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Crossability and cytology of hybrid progenies in the cross between *Brassica campestris* and three wild relatives of *B. oleracea, B. bourgeaui, B. cretica* and *B. montana*

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Summary

Crossability and cytology were examined in F_1, F_2, B_1 and hybrid plants of F_1 hybrids of *Brassica campestris* and three wild relatives of *B. oleracea, B. bourgeaui, B. cretica* and *B. montana,* respectively. The F_2 plants were obtained after self- and open pollination of the F_1 hybrids. The B_1 and hybrid plants were produced after the F_1 hybrids backcrosses with *B. campestris* and crossed with *B. napus,* respectively. After crossing the F_1 hybrids, many seeds of the F_2, B_1 and hybrid plants were harvested. Multivalent formation was high in the chromosome configuration for the PMCs of F_2, B_1 and hybrid plants, suggesting that crossing over might occur between them. Many different types of aneuploids were obtained in the progenies of the F_2, B_1 and hybrid plants. It is suggested that different types of normal egg cells may be produced by one-by-one or little-by-little chromosome addition. The possibility is discussed of gene transfer from *B. bourgeaui, B. cretica* and *B. montana,* to cultivated plants, *B. campestris* and *B. napus.*

Introduction

Wild relatives are often a valuable source of genes in crop improvement. *Brassicinae* consists of five genera, *Brassica, Diplotaxis, Eruca, Erucastrum* and *Sinapis* (Mizushima, 1952), *Brassica* being the most important. It is thus essential to investigate the possibility of gene transfer from the wild relatives of the crucifer to *Brassica* crops. Interspecific and intergeneric hybridization in the crucifers is not easy because of cross incompatibility and hybrid lethality. With the development of embryo rescue and somatic hybridization techniques, many interspecific and intergeneric hybrids have been obtained among crucifers (see Bajaj, 1990; Inomata, 1990; Pelletier, 1990; Vambing & Glimelius, 1990): however, little information is available on the progeny of F_1 hybrids.

There are nine subspecies and six varieties in *B. campestris* and *B. oleracea,* respectively (Hosoda, 1961). Hybrid production between them was very difficult using artificial pollination (Sarashima, 1964), but many interspecific hybrids have been produced through ovary culture (Inomata, 1977, 1978, 1985b, 1990; Matsuzawa, 1978). Crosses of the F_1 hybrids showed that unreduced normal egg cells are produced (Inomata, 1983). Different types of the progenies were obtained in the F_2, B_1 and hybrid plants in the cross of F_1 hybrids × *B. napus.* In these hybrids, a *B. campestris*-type plant with 20 chromo-

Table 1. Crossability of the F_1 hybrids of *Brassica campestris* and three wild relatives of *B. oleracea, B. bourgeaui, B. cretica* and *B. montana*

Cross combination of the F_1 hybrid[1]	Self-pollination		Open pollination		F_1 × *B. campestris*[2]		F_1 × *B. napus*[3]	
	No. of flowers pollinated	No. of seeds obtained	No. of flowers pollinated	No. of seeds obtained	No. of flowers pollinated	No. of seeds obtained	No. of flowers pollinated	No. of seeds obtained
ssp. *chinensis* × *B. bourgeaui*	139	0	585	14	145	2	122	1
ssp. *pekinensis* × *B. bourgeaui*	99	0	537	2	232	1	219	3
ssp. *chinensis* × *B. cretica*	95	0	122	8	118	3	108	3
ssp. *trilocularis* × *B. cretica*	0	–	3	0	8	0	9	24
ssp. *pekinensis* × *B. cretica*	150	0	465	58	112	2	112	16
ssp. *chinensis* × *B. montana*	75	0	403	0	157	0	140	4
ssp. *pekinensis* × *B. montana*	9	0	132	3	148	0	186	2

[1] Cultivars of *B. campestris* were ssp. *chinensis* cv. Seppaku-taina, ssp. *trilocularis* cv. Brown Sarson DS-2 and ssp. *pekinensis* cv. Nozaki-hakusai No. 2
[2] Cultivars of *B. campestris* used in the present experiment were the same as the production of the F_1 hybrid
[3] Cultivar of *B. napus* was ssp. *oleifera* cv. Miho-natane

somes, a *B. oleracea*-type plant with 18 chromosomes, and a *B. napus*-type plant with 38 chromosomes appeared. The genes of the *B. campestris* and *B. oleraceae* can exchange reciprocally, and the genes of both species can be introduced into *B. napus* (Inomata, 1983, 1991). Previous papers have reported the production of many interspecific hybrids between *B. campestris* and three wild relatives of *B. oleraceae – B. bourgeaui, B. cretica* and *B. montana* – through ovary culture (Inomata, 1985c, 1986, 1987).

The present paper deals with the crossability of the F_1 hybrids of *B. campestris* and *B. bourgeaui, B. cretica* and *B. montana.* It also reports the chromosome configuration for the PMCs of F_2, B_1 and hybrids in the cross of F_1 hybrids × *B. napus* and, moreover, the crossability of the F_2 plants, the B_1 plants from the F_1 hybrids backcrossed with *B. campestris* and the hybrid plants from the F_1 hybrid crossed with *B. napus.* Chromosome numbers are reported for some of the progenies of the F_2, B_1 and hybrid plants harvested.

