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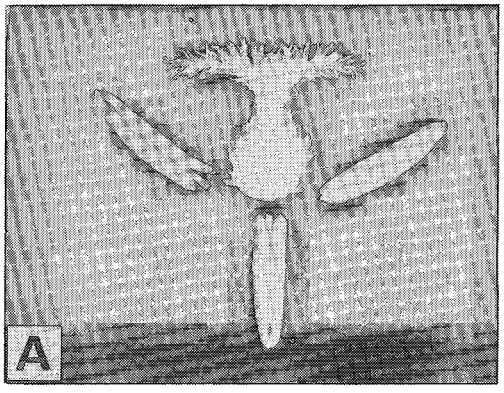

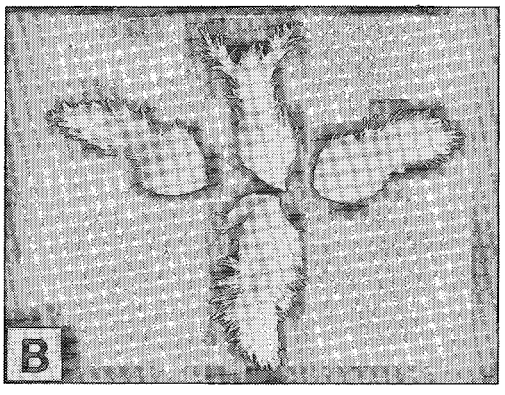

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WHEAT INFORMATION SERVICE

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Wheat Information Service
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IN MEMORY OF Dr. E. R. Sears (15 October 1910 – 15 February 1991)

(1983 in Okayama)

I sincerely wish to express my heartfelt condolences over the passing of Dr. E. R. Sears.

I remember clearly when Dr. Sears came to visit Japan on the occasion of the International Genetics Symposium held in Tokyo and Kyoto in 1956. I went to Haneda (Tokyo) Airport to meet Dr. Sears. He was already well-known by wheat geneticists in Japan. This was caused primarily by the fact that Dr. Sears in the U. S. A. and Dr. Kihara in Japan at about the same time independently discovered during the Second World War that the D genome of hexaploid wheat had been derived from *Aegilops squarrosa.*

During the years when I was a Ph. D. student with Dr. Sears, majoring in genetics at the University of Missouri, I had many good opportunities to discuss with him various problems of wheat cytogenetics almost every day of the week; these discussions sometimes lasted until midnight by continuing them in Dr. Sears' home. "McFadden and I" —— Dr. Sears started to say about the origin of hexaploid wheat many times. I found his deep appreciation of cytogenetics, not just on wheat cytogenetics, very impressive. He had a rare warmth of personality. He was the kindest and most generous of men. Also, he was a

scholar of great eminence, indeed. During our discussions, he talked always with a good sense of humor and with much appreciation for the achievement of his Japanese colleagues.

His wide knowledge and understanding of wheat are revealed in his work. Certainly the most important to all of us over the world, working with wheat, is the wheat aneuploid series developed by him in the cultivar Chinese Spring of common wheat. He has contributed also a great deal to the general understanding of the cytogenetics of wheat and relatives. These include the proof that the loci are duplicated on the homoeologous chromosomes which behave cytologically as though they would be non-homologous but genetically very similar by basic structure and function. There are genes with a drastic effect in duplicated loci, such as the *Q* gene for the origin of the squareheaded, non-spelting naked wheat. It was a corollary of the studies that the *Ph* gene was discovered. There were many other unique contributions of Dr. Sears, such as the nulli-tetra compensation combination, the chromosome substitution lines, telo- and isochromosomes, introduction of chromosome segments from foreign genomes, etc. He developed most of stocks with a pair of forceps and made great contributions to the many fields of modern genetics and agricultural sciences. There is no doubt that these are the fruits of Dr. Sears' assiduous labor. For these unique devotion I pay my sincere respect to Dr. Sears.

Once I had a chance to ask Dr. Sears for a few words as a motto. He suggested to me "patience, to be patient, work hard and study hard". He meant these as requisites for a cytogeneticist.

Dr. Sears, you have created for us a great jewel to use in plant cytogenetics. Although you are gone and we must say good-bye, "Sayonara" in Japanese, your good and precious contributions and most valuable teachings, given to us will never be forgotten.

