CHROMOSOME STUDIES ON AFRICAN PLANTS. 13. *LACHENALIA MUTABILIS*, *L. PUSTULATA* AND *L. UNICOLOR*

The genus *Lachenalia* Jacq.f. ex Murray consists of small bulbous geophytes and shows a great potential for use as pot plants (Niederwieser *et al.* 1997). Various chromosome numbers, and even different basic chromosome numbers, have been reported for this genus (Moffett 1936; De Wet 1957; Riley 1962; Mogford 1978; Ornduff & Watters 1978; Nordenstam 1982; Crosby 1986; Hancke & Liebenberg 1990; Hancke 1991; Johnson & Brandham 1997; Kleynhans 1997; Hancke & Liebenberg 1998; Kleynhans & Spies 1999).

Lachenalia mutabilis Sweet belongs to the *L. orchioides* group (Crosby 1986) and the chromosome numbers reported for this species vary from 2n = 10

(Ornduff & Watters 1978; Johnson & Brandham 1997) to 2n = 14 (De Wet 1957; Johnson & Brandham 1997) and 56 (De Wet 1957).

Lachenalia pustulata Jacq. and *L. unicolor* Jacq. are closely related species, growing in the southwestern Cape. Both species are classified as part of the *L. unicolor* group (Crosby 1986), and Duncan (1988) suggested that these species might be combined, due to their morphological similarities. Somatic chromosome numbers of 2n = 16 have been reported for both species by De Wet (1957), Hancke (1991), Johnson & Brandham (1997) and Kleynhans (1997).

The aim of this paper is to determine the chromosome numbers of the three species and to determine whether any correlation exists between chromosome number and geographical distribution.

MATERIALS AND METHODS

Specimens were collected in the field throughout the geographical distribution area of each species and vegetative material of each plant collected was grown separately in the nurseries of ARC Roodeplaat and the Department of Botany and Genetics, University of the Orange Free State, Bloemfontein. Voucher specimens are housed in the Geo Potts Herbarium (BLFU) at the university (Table 2).

Bulbs were grown on Deco gel™ and actively growing root tips were cut and placed in water at 4°C for 24 hours. The root tips were fixed in Carnoy's (1886) fixative for 24–36 hours, hydrolysed in 1N HCl at 60°C for 7 minutes and stained with 0.5% leucobasic fuchsin for two hours (Darlington & LaCour 1976). Squashes were made in 1% aceto-orcein (Darlington & LaCour 1976). Slides were permanently mounted in Euparal (Darlington & LaCour 1976).

RESULTS AND DISCUSSION

Lachenalia mutabilis: this species is morphologically extremely variable. It is naturally distributed throughout Namaqualand to south of Riviersonderend (Duncan 1988). During this study 35 specimens, representing 16 populations have been studied cytogenetically. The specimens studied represent the central and northern geographical distribution area of this species (Figure 2).

Somatic chromosome numbers of 12 (Figure 3A, B), 14 (Figure 3C–G) and 24 (Figure 3H) were observed during this study (Table 2). The numbers based on x = 7 confirm a previous report on this species (De Wet 1957). The numbers based on x = 6 present a new basic chromosome number for this species. Although collected in the same geographical area as the two specimens studied by Ornduff & Watters (1978), no specimens with x = 5 were observed during this study. Six further specimens from unknown localities with 2n = 10 were studied by Johnson & Brandham (1997).

