

## Meiotic analysis of two putative polyploid species of Verbenaceae from Brazil.

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**Abstract** — The natural occurrence of hybrids and polyploids hinders the precise identification of some taxa in Verbenaceae (Lamiales). Since meiotic studies are useful for understanding the chromosomal behavior of plant species, we have examined the meiotic stages and chromosomal behavior of *Lippia alba* (Mill) N. E. Brown and *Lantana camara* Plum. in Brazil. *Lippia alba* and *Lantana camara*, had normal stages of meiosis, including interphase, leptotene, zygotene, pachytene, diplotene, initial diakinesis, diakinesis, metaphase I, anaphase I, telophase I, prophase II and telophase II. Fifteen and 22 bivalents were observed during diplotene and diakinesis in *L. alba* and *L. camara*, respectively. The chromosomes segregated normally during anaphase I, telophase I and II. The regular meiotic behavior seen in *L. alba* and *L. camara* indicated that polyploidy was established by a diploid-like behavior in meiosis, although this did not exclude a possible diploid origin.

**Key words:** Chromosomal behavior, *Lantana*, *Lippia*, meiosis, Verbenaceae.

### INTRODUCTION

The relationships among several species of Verbenaceae have been extensively discussed and different taxonomic arrangements have been proposed (TRONCOSO 1974; BOTTA 1980; SILVA 1999; SALIMENA 2000; 2002). These divergences indicate that the taxonomic position of Verbenaceae species remains unresolved (RAJ 1983; SINHA and SHARMA 1984). Additionally, the natural occurrence of hybrids and polyploids hinders the precise classification of some taxa (SPIES 1984; SANDERS 1987; OJHA and DAYAL 1992; RUEDA 1993; SANDERS 2001).

Meiotic studies have helped to confirm the relationship between polyploidy and highly irregular meiosis in several plant species (NIRMALA and KAUL 1993; MILLER and VENABLE 2000; STACE 2000). There have been few meiotic studies in the Verbenaceae and some of these have reported abnormal chromosomal behavior. ANDRADA *et al.* (1998) observed meiotic abnormalities in *Lippia fissicalyx* Tronc., including univalents, multiva-

lents and a high frequency (82.4%) of abnormal pollen and suggested a relationship among these characteristics and the level of ploidy. In *Lantana* L. species, an unequal distribution of chromosomes has been observed during anaphase (SINHA and SHARMA 1984; SINHA *et al.* 1995). CORAZZA-NUNES *et al.* (1995) also reported abnormal segregation of the chromosomes and spindle alterations in meiotic cells of *Aloysia lycioides* Cham.

In this report, we provide the first description of the meiotic stages and the chromosomal behavior of *Lippia alba* and *Lantana camara*, both of which are part of the Brazilian flora.

### MATERIAL AND METHODS

The anthers of *L. camara* and *L. alba* were collected in the Botanical Experimental Area of the Institute of Biological Sciences at the Federal University of Juiz de Fora, Minas Gerais State, Brazil. Meiotic cells were obtained as described by BRANDAO *et al.* (2000). About 100 cells in each stage of meiosis were analyzed and identified using meiotic images of *Hordeum*, *Artemisia* and *Zea mays*, as described by SINGH (1993) and FUKUI AND NAKAYAMA (1996).

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## RESULTS

Only normal stages were observed, in the meiotic cells of *L. alba* and *L. camara*, including interphase, leptotene, zygotene, pachytene, diplotene, initial diakinesis, diakinesis, metaphase I, anaphase I, telophase I, prophase II and telophase II (Figs. 1a-k and 2a-l, respectively). Fifteen and 22 bivalents were observed during diplotene and diakinesis in *L. alba* and *L. camara*, respectively (Figs. 1e-g and 2e-g). In both species, the chromosomal segregation during anaphase I and telophase I and II was normal (Figs. 1i-l and 2i-l).

## DISCUSSION

In all sexually reproducing organisms, meiosis is a complex process that helps to maintain the chromosomal number constant from generation to generation and ensures the operation of Mendel's laws of heredity (SINGH 1993; WENDEL 2000). The halved chromosomal number and the occurrence of genetic recombination are the main features in diploidy and in the evolutionary success of sexual reproduction in eukaryotes (MILLER and VENABLE 2000; MERCIER *et al.* 2001). Meiosis also has an important role in repairing genetic defects