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Production of D addition lines in tetraploid wheat: I. Cytology of F_1 and F_2 hybrids involving tetraploid and hexaploid wheats

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Introduction

Spontaneous transfer of traits from some alien species to wheat under natural conditions has been established and some cultivated varieties of wheat, particularly those grown in Europe and USSR, are known to have whole chromosomes or segments of chromosomes from one or the other alien source (Gupta 1979). While on one hand the attempts to artificially incorporate such transfers have met with success in many cases, on the other they have generated valuable information regarding homoeologous relationships between wheat chromosomes and chromosomes from several alien species.

In hexaploid bread wheat, addition lines that carry a pair of alien chromosomes have been obtained using several related genera like *Aegilops, Haynaldia, Secale* and *Hordeum*. However, despite continued cultivation and utility, tetraploid wheats have received little attention for such chromosome manipulations. Joppa (1973) reported production of D genome substitution lines in durum wheats by crossing nullisomic-tetrasomic plants of hexaploid wheat with durum wheat. Yashvir and Kesavan (1979) obtained three monosomic addition and one double monosomic addition plants by selfing pentaploid hybrids produced by crossing hexaploid and tetraploid wheats. Joppa and McNeal (1972) and Joppa et al (1979) developed disomic addition lines for chromosomes 1D, 3D, 4D, 5D and 6D in durum wheat.

Since only limited work has been done towards production of addition or substitution lines in durums, there is a need to attempt addition or substitution of many more alien chromosomes to this crop. Such work may also generate valuable basic material for exploitation in wheat breeding programmes.

Materials and Methods

Reciprocal crosses were made between tetraploid wheat strains DD-21, Jairaj and ED-1570 and hexaploid wheat strains Sonora-64 and C-306. Nine F_1 plants reached maturity. They were selfed to obtain F_2 progeny. Emerging ears were bagged to avoid pollen contamination. Standard aceto-carmine squash technique was used for cytological studies.

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Results and Discussion

The data on chromosome associations at metaphase I of meiosis of nine F_1 plants are presented in Table 1. All the plants had expected number of chromosomes ($2n=35$). Mean number of bivalents ranged between 13.88 to 14.00. As expected, the mode of the number of bivalents was 14. Mean number of ring bivalents per cell ranged between 10.44 to 12.20, with 11 being the most common number. For rod bivalents, mean/cell ranged from 1.80 to 3.52, with two modes (2 and 3). As expected, number of univalents in all the cases was 7 (Fig. 1A), with the mean ranging between 7.00 and 7.24. The presence of a quadrivalent in AD2 (5) in a very low frequency could be due to homoeologous pairing or interchange heterozygosity.

In pentaploid F_1 hybrids ($2n=35$, $14''+7'$), the gametic chromosome number is expected to range between 14 and 21. Accordingly, the somatic chromosome number in F_2 plants was found to range between 28 and 42. However, since the aim of the present experiment was to obtain disomic additions of D genome chromosomes to tetraploid wheat, the plants with more than fifteen bivalents were of no consequence.

Table 1. Chromosome associations at meiotic metaphase I of F_1 hybrids involving 4*x* and 6*x* wheats

Plant	2*n*	Chromosome associations*				
		I	II			IV
			Total	Ring	Rod	
D (1)	35	7-9	13-14	10-13	0-4	
		(7, 7.08)	(14, 13.96)	(12, 12.06)	(2, 1.90)	
AD1 (2)	35	7-7	14-14	11-14	0-3	
		(7, 7.00)	(14, 14.00)	(12, 12.20)	(2, 1.80)	
AD1 (8)	35	7-7	14-14	9-14	0-5	
		(7, 7.00)	(14, 14.00)	(11, 11.64)	(3, 2.36)	
AD2 (3)	35	7-9	13-14	6-13	1-8	
		(7, 7.08)	(14, 13.96)	(11, 10.80)	(3, 3.16)	
AD2 (4)	35	7-7	14-14	9-14	0-5	
		(7, 7.00)	(14, 14.00)	(11, 11.56)	(3, 2.44)	
AD2 (5)	35	7-9	13-14	6-14	0-8	0-1
		(7, 7.04)	(14, 13.92)	(12, 10.72)	(2, 3.20)	(0, 0.04)
AD2 (9)	35	7-7	14-14	9-14	0-5	
		(7, 7.00)	(14, 14.00)	(12, 11.72)	(2, 2.28)	
AD3 (1)	35	7-9	13-14	6-13	0-8	
		(7, 7.08)	(14, 13.96)	(11, 10.44)	(3, 3.52)	
AD3 (2)	35	7-11	12-14	9-14	0-5	
		(7, 7.24)	(14, 13.88)	(11, 11.40)	(2, 2.40)	