Different specimens collected in a given locality showed no variation in chromosome number. Specimens

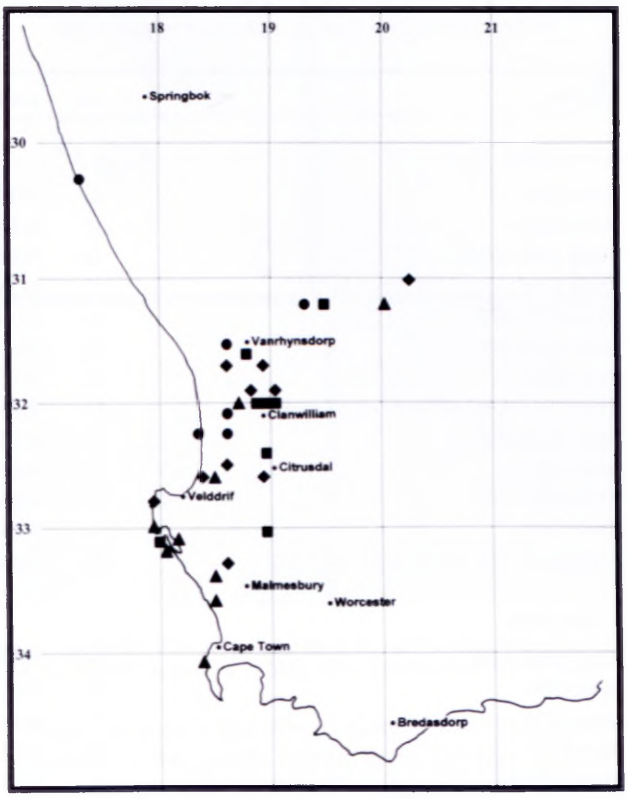

FIGURE 2.—Geographical distribution of the *Lachenalia* populations. *Lachenalia mutabilis* specimens, x = 6, ●; x = 7, ■. *L. pustulata*, ▲; *L. unicolor*, ◆.

with lower chromosome numbers were mostly collected in the northern and western areas of the geographical distribution area of this species (Figure 2). However, an insufficient number of specimens has been studied to test the validity of this observation.

Three hypotheses regarding the origin of the different basic chromosome numbers in one species can be postulated: 1, the original basic chromosome number is 5 and the misidentification of B-chromosomes is responsible for the higher basic numbers described; 2, the original basic chromosome number is 7 and dysploidy leads to the formation of lower basic chromosome numbers; or 3, an aneuploid series occurs.

The presence of B-chromosomes in various *Lachenalia* species has been reported by Hancke & Liebenberg (1990) and Johnson & Brandham (1997). Initial meiotic studies indicate that the same number of chromosomes is present in the anthers and in the root tips used during this study. The initial meiotic study also indicates normal meiosis with the formation of bivalents only. Therefore, we reject the hypothesis that misinterpretation of B-chromosomes attributed to the 'different basic chromosome numbers' observed in this species.

Johnson & Brandham (1997) suggested that the different basic chromosome numbers in *L. mutabilis* can be attributed to Robertsonian translocations resulting in dysploidy. However, the chromosome morphology of the specimens varies. Most specimens contain 4 to 8 very short chromosomes. The number of short chromosomes varies between different localities and even between specimens collected at the same locality. There is no indication of longer chromosomes (as a result of Robert-

TABLE 2.—Somatic chromosome numbers of *Lachenalia* specimens with their voucher numbers and localities. Specimens are listed according to their locality from north to south and from west to east