Materials and methods

The F_1 hybrids used in the present experiment were obtained by ovary culture in the cross between *B. campestris* and *B. bourgeaui, B. cretica* and *B. montana,* three wild relatives of the *B. oleracea* (Inomata, 1985c, 1986, 1987), viz. *B. bourgeaui* 120, *B. cretica* ssp. *cretica* 35 and *B. montana* 89 (S. Snoge-

Table 2. Seed setting of the F_1 hybrid and chromosome number of the F_2, B_1 and hybrid plants derived from the F_1 hybrids of *Brassica campestris* and three wild relatives of *B. oleracea, B. bourgeaui, B. cretica* and *B. montana*

Cross combination	No. of seeds obtained[1]	No. of seeds sown	No. of plants examined	Crhomsome number in root tip cell ($2n$)[2]
Open pollination	85	38	26	25(1), 28(2), 29(4), 34(1), 36 & 58 (1), 38(2), 46(2), 48(1), 48 & 56 (1), 53 & 56 (1), 54 (1), 56 (7), ? (2)
F_1 × *B. campestris*[3]	8	8	7	29 (3), 48 (2), ? (2)
F_1 × *B. napus*[4]	53	29	20	38 (13), 42 (1), 47 (2), ? (4)

[1] Refer to Table 1
[2] Number in parentheses shows the number of plants observed and ? shows a chromosome number that could not be determined
[3] *B. campestris* contains three subspecies, *chinensis, trilocularis* and *pekinensis*
[4] *B. napus* contains one subspecies

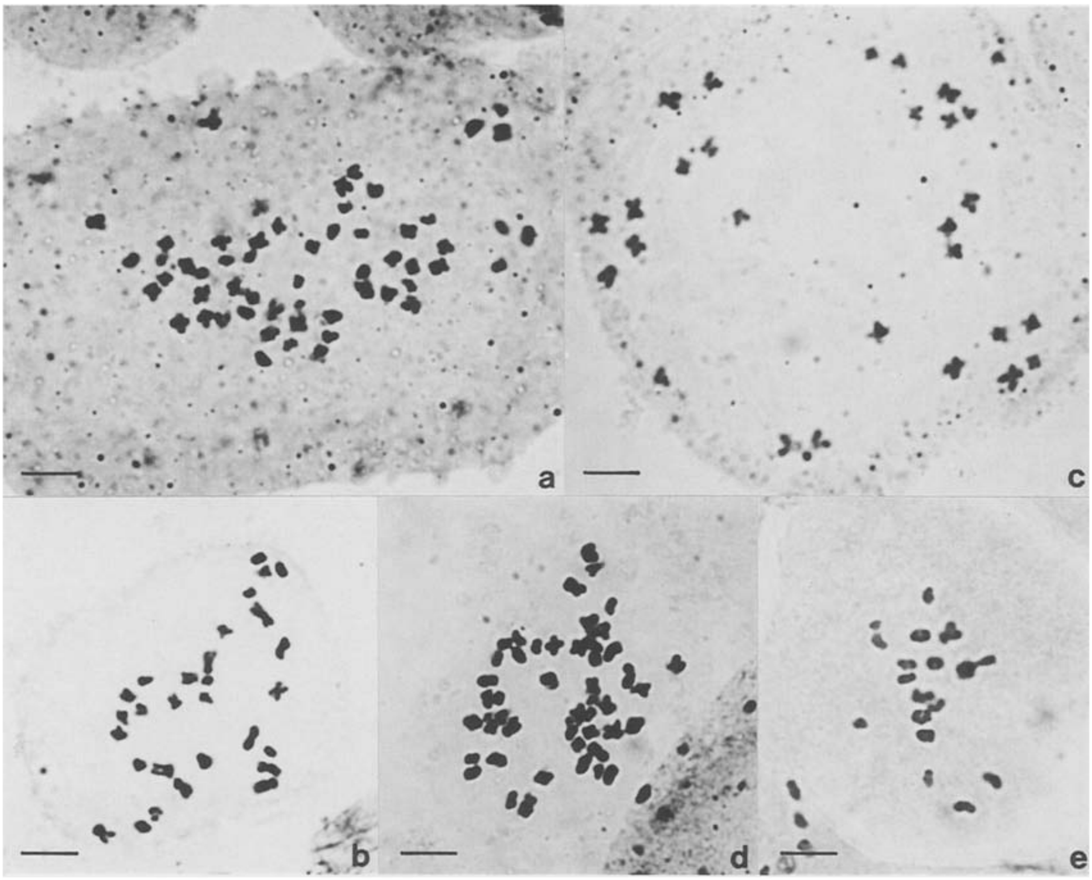


Fig. 1. Mitotic (a–d) and meiotic chromosomes (e) in F_2 and B_1 plants. a: $2n = 56$, b: $2n = 29$, c: $2n = 28$, d: $2n = 48$. e: $1_{III} + 9_{II} + 7_I$. —: indicates 5 microns.

