**Differential amphiplasty and its recovery in hybrid
offspring of *Crepis capillaris* × *C. vesicaria*
ssp. *taraxacifolia***

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ABSTRACT

A study was undertaken to determine the relationship between differential amphiplasty and chromosome complements in hybrid offspring of *C. capillaris* × *C. vesicaria* ssp. *taraxacifolia*. In cells of offspring plants with somatic chromosome complement of *capillaris* and *vesicaria*, the satellite of 4v chromosome of *vesicaria* disappeared. In a plant without 1c chromosome of *capillaris* ($2n=10=2c+3c+1v+1v+2v+2v+3v+3v+4v+4v$), disappearance of the satellite of 4v chromosome was observed. The satellite of 4v chromosomes which had disappeared in F_1 hybrid reappeared in the B_1 offspring by eliminating the chromosomes of *capillaris*. Further, it was confirmed that differential amphiplasty recovered under the condition of nucleus substitution. As the result, 2c or 3c chromosome of *capillaris* may have induced differential amphiplasty.

1. INTRODUCTION

Differential amphiplasty was observed in 18 hybrids among various species in the genus *Crepis* by Navashin (1927, 1928, 1934), Avery (1930), Hollingshead (1930), Babcock and Emsweller (1936), and Sherman (1946). On the basis of these data, the hierarchy of dominance under the conditions of differential amphiplasty was presented in a figure by Wallace and Langridge (1971), but they failed to report on the hybrid between *capillaris* and *vesicaria* ssp. *taraxacifolia*. According to Navashin (1934) the amphiplastic change is reversible. In this study, differential amphiplasty in F_2 , B_1 , and B_2 offsprings of *Crepis capillaris* × *C. vesicaria* ssp. *taraxacifolia* was observed and its recovery was cytogenetically examined.

2. MATERIALS AND METHODS

F_1 hybrids were obtained by crossing between stigma of *C. capillaris* (or *vesicaria* ssp. *taraxacifolia*) and pollen of *C. vesicaria* ssp. *taraxacifolia* (or *capillaris*) in the net room. The chromosome complement of *capillaris* was $2n=6=1c+1c+2c+2c+3c+3c$, that of *vesicaria* ssp. *taraxacifolia* was

$2n=8=1v+1v+2v+2v+3v+3v+4v+4v$ and that of F_1 was $2n=7=1c+2c+3c+1v+2v+3v+4v$ (Ikeda-Komatsu and Tanaka, 1983; Ikeda-Komatsu, the accompanying paper).

In order to obtain F_2 achenes, the eight F_1 hybrids of *capillaris* $\times$ *vesicaria* were isolated, and then intercrossed by open pollination. The F_2 achenes obtained from open pollination were sown, and eleven F_2 seedlings were raised.

B_1 offspring of (*capillaris* $\times$ *vesicaria*) $\times$ *vesicaria* was obtained by crossing between stigma of F_1 hybrid (*capillaris* $\times$ *vesicaria*) and pollen of *vesicaria*. The other B_1 offspring were obtained by the same method.

B_2 offspring of [(*capillaris* $\times$ *vesicaria*) $\times$ *vesicaria*] $\times$ *vesicaria* was produced by crossing between stigma of B_1 offspring [(*capillaris* $\times$ *vesicaria*) $\times$ *vesicaria*] with $2n=11=1c+2c+3c+1v+1v+2v+2v+3v+3v+4v+4v$ and pollen of *vesicaria*.

Aceto-orcein staining for root tips were carried out according to the methods described (Tanaka and Komatsu, 1977; Komatsu and Tanaka, 1978; Ikeda-Komatsu and Tanaka, 1983). Each chromosome was identified by aceto-orcein staining.

3. RESULTS

Various offspring plants from F_1 hybrids between *capillaris* and *vesicaria* were obtained by different crossings. Table 1 shows the chromosome complements and the maximum number of nucleoli per nucleus in them.