Voucher	2n	Locality
L. mutabilis		
Spies 6750	24	NORTHERN CAPE.—3017 (Hondeklipbaai): near Hondeklipbaai, (–AD).
Spies 6744	12	WESTERN CAPE.—3118 (Vanrhynsdorp): near Vanrhynsdorp, (–DA).
Spies 6748, 6753	14	WESTERN CAPE.—3118 (Vanrhynsdorp): near Vanrhynsdorp, (–DA).
Spies 6746	12	NORTHERN CAPE.—3119 (Calvinia): on top of Vanrhyns Pass, (–AC).
Spies 6757–6761	14	NORTHERN CAPE.—3119 (Calvinia): along road to Oorlogskloof, (–AC).
Spies 6774, 6775	12	WESTERN CAPE.—3218 (Clanwilliam): Clanwilliam Nature Reserve, (–AB).
Spies 6751	12	WESTERN CAPE.—3218 (Clanwilliam): near Elandsbaai, (–AD).
Spies 6747, 6767–6770, 6773, 6776–6778	14	WESTERN CAPE.—3218 (Clanwilliam): near Clanwilliam, (–BB).
Spies 6779–6781	12	WESTERN CAPE.—3218 (Clanwilliam): near Sandberg, (–BC).
Spies 6745	14	WESTERN CAPE.—3219 (Wuppertal): near Citrusdal, (–CA).
Spies 6784, 6785	14	WESTERN CAPE.—3318 (Cape Town): Langebaan Nature Reserve, (–AA).
Spies 6752	14	WESTERN CAPE—3318 (Cape Town): near Porterville, (–BB).
Spies 6749, 6754–6756, 6782, 6783	14	Unknown.
L. pustulata		
Spies 6806, 6807	16	NORTHERN CAPE.—3119 (Calvinia): Oorlogskloof, (–AC).
Spies 6792	16	WESTERN CAPE.—3218 (Clanwilliam): Algeria, (–BB).
Spies 6789	16	NORTHERN CAPE.—3220 (Sutherland): near Aurora, (–DC).
Spies 6788	16	WESTERN CAPE.—3317 (Saldanha): near Saldanha, (–BB).
Spies 6808–6811	16	WESTERN CAPE.—3318 (Cape Town): Langebaan, (–AA).
Spies 6798–6804, 6816–6823	16	WESTERN CAPE.—3318 (Cape Town): Langebaan Nature Reserve, (–AA).
Spies 6790	16	WESTERN CAPE.—3318 (Cape Town): near Postberg, (–AA).
Spies 6791	16	WESTERN CAPE.—3318 (Cape Town): near Darling, (–AD).
Spies 6797	16	WESTERN CAPE.—3318 (Cape Town): Mamre Road, (–BC).
Spies 6787	16	WESTERN CAPE.—3318 (Cape Town): near Kampsbaai, (–CD).
Spies 6786, 6793, 6795, 6796, 6805, 6812–6815	16	Unknown.
Spies 6794	16/32	Unknown.
L. unicolor		
Spies 6827	16	WESTERN CAPE.—3118 (Vanrhynsdorp): Vredendal, (–CB).
Spies 6828	16	WESTERN CAPE.—3118 (Vanrhynsdorp): Unionskraal, (–DB).
Spies 6831	16	NORTHERN CAPE.—3119 (Calvinia): Nieuwoudtville, (–AC).
Spies 6853–6856	16	WESTERN CAPE.—3217 (Vredenburg): Abdolsbaai, (–DD).
Spies 6829	16	WESTERN CAPE.—3218 (Clanwilliam): Verlorevlei, (–AD).
Spies 6857	16	WESTERN CAPE.—3218 (Clanwilliam): near Clanwilliam, (–BB).
Spies 6837, 6838	16	WESTERN CAPE.—3218 (Clanwilliam): Velddrif, (–CC).
Spies 6825	16	WESTERN CAPE.—3218 (Clanwilliam): Piketberg, (–DD).
Spies 6844–6846	16	NORTHERN CAPE.—3220 (Sutherland): Aurora, (–DC).
Spies 6835	16	WESTERN CAPE.—3318 (Cape Town): Darling, (–AD).
Spies 6833, 6834, 6843	16	WESTERN CAPE.—3318 (Cape Town): Porterville, (–BB).
Spies 6830	16	WESTERN CAPE.—3319 (Worcester): Tulbach, (–AC).
Spies 6824, 6826, 6832, 6836, 6839–6842, 6847–6852	16	Unknown.

sonian translocations) in any specimen or in specimens with a particular basic chromosome number. Therefore, no evidence supports the second hypothesis.