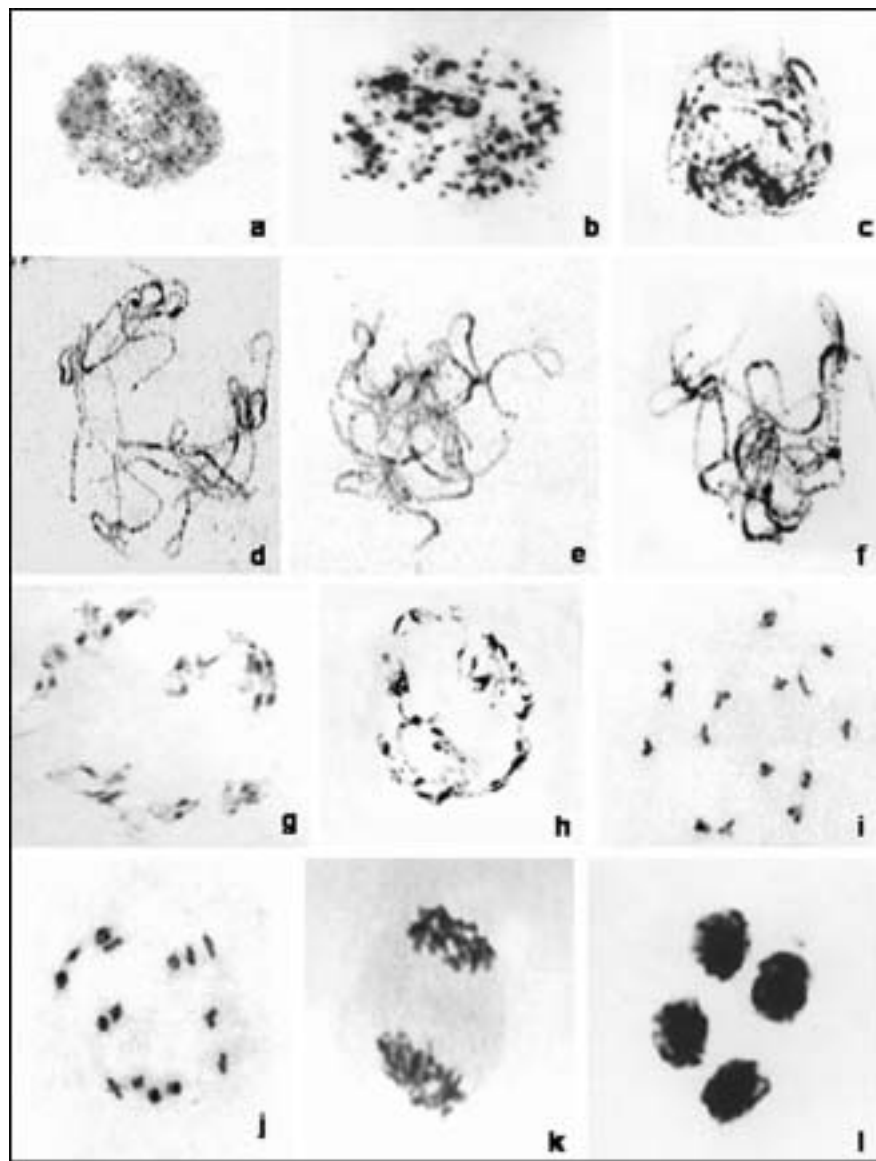


Fig 1 — Meiotic cells of *L. alba*: interphase (a), leptotene (b), zygotene (c), initial pachytene (d), middle pachytene (e), final pachytene (f), initial diplotene (g), middle diplotene (h), diakinesis (i), metaphase I (j), anaphase I (k) and telophase II (l). Bar = 10  $\mu$ m.