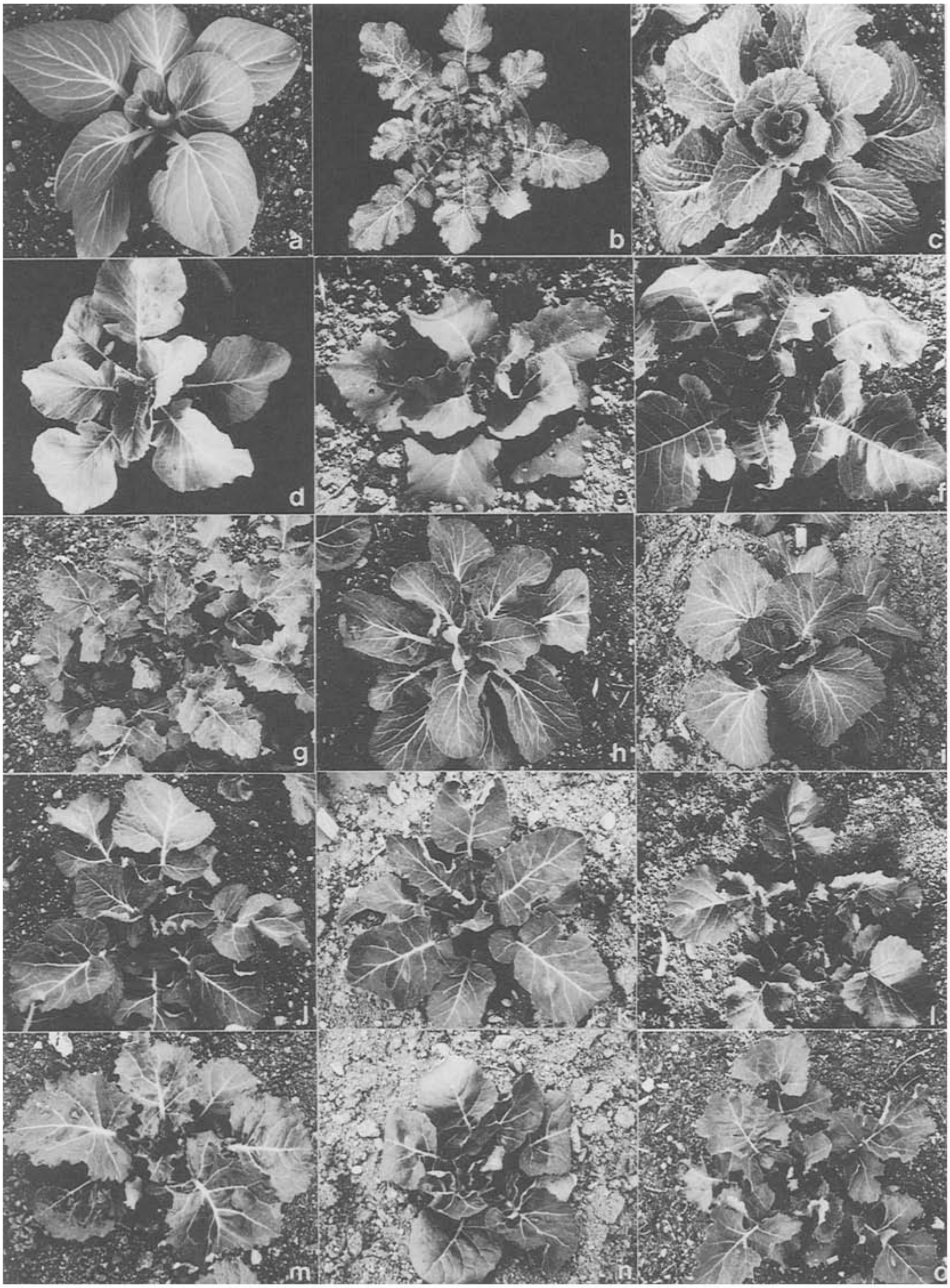
rup, personal communication). The pollination experiment was the same as experiments reported in a previous paper (Inomata, 1983). Three cultivars of *B. campestris* – ssp. *chinensis* cv. Seppaku-taina, ssp. *pekinensis* cv. Nozaki-hakusai No. 2 and ssp. *trilocularis* cv. Brown Sarson DS-2 – were used and these were the same as used in the production of the F_1 hybrid. One cultivar of *B. napus* used in the experiment was ssp. *oleifera* cv. Mihi-natane.

Investigation showed that pollen fertility and chromosome configuration for the PMCs were the same as those reported in a previous paper (Inomata, 1985a). The number of chromosomes in the root tip cells of the progenies was counted by the Feulgen squash method (Inomata, 1977).

Results

Crossability of the F_1 hybrids and their progenies

Table 1 shows the results of crossability studies on the F_1 hybrids. No seed was obtained from self-pollination. A few seeds were obtained from backcrosses of the F_1 hybrid with *B. campestris.* Many seeds were harvested after open pollination and from the crosses of F_1 hybrids × *B. napus.* Table 2 shows the results of a study of the chromosome number in some progenies of F_2, B_1 and hybrid plants. Many types of aneuploids appeared in open pollinated progeny and most of their seedlings had 56 (Fig. 1a), followed by 29 chromosomes (Fig. 1b). In the cross of F_1 × *B. napus,* the most commonly occurring type of the plants had 38 chromosomes.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Morphology of the F_2, B_1 and hybrid plants

The leaf characteristics of the F_1 hybrids were intermediate between *B. campestris* and *B. bourgeaui, B. cretica* and *B. montana* (Inomata, 1993).