At this stage it seems as if an aneuploid series exists in *L. mutabilis*. To test this hypothesis, thorough meiotic studies, including hybrids between some of these specimens, should be completed. The use of *in situ* hybridization techniques will also help to determine the mode of chromosome evolution in this species. Simultaneously, the species delimitation should be investigated to determine whether *L. mutabilis* represents one variable species or more than one species, each with its own basic chromosome number.

Polyploidy was observed in a specimen with x = 6 during this study and De Wet (1957) observed it in one with x = 7. The tetraploid number obtained during this study was restricted to a single bulb. All other bulbs obtained from the original specimen proved to be diploid. The polyploidization process occurred consequently, during cultivation. In contrast, De Wet (1957) described an octoploid specimen, suggesting that various polyploidization processes occurred in nature. More specimens should be studied before any conclusions regarding polyploidy in this species can be made. Polyploidy is relatively scarce in this species with only two specimens (5%) exhibiting this phenomenon. Further studies are needed to determine the mode of chromosome evolution in *L. mutabilis*.

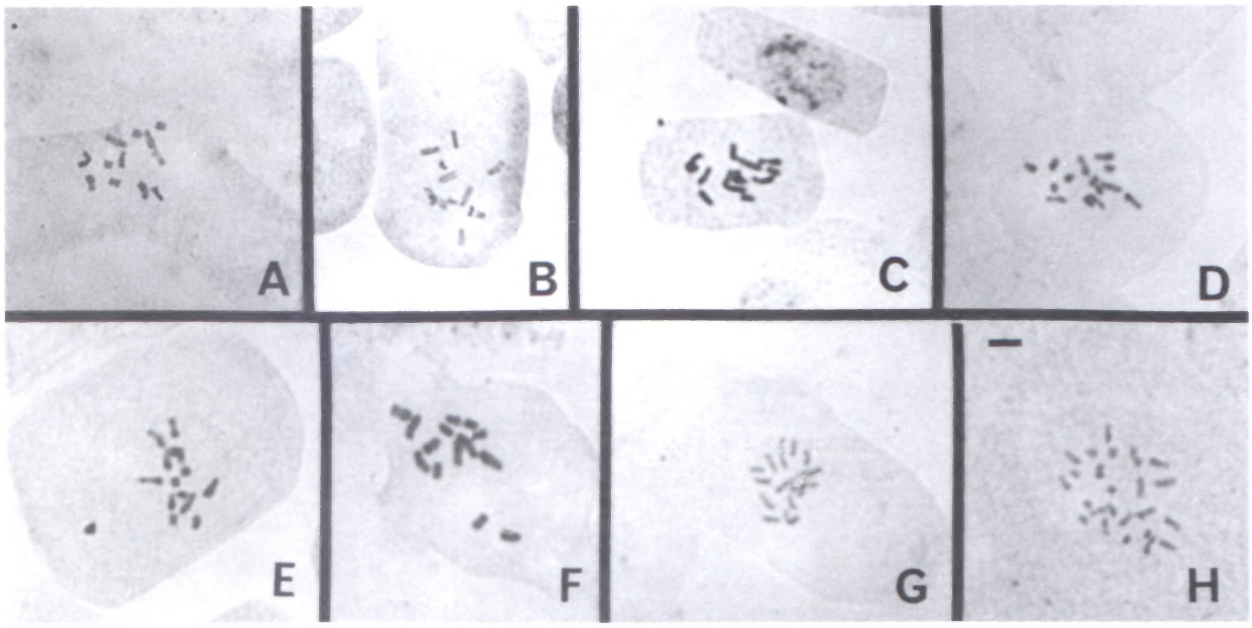

FIGURE 3.—Mitotic chromosomes in some *Lachenalia mutabilis* specimens. A, *Spies 6751*, 2n = 12; B, *Spies 6744*, 2n = 12; C, *Spies 6760*, 2n = 14; D, *Spies 6748*, 2n = 14; E, *Spies 6757*, 2n = 14; F, *Spies 6789*, 2n = 14; G, *Spies 6749*, 2n = 14; H, *Spies 6750*, 2n = 24. Scale bar: 6.5 µm.

