marlothii*; 8, sterile plant.

karyotype and epidermal structure make it reasonable to suggest that *A. marlothii* might be one of the parents. Although no other possible parent was found in the vicinity of the supposed hybrid plant, a hybrid origin for the sterile plant is still possible. The second parent might have died by the time the hybrid was discovered. It is also possible that the hybrid was introduced into the area where it was found.

The results of this study indicate, therefore, that the sterile plant is a hybrid between *A. marlothii* and another species of *Aloe*.



FIGS 9-12.—Drawings of epidermal patterns. 9, *Bulbine latifolia*; 10, *Aloe arborescens*; 11, *A. marlothii*; 12, sterile plant.

UITTREKSEL

'n Ongeïdentifiseerde steriele plant van die familie Liliaceae en drie moontlik verwante plante is kariatopies en anatomies bestudeer. Al hierdie plante het 'n somatiese chromosoomgetal van $2n = 14$ gehad. Die waarskynlikheid dat die steriele plant 'n

baster is, is groter as die moontlikheid van 'n mutasie oorsprong. Op grond van chromosoomorfologie word *Bulbine latifolia* (L.f.) R & S. en *Aloe arborescens* Mill. as moontlike ouers uitgeskakel. Die steriele plant en *A. marlothii* Berger het ooreenstemmende kariatipes en daarom kan *A. marlothii* een van die ouers wees. Die verwantskap tussen die steriele plant en die genus *Aloe* word verder bevestig deur ooreenstemmende epidermale strukture.

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