Figure 2 shows the leaf characteristics of the F_2, B_1 and hybrid plants, and of the male and female plants used in the production of the hybrids. No morphological differences were observed among the same cross combinations. The leaf characteristics of the B_1 (Fig. 2i) and hybrid plants (Figs. 2j, k, l, o) were more similar to *B. campestris* and *B. napus* than to those of F_1 hybrids, respectively. The F_2 plant with 28 chromosomes (Fig. 1c), obtained from the open pollination of the F_1 hybrids, was more similar to *B. campestris* (Fig. 2h) than to those of F_1 hybrid. The plant having 34 chromosomes, derived from the open pollination of the F_1 hybrid, was more similar to *B. napus* (Fig. 2n). The B_1 plant with 48 chromosomes (Fig. 1d), derived from the F_1 hybrid backcrossed with *B. campestris,* was more similar to *B. campestris* (Fig. 2m).

Chromosome configuration for the PMCs of F_2, B_1 and hybrid plants

Tables 3 and 4 show the results of studies on chromosome configurations for the PMCs of the F_2, B_1 and hybrid plants with 29 and 38 chromosomes, respectively. In the plants with 29 chromosomes, the pollen fertility ranged from 0–38.0%, with a mean of 15.8% in F_2 plants, and from 26.9–89.0%, with a mean of 53.0% in B_1 plants. The mode of chromosome configuration for the PMCs was $12_{II} + 5_I$ followed by $13_{II} + 3_I$ and $11_{II} + 7_I$. The range of chromosome configurations for the PMCs showed $(0–1)_{IV}$ + $(0–1)_{III} + (9–14)_{II} + (1–9)_I$. In the hybrid plants with 38 chromosomes the pollen fertility ranged from 38.0–92.0%, with a mean of 74.7%. The mode of chromosome configuration for the PMCs was 19_{II} followed by $1_{III} + 17_{II} + 1_I$ and $18_{II} + 2_I$. The range of chromosome configurations for the PMCs showed $(0–1)_{IV} + (0–2)_{III} + (14–19)_{II} + (0–7)_I$. Bivalent formation was high in the hybrid plants with 29 and 38 chromosomes, respectively.

The pollen fertility was 4.4% in the plant with 25 chromosomes (No. 4 in Table 6). The mode of chromosome configuration for the PMCs was $11_{II} + 3_I$ followed by $10_{II} + 5_I$ and $9_{II} + 7_I$, and reached 83.3%. The range of chromosome configurations for the PMCs showed $(0–1)_{III} + (8–10)_{II} + (2–9)_I$. The pollen fertility was 0% in the plant with 28 chromosomes and the chromosome configurations for the PMCs were $(0–2)_{III} + (8–12)_{II} + (1–10)_I$ (Fig. 1e). The pollen fertility was 54.2% in the plant with 34 chromosomes (No. 12 in Table 6), and the mode of chromosome configuration for the PMCs was $16_{II} + 2_I$ followed by $15_{II} + 4_I$ and $14_{II} + 6_I$, and reached 61.3%. The range of chromosome configurations for the PMCs were $(0–1)_{IV} + (0–1)_{III} + (13–17)_{II} + (1–6)_I$. The pollen fertility was 90.6% in the plant with 47 chromosomes (No. 9 in Table 6) and the range of chromosome configurations for the PMCs were $(0–1)_{IV} + (0–1)_{III} + (19–23)_{II} + (1–6)_I$.

Crossability of the F_2, B_1 and hybrid plants, and chromosome number in their progenies

Table 5 shows the cross combination and the number of plants examined with regard to crossability.

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Fig. 2. Open pollination of the F_1 hybrids of *Brassica campestris* and wild relatives of *B. oleracea, B. bourgeaui, B. cretica* and *B. montana,* B_1 plants in the F_1 hybrids backcrossed with *B. campestris,* hybrid plants in the cross of the F_1 hybrid × *B. napus,* and their parents. a: *B. campestris* ssp. *chinensis* cv. Seppaku-taina ($2n$ = 20). b: *B. campestris* ssp. *trilocularis* cv. Brown Sarson DS-2 ($2n$ = 20). c: *B. campestris* ssp. *pekinensis* cv. Nozaki-hakusai No. 2 ($2n$ = 20). d: *B. bourgeaui* 120 ($2n$ = 18). e: *B. cretica* ssp. *cretica* 35 ($2n$ = 18). f: *B. montana* 89 ($2n$ = 18). g: *B. napus* ssp. *oleifera* cv. Miho-natane ($2n$ = 38). h: F_2 plants ($2n$ = 28) in open pollination of the F_1 hybrid in Seppaku-taina × *B. bourgeaui.* i: B_1 plant ($2n$ = 29) in the F_1 hybrid (Seppaku-taina × *B. bourgeaui*) backcrossed with Seppaku-taina. j: Hybrid plant ($2n$ = 38) in the cross of the F_1 hybrid (Seppaku-taina × *B. bourgeaui*) × Miho-natane. k: Hybrid plant ($2n$ = 38) in the cross of the F_1 hybrid (Nozaki-hakusai No. 2 × *B. bourgeaui*) × Miho-natane. 1: Hybrid plant ($2n$ = 38) in the cross of the F_1 hybrid (Brown Sarson DS-2 × *B. cretica*) × Miho-natane. m: B_1 plant ($2n$ = 48) in the F_1 hybrid (Nozaki-hakusai No. 2 × *B. cretica*) backcrossed with Nozaki-hakusai No. 2. n: F_2 plant ($2n$ = 34) in open pollination of the F_1 hybrid in Nozaki-hakusai No. 2 × *B. montana.* o: Hybrid plant ($2n$ = 38) in the cross of the F_1 hybrid (Nozaki-hakusai No. 2 × *B. montana*) × Miho-natane.