Lachenalia pustulata and *L. unicolor*: during this study somatic chromosome numbers of 38 *L. pustulata* specimens, representing at least 10 different localities, and 34 *L. unicolor* specimens, representing at least 12 localities, were determined (Table 2). All specimens were diploid with 2n = 2x = 16 (Figure 4), except one *L. pustulata* specimen, *Spies 6794*, which was diploid (19 cells) and a single tetraploid cell was observed. This study confirms published reports on a chromosome number for *L. pustulata* (Johnson & Brandham 1997; Kleynhans 1997) and *L. unicolor* (De Wet 1957; Hancke 1991; Johnson & Brandham 1997; Kleynhans 1997).

The karyotypes of the two species correspond. Both species have four chromosomes that are significantly longer than the other chromosomes. The chromosome numbers of the species are the same. In order to determine whether Duncan's (1988) suggestion of combining the two species is correct, meiotic chromosome behaviour of crosses between these species and pollen viability of the hybrids should be studied.

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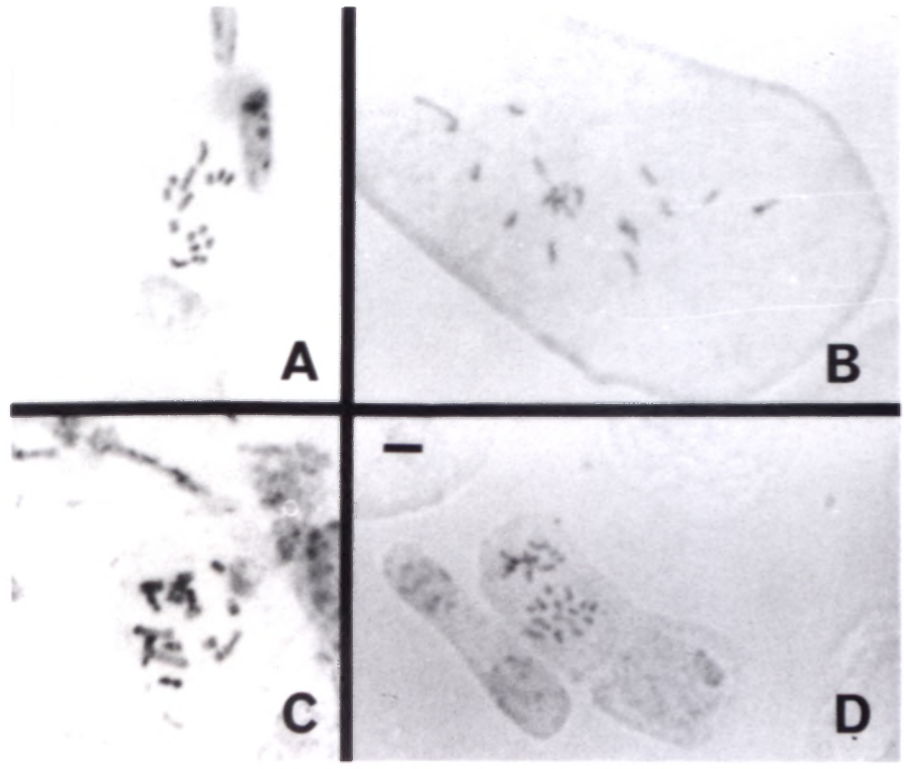

FIGURE 4.—Mitotic chromosomes in some *Lachenalia pustulata* and *L. unicolor* specimens. A, B, *L. pustulata*: A, *Spies 6803*, 2n = 16; B, *Spies 6798*, 2n = 16. C, D, *L. unicolor*: C, *Spies 6838*, 2n = 16; D, *Spies 6840*, 2n = 16. Scale bar: 6.5 µm.

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