* In each column, the figures in first row are range and those in second row in parentheses are mode and mean respectively.

Table 2. Chromosome associations at meiotic metaphase I of F_2 hybrids involving 4*x* and 6*x* wheats

Plant	2*n*	Chromosome associations* I	II	III	IV
D (1) 1	31	3-5 (3, 3.00)	12-15 (14, 13.84)	–	0-1 (0, 0.08)
D (1) 4	34	4-4 (4, 4.00)	15-15 (15, 15.00)	–	–
AD1 (1) 1	29	0-2 (1, 0.68)	12-14 (14, 13.44)	0-1 (0, 0.48)	–
AD1 (1) 4	29	1-3 (1, 1.08)	12-14 (14, 13.88)	–	0-1 (0, 0.04)
AD1 (2) 2	37	3-8 (5, 5.32)	13-17 (15, 15.24)	0-1 (0, 0.40)	–
AD1 (2) 6	30	0-2 (2, 1.88)	13-15 (14, 14.00)	0-1 (0, 0.04)	–
AD1 (3) 4	30	0-4 (2, 1.84)	13-15 (14, 14.08)	–	–
AD1 (3) 7	34	2-8 (4, 4.16)	8-16 (15, 14.32)	0-2 (0, 0.24)	0-1 (0, 0.12)
AD1 (4) 3	30	0-4 (2, 2.44)	13-15 (14, 13.72)	0-1 (0, 0.04)	–
AD1 (4) 6	36	4-8 (6, 6.12)	14-16 (15, 14.88)	0-1 (0, 0.04)	–
AD1 (4) 7	35	4-7 (5, 5.56)	13-15 (15, 14.60)	0-1 (0, 0.08)	–
AD1 (8) 1	31	1-3 (3, 2.80)	13-15 (14, 13.92)	0-1 (0, 0.12)	–
AD2 (1) 1b	30	1-4 (2, 2.04)	12-14 (14, 13.80)	0-1 (0, 0.12)	–
AD2 (1) 2	31	3-5 (3, 3.08)	13-14 (14, 13.96)	–	–
AD2 (2) 1	34	4-6 (6, 5.56)	12-15 (14, 14.04)	0-2 (0, 0.12)	–
AD2 (2) 3	32	2-8 (4, 4.68)	11-15 (14, 13.44)	0-1 (0, 0.04)	0-1 (0, 0.08)
AD2 (2) 3b	33	3-7 (5, 5.08)	13-15 (14, 13.96)	–	–
AD2 (2) 6	28	0-2 (0, 0.08)	13-14 (14, 13.96)	–	–
AD2 (3) 4	36	6-8 (6, 6.16)	14-15 (15, 14.92)	–	–
AD2 (4) 4	35	5-9 (7, 7.00)	12-15 (14, 13.88)	0-1 (0, 0.08)	–

(Table 2. continued)