Table 3. Chromosome configuration at the PMCs of F_2 and B_1 plants with 29 chromosomes derived from open pollination of the F_1 hybrids of *Brassica campestris* and *B. bourgeaui*, and from B_1 plants from the F_1 hybrid backcrossed with *B. campestris*, respectively

Plant No.[1]	First meiotic division					
	No. of PMCs observed	$13_{II}+3_I$	$12_{II}+5_I$	$11_{II}+7_I$	$10_{II}+9_I$	Other types
1	31	8	11	3	3	6
1	30	4	10	0	0	16
1	30	7	11	3	7	2
2	30	0	15	5	0	10
5	30	12	8	4	4	2
6	33	5	10	7	3	8
	30	3	9	8	6	4
Total	214	39	74	30	23	48
(%)	(99.9)	(18.2)	(34.6)	(14.0)	(10.7)	(22.4)

[1] Plant number corresponds to the plant number in Table 6
Plant without plant number does not correspond to Table 6 and the B_1 plant derived from the backcross of F_1 hybrid (Seppaku-taina × *B. cretica*) with Seppaku-taina

Table 6 shows the results of studies of the crossability of F_2, B_1 and hybrid plants and the chromosome number in the progenies. Six plants with 29 chromosomes, nine plants with 38 chromosomes and six plants with other than 29 or 38 chromosomes were investigated. Of the six plants with 29 chromosomes, four were from F_2 plants from open pollination of the F_1 hybrid, and the remaining two plants were from the F_1 hybrid backcrossed with *B. campestris*. A few seeds were obtained via self-pollination, and the chromosome number ranged from 24–36. Many seeds were harvested after open pollination and from B_1 plants. The chromosome number ranged from 20–49 from open pollination and from 17–32 from B_1 plants. Most of their seedlings had 24 or 26 chromosomes and progenies were obtained with one-by-one or little-by-little chromosome addition.

Of the nine plants with 38 chromosomes, three were from the hybrid plant resulting from the cross of the F_1 hybrid (Seppaku-taina × *B. bourgeaui*) × *B. napus*. Five plants were from the hybrid plants resulting from the cross of the F_1 hybrid (Brown Sarson × *B. cretica*) × *B. napus*. The remaining plant

Table 4. Chromosome configuration at the PMCs of hybrid plants with 38 chromosomes derived from the F_1 hybrids (*Brassica campestris* × three wild relatives of the *B. oleracea, B. bourgeaui, B. cretica* and *B. montana*) crossed with *B. napus*

Plant No.[1]	First meiotic division					
	No. of PMCs observed	$1_{III}+17_{II}+1_I$	$17_{II}+4_I$	$18_{II}+2_I$	19_{II}	Other types
3	31	1	9	14	3	4
7	30	14	0	3	13	0
7	31	4	0	5	22	0
8	31	0	0	1	30	0
13	34	3	0	13	15	3
Total	157	22	9	22	83	21
(%)	(100)	(14.0)	(5.7)	(14.0)	(52.9)	(13.4)

[1] Plant number corresponds to the plant number in Table 6

was from the hybrid plant resulting from the cross of the F_1 hybrid (Nozaki-hakusai No. 2 × *B. montana*) × *B. napus*. In each case many seeds were harvested from self-, open pollination and hybrid plants. The chromosome number ranged from 28–44 from self-pollination, 27–44 from open pollination, 29–45 in hybrid plants and 33–52 from sib-cross. Most of their seedlings had 38 chromosomes in all cross combinations. Many aneuploids were also observed with one-by-one or little-by-little chromosome addition.