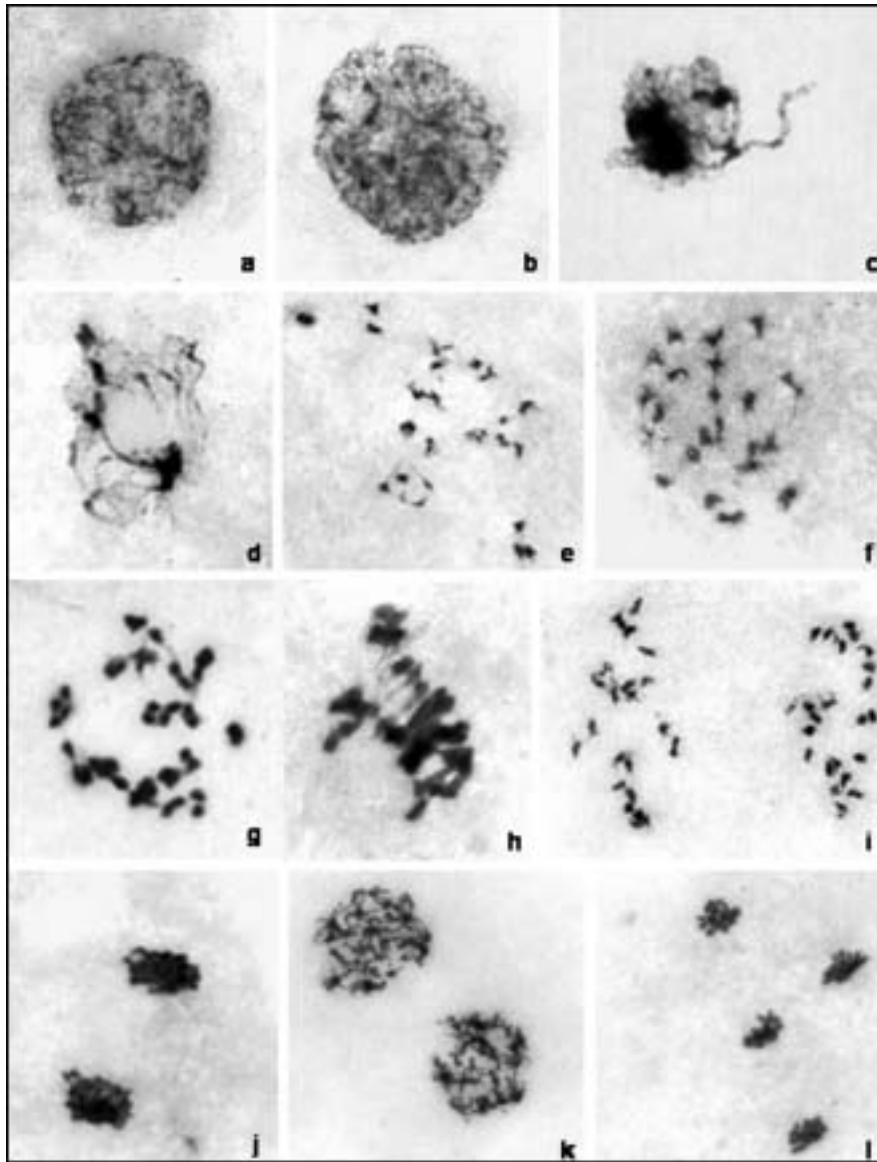


Fig. 2 — Meiotic cells of *L. camara*: interphase (a), leptotene (b), zygotene (c), pachytene (d), diplotene (e), initial diakinesis (f), diakinesis (g), metaphase I (h), anaphase I (i), telophase I (j), prophase II (k) and telophase II (l). Bar = 10  $\mu$ m.

in germ line cells (HOLLIDAY 1984; FUKUI and NAKAYAMA 1996). Meiotic studies allow the analysis of a large variety of chromosomal alterations, such as polyploidy, which is very common in plants (OTTO and WHITTON 2000). Non-reduced gametes can be identified by their meiotic configuration during chromosomal pairing and can allow the detection and accurate determination of chromosomal rearrangements. Such information can be used to construct cytogenetic maps and determine the evolutionary relationships among related species (DE WET 1979; STEVENS and BOUGOURD 1991; FUKUI and NAKAYAMA 1996; SEIJO and FERNANDEZ 2001).

QU *et al.* (1998) used meiotic analysis to study *Vaccinium* species and observed tetrasomic inheritance, as well as the formation of bivalents and multivalents by random pairing among four homologues and a high fertility, all in the same newly formed hybrid. Studies with *Lantana camara* populations from South Africa, the Caribbean and India have shown that this species is tetraploid ( $x = 11$ ) and always has uni-, bi-, tri- and multivalents, (TJIO 1948; SEN and SAHNI 1955; RAGHAVAN and ARORA 1960; ARORA 1961; SINHA and SHARMA 1984; SPIES 1984; SANDERS 1987; OJHA and DAYAL 1992; SINHA *et al.* 1995).

In *Lippia* L., the high degree of ploidy has been related to the occurrence of chromosomal abnormalities and, consequently, to abnormal development of the seeds (POGGIO *et al.* 1993). No such abnormalities were seen in the present study.

Based on the putative polyploidy hypothesis proposed by SANDERS (2001) for both species (*Lippia alba* - hexaploid and *Lantana camara* - tetraploid), our data suggest that natural selection may have eliminated the problems caused by polyploidization and produced a normal meiotic chromosomal behavior. The meiotic success of both species could be explained by processes such as genic control of the pairing of distinct genomes in tetraploids (two) and hexaploid (three), or the high degree of homology between the genomes (RILEY and CHAPMAN 1958; SYBENGA 1996; DAGNE 2001). The regular meiotic behavior seen in *L. alba* and *L. camara* indicated a polyploid state sustained by diploid-like behavior at meiosis (DE WET 1979; WIDEN and WIDEN 2000; OZKAN *et al.* 2001; OLEARY *et al.* 2004) although a possible diploid origin can not be excluded.

**Acknowledgements** — The authors thank Rosana Pezzin, Mateus R. Pereira and Patricia M. O. Pierre for collecting the specimens of *Lippia alba* and *Lantana camara*. This work was supported by FAPESP (proc. no. 00/11842-2 - scholarship to A. D. Brandão), FAPEMIG (grant no. CBB 2728/98) and FINEP (grant no. 0930/01).

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Received XII.02.2004; accepted X.10.2005