AD2 (4) 13	35	3-7 (5, 4.68)	14-16 (15, 15.16)	–	–
AD2 (4) 15	32	2-8 (4, 4.24)	12-15 (14, 13.76)	0-1 (0, 0.08)	–
AD2 (5) 6	31	2-7 (2, 3.24)	12-14 (14, 13.76)	0-1 (0, 0.08)	–
AD2 (5) 8	30	1-2 (2, 1.88)	13-14 (14, 13.88)	0-1 (0, 0.12)	–
AD2 (6) 7	37	4-9 (7, 6.88)	13-16 (15, 14.88)	0-1 (0, 0.12)	–
AD2 (6) 8	29	0-1 (1, 0.92)	13-14 (14, 13.92)	0-1 (0, 0.08)	–
AD2 (7) 4	30	2-4 (2, 2.32)	13-14 (14, 13.84)	–	–
AD2 (8) 1	34	4-10 (6, 6.04)	12-16 (14, 13.88)	0-1 (0, 0.12)	–
AD2 (8) 3	36	6-10 (6, 6.40)	13-15 (15, 14.88)	–	–
AD2 (9) 1	31	3-5 (3, 3.12)	12-14 (14, 13.88)	0-1 (0, 0.04)	–
AD2 (10) 3	33	3-5 (5, 4.60)	14-15 (14, 14.20)	–	–
AD2 (10) 6	34	6-10 (6, 6.48)	12-14 (14, 13.76)	–	–
AD2 (11) 8	36	4-8 (6, 6.16)	13-16 (15, 14.80)	0-1 (0, 0.08)	–
AD2 (11) 9	31	3-5 (3, 3.08)	13-14 (14, 13.96)	–	–
AD2 (11) 11	30	0-4 (0, 1.04)	13-15 (15, 14.24)	0-1 (0, 0.16)	–
AD2 (11) 12	30	0-4 (2, 1.84)	13-15 (14, 13.84)	0-1 (0, 0.16)	–
AD2 (11) 15	28	–	14-14 (14, 14.00)	–	–
AD2 (12) 3	31	3-3 (3, 3.00)	14-14 (14, 14.00)	–	–
AD2 (15) 1	33	3-7 (5, 4.80)	13-15 (14, 14.04)	0-1 (0, 0.04)	–
AD1 (8) 3	33	4-7 (5, 5.28)	10-14 (14, 13.68)	0-2 (0, 0.12)	–

* same as in Table 1

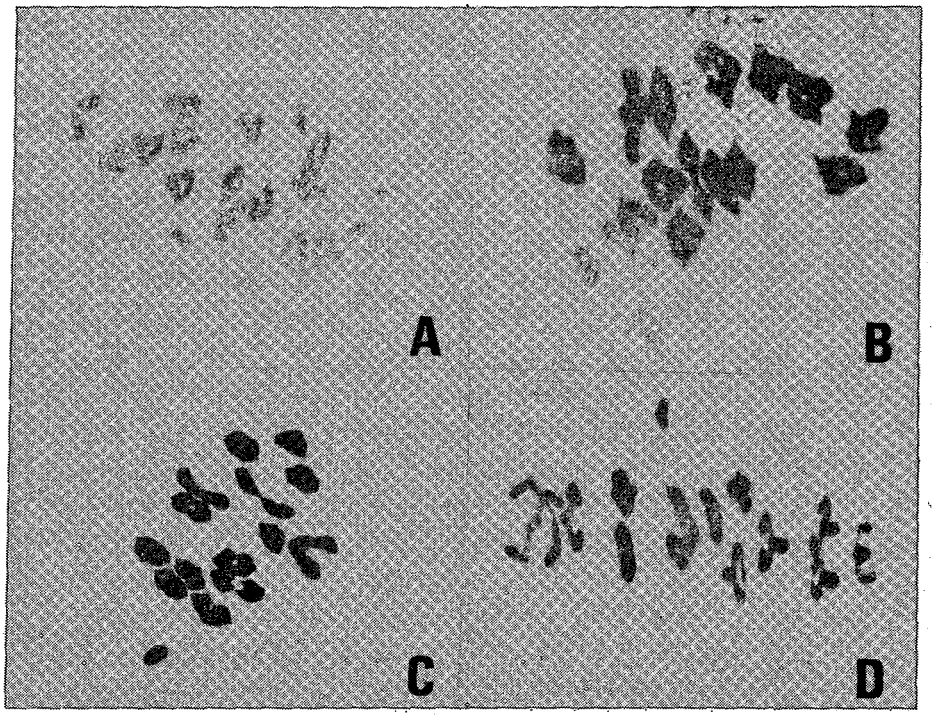

Fig. 1. Meiotic metaphase I cells of the F_1 and F_2 of 6*x* and 4*x* wheats. A: AD2(5) showing 7 univalents, 13 ring bivalents and 1 rod bivalent ($2n=35$). B: D(1)1 showing 3 univalents, 13 ring bivalents and 1 rod bivalent ($2n=31$). C: AD2(1)1b showing 1 univalent, 11 ring bivalents, 2 rod bivalents and 1 chain trivalent ($2n=30$). D: AD1(1)4 showing 1 univalent, 11 ring bivalents and 3 rod bivalents ($2n=29$)