The F_2 plant with 25 chromosomes was from open pollination of the F_1 hybrid in *B. campestris* × *B. bourgeaui*. Many seeds were harvested from both open pollination and backcross with *B. campestris*. Most of their seedlings had 24 chromosomes in both crosses. The hybrid plant with 47 chromosomes was from the cross of F_1 hybrid × *B. napus*, and most of their seedlings had 48 followed by 52 chromosomes from self-pollination and 41 followed by 38 chromosomes from open pollination. Two B_1 plants with 48 chromosomes were from the backcross of the F_1 hybrid with *B. campestris* and most of their seedlings had 38 followed by 42 chromosomes from open pollination. The hybrid plant with 42 chromosomes was from the cross of F_1 hybrid × *B. napus*, and most of their seedlings had 43 followed by 45 chromosomes after both open pollination and cross with *B. napus*. The F_2 plant with 34 chromosomes was from open pollination of the F_1 hybrid in *B. campestris* × *B. montana* and most of their seedlings had 38 chromosomes after both open pollination and crossed with *B. napus*. The progenies of the plants with 25, 47, 48 and 34 chromosomes showed one-by-one or little-by-little chromosome addition from 20 to 65 chromosomes.

Discussion

Although most pollen grains were sterile in the F_1 hybrids of *B. campestris* and *B. oleracea*, many seeds were harvested from B_1 plants of the F_1 hybrids backcrossed with *B. campestris* and from hybrid plants of the F_1 hybrids crossed with *B. napus*. Most of the B_1 and hybrid plants obtained had 29 and 38 chromosomes, respectively. The most frequent type of normal egg cell in the F_1 hybrids was unreduced gamete (Inomata, 1983). Almost all pollen grains were sterile in the F_1 hybrids of *B. campestris* and *B. bourgeaui*, *B. cretica* and *B. montana* obtained by ovary culture (Inomata, 1985c, 1986, 1987). In the present experiment, five B_1 plants from the F_1 hybrids backcrossed with *B. campestris* were examined, and three of them had 29 chromosomes, and most hybrid plants in the F_1 hybrids crossed with *B. napus* showed 38 chromosomes. The most frequent type of normal egg cell in the F_1 hybrids might be unreduced gamete.

The mean frequency of chromosome configuration at the PMCs of the F_1 hybrids of *B. campestris* and *B. oleracea* showed $9_{II} + 1_I$, $1_{III} + 8_{II}$ and $8_{II} + 3_I$ was 44.7%, 16.9% and 11.4%, respectively (Inomata, 1980). Mean bivalent formation at the PMCs was 7.3 in the F_1 hybrids of *B. campestris* and *B. oleracea* (Attia & Röbbelen, 1986). In the previous papers on the F_1 hybrids of *B. campestris* and *B. bourgeaui*, *B. cretica* and *B. montana*, the mean frequency of chromosome configuration at the PMCs showed $9_{II} + 1_I$, $1_{III} + 8_{II}$ and $8_{II} + 3_I$ was 29.1%, 23.0% and 7.9%, respectively (Inomata, 1985c, 1986, 1987). It seemed that there was high homology in the chromosomes of the hybrids of *B. campestris* and wild relatives of the *B. oleracea*. The recombination might occur reciprocally.

Table 5. Cross combination and number of plants examined in crossability of the F_2, B_1 and hybrid plants

Cross combination of the F_1 hybrid[1]	Open pollination	F_1 × *B. campestris*[1]	F_1 × *B. napus*[1]
ssp. *chinensis* × *B. bourgeaui*	3	1	1
ssp. *pekinensis* × *B. bourgeaui*	2	1	2
ssp. *trilocularis* × *B. cretica*	0	0	6
ssp. *pekinensis* × *B. cretica*	0	2	1
ssp. *pekinensis* × *B. montana*	1	0	1

[1] Refer to Table 1

Table 6. Chromosome number and crossability of the F_2, B_1 and hybrid plants derived from the F_1 hybrids of *Brassica campestris* and three wild relatives of *B. oleracea, B. bourgeaui, B. cretica* and *B. montana*, and the chromosome number in some of their progenies