F₂ hybrid: (capillaris $\times$ *vesicaria)* $\times$ (*capillaris* $\times$ *vesicaria*)

The chromosome number of F_2 hybrids was $2n=6, 7, 9, 10$, or 11 . The plant with $2n=6$ had the same complement as *capillaris*. The plants with $2n=7$ had a similar chromosome complement as F_1 hybrid. Differential amphiplasty had developed in $4v$ chromosome. In $2n=9$ plant, differential amphiplasty had developed in $4v$ chromosome (Fig. 1). All of the 4 plants with $2n=10$ was composed of two sets of *capillaris* and one set of *vesicaria* chromosomes. Differential amphiplasty developed in $4v$ chromosomes. The $2n=11$ plants were all composed of one set of *capillaris* chromosomes and two sets of *vesicaria* chromosomes. Differential amphiplasty occurred in two $4v$ chromosomes.

B₁ offspring: (capillaris $\times$ *vesicaria)* $\times$ *vesicaria*

Twenty three backcross offspring plants of *capillaris* $\times$ *vesicaria* F_1 hybrid to *vesicaria* were obtained. The chromosome number of these plants was $2n=7, 10$, or 11 . The $2n=7$ plants had the same chromosome complement as F_1 hybrid. Differential amphiplasty occurred in $4v$ chromosomes. Eight plants with $2n=10$ were classified to five kinds of complements (Fig. 2A-C). One of the plants with $2n=10$ had the chromosome complement without $1c$ chromosome of *capillaris* (Fig. 2C). Differential amphiplasty occurred in $4v$ chromosomes of all eight

Table 1. Chromosome complements and the maximum number of nucleoli per nucleus in hybrid offspring of *Crepis capillaris* and *C. vesicaria* spp. *taraxacifolia*

Generation	Chromosome number	Chromosome complement	Max. No. of nucleoli/nucleus	No. of plants
F ₂ : cv.cv	6	1c+1c+ <u>2c</u> +3c+3c	2	1
	7	1c+ <u>2c</u> +3c+1v+2v+3v+4v*	1	2
	9	1c+ <u>2c</u> + <u>2c</u> +3c+3c+1v+2v+3v+4v*	2	1
	10	1c+1c+ <u>2c</u> + <u>2c</u> +3c+3c+1v+2v+3v+4v*	2	4
	11	1c+ <u>2c</u> +3c+1v+1v+2v+2v+3v+3v+4v*+4v*	1	3
B ₁ : cv.v	7	1c+ <u>2c</u> +3c+1v+2v+3v+4v*	1	7
	10	1c+ <u>2c</u> +3c+1v+2v+2v+3v+3v+4v*+4v*	1	1
		1c+ <u>2c</u> +3c+1v+1v+2v+3v+3v+4v*+4v*	1	1
		1c+ <u>2c</u> +3c+1v+1v+2v+2v+3v+4v*+4v*	1	4
		1c+ <u>2c</u> +3c+1v+1v+2v+2v+3v+3v+4v*	1	1
		<u>2c</u> +3c+1v+1v+2v+2v+3v+3v+4v*+4v*	1	1
	11	1c+ <u>2c</u> +3c+1v+1v+2v+2v+3v+3v+4v*+4v*	1	6
		1c+ <u>2c</u> +3c+1v+2v+2v+3v+3v+4v*+V ₁ +V ₂	1	1
		1c+ <u>2c</u> +3c+1v+2v+2v+3v+4v*+4v*+V ₃ +V ₄	1	1
B ₁ : cv.c	6	1c+1c+ <u>2c</u> + <u>2c</u> +3c+3c	2	2
	7	1c+ <u>2c</u> +3c+1v+2v+3v+4v*	1	2
	10	1c+1c+ <u>2c</u> + <u>2c</u> +3c+3c+1v+2v+3v+4v*	2	1
	11	1c+ <u>2c</u> + <u>2c</u> +3c+3c+2v+2v+3v+3v+4v*+4v*	2	1
B ₁ : vc.v	7	1c+ <u>2c</u> +3c+1v+2v+3v+4v*	1	7
	8	1c+ <u>2c</u> +3c+1v+2v+3v+4v*+V ₅	1	1
		1v+1v+2v+2v+3v+3v+ <u>4v</u> + <u>4v</u>	2	4
B ₂ : cv.v.v	8	1v+1v+2v+2v+3v+3v+ <u>4v</u> + <u>4v</u>	2	3
	9	1c+ <u>2c</u> +3c+1v+2v+2v+3v+4v*+4v*	1	1
	11	1c+ <u>2c</u> +3c+1v+1v+2v+2v+3v+3v+4v*+4v*	1	6
	12	1c+ <u>2c</u> +3c+1v+1v+1v+2v+2v+3v+3v+4v*+4v*	1	1