Block number of F_2, B_1 and hybrid plant[1]	No. of plants observed	Chromosome number ($2n$)	Type of pollination of F_2, B_1 and hybrid plant	Progeny				
				No. of flowers pollinated	No. of seeds obtained	No. of seeds sown	No. of seeds germinated	Chromosome number in root tip cell ($2n$)[2]
1	3	29	Self-pollination	250	27	10	9	29(2), 32(3), 35(1), 36(1), ?(2)
			Open pollination	173	251	80	75	20(3), 21(1), 22(14), 23(10), 24(18), 25(7), 26(1), 27(1), 28(2), 29(2), 30(1), 31(1), 32(4), 34(2), 42(1), 48(1), ?(6)
			Sib-cross	34	2	2	2	27(1), ?(1)
			B. campestris	48	166	50	44	17(1), 20(4), 21(2), 22(10), 23(6), 24(10), 25(3), 28(1), 29(2), ?(5)
2	1	29	Self-pollination	37	0	–	–	
			Open pollination	47	98	50	49	20(2), 21(3), 22(6), 23(1), 24(14), 25(6), 26(8), 27(3), 29(1), 32(2), 36(1), 38(1), 49(1)
			B. campestris	21	32	20	18	20(2), 21(1), 23(1), 24(6), 26(6), 28(1), 29(1)
3	1	38	Self-pollination	68	410	70	66	29(6), 30(2), 31(1), 32(3), 34(3), 37(1), 38(30), 39(1), 40(1), 41(2), 42(12), 44(2), ?(2)
			Open pollination	61	625	100	100	29(8), 38(58), 39(1), 40(7), 41(3), 42(13), 43(1), 44(1), ?(8)
			B. napus	21	340	50	49	29(2), 36(1), 38(38), 41(1), 42(3), 43(1), 45(1), ?(2)
4	1	25	Self-pollination	37	0	–	–	
			Open pollination	43	75	50	39	20(5), 23(2), 24(8), 25(2), 26(3), 29(3), 32(5), 33(1), 34(1), 44(1), ?(8)
			B. campestris	18	72	50	47	20(5), 21(1), 22(9), 23(5), 24(10), 25(1), 26(3), ?(13)
5	1	29	Self-pollination	44	4	4	2	?(2)
			Open pollination	79	37	37	22	20(1), 21(2), 23(1), 24(2), 26(4), 29(1), 30(1), 31(1), 32(1), 34(1), ?(7)
			B. campestris	22	20	20	12	20(1), 21(1), 22(2), 23(1), 24(2), 25(1), 27(2), 29(1), 32(1)
6	1	29	Self-pollination	21	4	4	4	24(1), 26(2), 36(1)
			Open pollination	96	315	50	48	21(3), 22(6), 23(3), 24(12), 25(11), 26(9), 28(1), 31(1), ?(2)
			B. campestris	17	54	25	25	21(1), 22(2), 23(1), 23 & 24(1), 24(5), 25(1), 26(8), 28(2), 29(1), ?(3)
7	2	38	Self-pollination	82	248	100	95	29(2), 34(11), 35(2), 36(7), 38(58), ?(15)
			Open pollination	107	688	100	96	26(1), 28(2), 29(1), 32(7), 34(10), 35(10), 36(9), 37(7), 38(42), ?(7)
			Sib-cross	43	296	100	95	33(1), 34(4), 35(16), 36(4), 38(59), 52(1), ?(10)
			B. napus	44	338	100	88	29(1), 32(2), 33(1), 34(1), 35(11), 36(9), 37(8), 38(50), ?(5)
8	5	38	Self-pollination	141	247	55	45	29(1), 34(2), 37(2), 38(38), ?(2)
			Open-pollination	144	1057	205	189	28(1), 29(76), 32(3), 33(1), 34(1), 35(8), 37(2), 37 & 39(1), 38(84), 41(1), ?(11)
			B. napus	152	2467	205	184	29(1), 36(3), 37(5), 38(159), ?(16)
9	1	47	Self-pollination	43	38	38	26	44(1), 46(1), 48(9), 49(2), 50(1), 52(6), 55(2), 56(1), 57(1), 58(1), ?(1)
			Open-pollination	30	104	50	49	38(7), 39(2), 40(1), 41(8), 43(6), 44(2), 45(4), 47(2), 48(2), 51(3), 55(1), 57(1), ?(10)
10	2	48	Self-pollination	82	4	4	1	42(1)
			Open-pollination	96	124	55	52	36(1), 38(27), 38 & 41(1), 40(2), 41(3), 42(7), 43(1), 44(1), 45(3), 56(1), 58(1), 65(1), ?(3)
			B. campestris	87	12	12	2	29(2)

Table 6. Continued

Block number of F_2, B_1 and hybrid plant[1]	No. of plants observed	Chromosome number (2*n*)	Type of pollination of F_2, B_1 and hybrid plant	Progeny				
				No. of flowers pollinated	No. of seeds obtained	No. of seeds sown	No. of seeds germinated	Chromosome number in root tip cell (2*n*)[2]
11	1	42	Self-pollination	19	0	–	–	
			Open pollination	56	303	205	180	26(1), 35(1), 37(3), 38(9), 39(14), 40(1), 41(27), 42(11), 43(38), 44(3), 45(20), 46(6), 47(7), 48(3), 51(7), 52(1), 53(3), 54(2), 55(2), 56(2), 57(1), 59(1), ?(17)
			B. napus	30	232	105	97	33(1), 37(4), 38(5), 39(7), 40(1), 41(10), 42(9), 43(19), 44(6), 45(15), 46(5), 47(8), 49(3), ?(4)
12	1	34	Self-pollination	55	1	1	0	
			Open pollination	135	136	61	54	26(2), 29(1), 30(1), 31(1), 32(4), 34(3), 35(4), 36(8), 38(14), 40(3), 42(1), ?(12)
			B. napus	22	35	35	32	26 & 38(1), 33(1), 34(2), 37(1), 38(18), 39(2), 40(3), 41(1), 42(2), 43(1)
13	1	38	Self-pollination	63	399	100	92	28(1), 29(3), 32(1), 34(1), 38(63), ?(23)
			Open pollination	105	1497	100	100	27(4), 29(23), 32(5), 33(1), 34(1), 35(3), 36(4), 37(5), 38(20), ?(34)
			B. napus	24	558	100	100	29(2), 32(1), 35(3), 35 & 37(1), 37(14), 38(60), ?(19)

[1] 1: Three F_2 plants in open pollination of the F_1 hybrid of *B. campestris* ssp. *chinensis* cv. Seppaku-taina and *B. bourgeaui* 120
2: One B_1 plant in the backcross of the F_1 hybrid (Seppaku-taina × *B. bourgeaui*) with cv. Seppaku-taina
3: One hybrid plant in the cross of the F_1 hybrid (Seppaku-taina × *B. bourgeaui* 120) and Miho-natane
4 and 5: Two F_2 plants in open pollination of the F_1 hybrid of *B. campestris* ssp. *pekinensis* cv. Nozaki-hakusai No. 2 and *B. bourgeaui* 120
6: One B_1 plant in the backcross of the F_1 hybrid (Nozaki-hakusai No. 2 × *B. bourgeaui* 120) with Nozaki-hakusai No. 2
7: Two hybrid plants in the cross between F_1 hybrids (Nozaki-hakusai No. 2 × *B. bourgeaui* 120) and Miho-natane
8 and 9: Six hybrid plants in the cross of the F_1 hybrid (*B. campestris* ssp. *trilocularis* cv. Brown Sarson DS-2 × ssp. *cretica* 35) and Miho-natane
10: Two B_1 plants in the backcross of the F_1 hybrid (Nozaki-hakusai No. 2 × *cretica* 35) with Nozaki-hakusai No. 2
11: One hybrid plant in the cross of the F_1 hybrid (Nozaki-hakusai No. 2 × ssp. *cretica* 35) with Miho-natane
12: One F_2 plant in open pollination of the F_1 hybrids of Nozaki-hakusai No. 2 and *B. montana* 89
13: One hybrid plant in the cross of the F_1 hybrid (Nozaki-hakusai No. 2 × *B. montana* 89) with Miho-natane

[2] Number in parentheses shows the number of plants observed, and ? shows a chromosome number that could not be determined

In the B_1 plants with 29 chromosomes obtained from the F_1 hybrids (*B. campestris* × *B. oleracea*) backcrossed with *B. campestris,* the frequency of chromosome configurations for the PMCs showing $10_{II} + 9_I$, $11_{II} + 7_I$ and $12_{II} + 5_I$ was 44.3%, 40.8% and 11.8%, respectively (Inomata, 1985a). In the present experiment, chromosome configurations at the PMCs of the F_2 and B_1 plants with 29 chromosomes were examined and the frequency showing $12_{II} + 5_I$, $13_{II} + 3_I$, $11_{II} + 7_I$ and $10_{II} + 9_I$ was 34.6%, 18.2%, 14.0% and 10.7%, respectively. If genome constitution of the F_2 and B_1 plants with 29 chromosomes was AAC, chromosome configuration at the PMCs might be only $10_{II} + 9_I$, but the frequency was only 10.7% and tri- and tetravalent formation was observed. Meanwhile, the mean frequency showing 19_{II} reached 71.5% in the hybrid plants with 38 chromosomes in the cross of the F_1 hybrid (*B. campestris* × *B. oleracea*) and *B. napus* (Inomata, 1985a). In the present experiment the mean frequency showing 19_{II} reached 52.9% in the hybrid plants with 38 chromosomes. Pollen fertility varied in the plants with the same number of chromosomes. It seemed that the complex chromosome configuration occurred among multivalents in *B. campestris* and wild relatives of the *B. oleracea.*

In a previous paper on the progenies of the F_2 and B_1 hybrids of *B. campestris* and *B. oleracea*, most of the seedlings of the F_3 and B_2 plants backcrossed with *B. campestris* had 22 and 24 chromosomes, but plants of the *B. campestris* type with 20 chromosomes, *B. oleracea* type with 18 chromosomes and *B. napus* type with 38 chromosomes, appeared. It was possible that the genes of the *B. campestris* and *B. oleracea* could easily be introduced into *B. napus*, and that the genes of *B. campestris* and *B. oleracea* could exchange reciprocally (Inomata, 1991). In the progenies of the interspecific F_1 hybrids of *B. napus* and *B. campestris,* plants with 22, 24 and 26 chromosomes appeared in the F_1 hybrids backcrossed with *B. campestris* (Nwankiti, 1971; Lee & Namai, 1992). The present experiment showed the same tendency in the progenies of the F_1 hybrids of *B. campestris* and *B. bourgeaui, B. cretica* and *B. montana.* The genes of *B. bourgeaui, B. cretica* and *B. montana* could be introduced into *B. campestris* and *B. napus.*

From reports of germplasm conservation of wild relatives of the *B. oleracea* we know that many species and subspecies are distributed in Cyprus, the Mediterranean and the West Coast of Europe (Snogerup, 1980; Gustafsson, 1982; Gustafsson et al., 1983). A new subspecies of *B. cretica* was collected in Peloponnisos (Gustafsson & Snogerup, 1983). The taxonomy and variation of the wild relatives of *B. oleracea* was reported recently (Snogerup et al., 1990). F_1 hybrids of *B. campestris* and other wild relatives of *B. oleraceae* may be produced by ovary culture, and B_1 plants backcrossed with *B. campestris* and hybrid plants crossed with *B. napus* may be obtained. The wild relatives of the *B. oleracea* group are an important source of germplasm for the improvement of *Brassica* crops.

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