cv.cv: (*capillaris* × *vesicaria*) × (*capillaris* × *vesicaria*)cv.v: (*capillaris* × *vesicaria*) × *vesicaria*cv.c: (*capillaris* × *vesicaria*) × *capillaris*vc.v: (*vesicaria* × *capillaris*) × *vesicaria*cv.v.v: [(*capillaris* × *vesicaria*) × *vesicaria*] × *vesicaria*

Underline shows the SAT-chromosome.

The symbol * shows the chromosome incurred differential amphiplasty. V_n shows a variant chromosome.

plants. Eight plants with 2n=11 were distinguished to three kinds of complements. Six out of eight plants had the same chromosome complement but remaining two were different with respect to having two variant chromosomes (V₁-V₂ and V₃-V₄) (Fig. 2D, E). Differential amphiplasty occurred in 4v chromosomes of all eight plants.

Fig. 1. Chromosome complements of the plant with $2n=9$ in F_2 hybrids: $(capillaris \times vesicaria) \times (capillaris \times vesicaria)$. The arrow indicates the region of chromosome incurred differential amphiplasty. The bar indicates $5 \mu m$.

Fig. 2. Chromosome complements of B_1 offspring: $(capillaris \times vesicaria) \times vesicaria$. A-C, $2n=10$. D, E, $2n=11$. A, plant with two $4v$ chromosomes of *vesicaria*. B, plant with a $4v$ chromosome of *vesicaria*. C, plant without $1c$ chromosome of *capillaris*. D, E, plant with the variant chromosomes. The arrows indicate the regions of the chromosomes incurred differential amphiplasty. The bar indicates $5 \mu m$.

*B*₁ offspring: (*capillaris* × *vesicaria*) × *capillaris*

Six plants were obtained. They had the chromosome number of $2n=6, 7, 10$ (Fig. 3A), or 11 (Fig. 3B). Differential amphiplasty occurred in 4v chromosomes of all the plants.

Fig. 3. Chromosome complements of *B*₁ offspring: (*capillaris* × *vesicaria*) × *capillaris*. A, $2n=10$. B, $2n=11$. The arrows indicate the region of the chromosomes incurred differential amphiplasty. The bar indicates 5 μm

*B*₁ offspring: (*vesicaria* × *capillaris*) × *vesicaria*

Twelve plants were obtained. They had the chromosome number of $2n=7$ or 8. The $2n=7$ plants had the same chromosome complement as *F*₁ hybrid. Differential amphiplasty occurred in 4v chromosomes of all seven plants. One of five plants with $2n=8$ had the same chromosome complement of *F*₁ hybrid and variant chromosome (*V*₅) (Fig. 4A). Differential amphiplasty occurred in 4v chromosome. The remaining four plants with $2n=8$ had the same chromosome complement as *vesicaria* (Fig. 4B). No differential amphiplasty occurred in these plants.

Fig. 4. Chromosome complements of *B*₁ offspring: (*vesicaria* × *capillaris*) × *vesicaria*. A, B, $2n=8$. A, plant with a variant chromosome. The arrows indicate the region of the chromosomes incurred differential amphiplasty. The bar indicates 5 μm .

B₂ offspring: [(capillaris × vesicaria) × vesicaria] × vesicaria

Eleven plants were obtained. They had the chromosome number of $2n=8$, 9 (Fig. 5B), 11, or 12. Cytoplasm of these plants was owned by *capillaris* as a result of crossing. The $2n=8$ plants had the same chromosome complement as *vesicaria* (Fig. 5A). Nucleus substitution occurred in *B₂* offspring for the first time. Differential amphiplasty had not occurred. Six plants with $2n=11$ were the same chromosome complement as the female plant. Differential amphiplasty occurred in all 4v chromosomes of the plants with $2n=9$, 11, or 12.

Fig. 5. Chromosome complements of *B₂* offspring: *[(capillaris × vesicaria) × vesicaria] × vesicaria*. A, $2n=8$. B, $2n=9$. A, plant with substituted nucleus. The arrows indicate the region of the chromosomes incurred differential amphiplasty. The bar indicates 5 μ m.

4. DISCUSSION

It has been reported that differential amphiplasty had developed in 4v chromosome in the *F₁* hybrid of *capillaris* × *vesicaria* ssp. *taraxacifolia* and their reciprocal *F₁* hybrid (Ikeda-Komatsu and Tanaka, 1983). In this study, 4v chromosome was also incurred differential amphiplasty in various offspring plants from *F₁* hybrids between *capillaris* and *vesicaria*.

Navashin (1934) has reported that dominance of differential amphiplasty was evident in trihybrid, and the most dominant species extinguished the secondary constrictions of the other two species. In this study, allotriploids were obtained in *B₁* offspring *[(capillaris × vesicaria) × vesicaria]* and *B₂* offspring *[(capillaris × vesicaria) × vesicaria × vesicaria]*. In the plant having 1 set of *capillaris* chromosomes plus 2 sets of *vesicaria* chromosomes, it was observed that two secondary constrictions of *vesicaria* had disappeared. It was further observed that the satellite of *vesicaria* disappeared in the plant having 2 sets of *capillaris* chromosomes and 1 set of *vesicaria* chromosomes, and that in every hybrid derivatives the direction of differential amphiplasty was defined. This

was similar to Hollingshead's results (1930) on allotriploid and amphidiploid of *capillaris* and *tectorum*.

In recent years Lacadena and his co-workers (Orellana et al., 1983; Cermeño et al., 1984a, 1984b; Lacadena et al., 1984a, 1984b; Santos et al., 1984) have ascertained that it was a single nucleolar chromosome which induced differential amphiplasty, and that nucleolar competition was evident among the nucleolar chromosomes in the clone having some nucleolar chromosomes. Regretfully, in this study it could not be ascertained which chromosome induced the differential amphiplasty. In the plant without 1c chromosome of *capillaris* ($2n=10=2c+3c+1v+1v+2v+2v+3v+3v+4v+4v$) which appeared in B_1 offspring of (*capillaris* × *vesicaria*) × *vesicaria*, differential amphiplasty occurred in two 4v chromosomes of *vesicaria*. This confirmed that 1c chromosome of *capillaris* did not trigger differential amphiplasty. Probably 2c or 3c chromosome may have induced differential amphiplasty, or it may have been induced by the reciprocal interaction of these two.

On the other hand, the number of the SAT-chromosomes corresponded to the maximum number of nucleoli at interphase (Tabel 1). This shows that the secondary constriction of SAT-chromosome involves the organization of nucleolus.

Navashin (1934) has pointed out that differential amphiplasty is a reversible phenomenon. This was also confirmed in the present study. The secondary constriction of 4v chromosome which had disappeared in F_1 hybrid reappeared again in B_1 offspring by eliminating the chromosomes of *capillaris*. Cytoplasm of these plants owned by *vesicaria*. Furthermore, the plants with substituted nucleus, cytoplasm of *capillaris*, and nucleus of *vesicaria*, were obtained in B_2 offspring. In these plants, differential amphiplasty was restored. Thus, reversibility was not influenced by cytoplasm.

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