Eleven plants were found to have 15 bivalents (Table 2). The number of univalents ranged between 0 to 7 per cell. Two plants, AD2 (4) 13 and AD1 (4) 7 had thirty five chromosomes and formed 15 bivalents at metaphase I in most of the cells. These plants thus had 14″ from A and B genomes and another bivalent from D genome. More interesting from this point of view was the plant D (1) 4 with only four univalents from D genome ($2n=34$, $15''+4'$). AD1 (3) 7, another plant with thirty four chromosomes and 15″, had several multivalents (III = 0.24/cell, IV = 0.12/cell). Therefore, there are lesser chances of obtaining a disomic addition plant in its progeny.

AD2 (11) 12 was found to be disomic addition plant ($2n=30$, 15″). Efforts are presently underway to stabilize and identify the added chromosomes.

Twenty nine plants had 14″. The number of chromosomes ranged between 28 and 35 and number of univalents between 0 and 7. D (1) 1 (Fig. 1B), AD1 (8) 1, AD2 (1) 2, AD2 (5) 6, AD2 (9) 1, AD2 (11) 9 and AD2 (12) 3 were triple monosomic addition plants. AD1 (2) 6, AD1 (3) 4, AD1 (4) 3, AD2 (1) 1b (Fig. 1C), AD2 (5) 8, AD2 (7) 4 and, AD2 (11) 12 had thirty chromosomes each and all of them were double monosomic addition plants. AD1 (1) 1, AD1 (1) 4 and AD2 (6) 8 were monosomic addition plants (Fig. 1D). AD1 (1) 1 had a high frequency of trivalents (0.48/cell). It is possible that the added chromosome is involved in the trivalent, resulting due to interchange heterozygosity or homoeologous pairing. The three chromosomes involved in the trivalent are expected to segregate into 2:1 fashion. The chances of obtaining a disomic addition plant in its progeny can thus be rated high since the extra chromosome will be involved in a 2:1 segregation giving gametes with $n=15$, 14 and thus reducing the chances of loss of extra chromosome by elimination as laggard at anaphase I.

Acknowledgments

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Hybrid weakness in *Triticum dicoccum* Schubl.

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Introduction

Genetical traits such as hybrid necrosis and hybrid chlorosis are controlled by two independent genetic systems. In both kinds of hybrid weakness the F_1 hybrids are lethal or semi-lethal resulting in gradual death or debility. Hybrid necrosis and hybrid chlorosis are frequently met with in inter- and intraspecific wheat crosses and are serious barriers to the transfer of genes in a planned hybridization programme. Hybrid necrosis is governed by two complementary genes, *Ne*1 and *Ne*2, located on chromosomes 5B and 2B respectively (Tsunewaki 1960) while hybrid chlorosis is controlled by two complementary genes *Ch*1 located on 2A (Hermsen & Waninge 1972) and *Ch*2 on 3D (Tsunewaki & Kihara 1961). The present investigations were carried out to identify the genes for necrosis and chlorosis in nine Indian varieties of *Triticum dicoccum*.

Materials and Methods

Nine varieties of *Triticum dicoccum* were crossed to two *T. aestivum* testers, C 306 (*Ne*1*ne*2*ch*1*Ch*2) and Sonalika (*ne*1*Ne*2*ch*1*Ch*2). The F_1 hybrids and parents were raised in the greenhouse under optimum conditions of growth. The F_1 hybrids were critically observed for the occurrence of hybrid necrosis and hybrid chlorosis and genotype of the parents with respect to the genes for necrosis and chlorosis were determined from the phenotype of the F_1 hybrids.

Results and Discussion

The results obtained in the study are presented in Table 1. With the exception of HW 178-A and HW 489, all the varieties of *T. dicoccum* when crossed to C 306 (*Ne*1*Ch*2-carrier) produced strong chlorotic F_1 hybrids, indicating that the varieties carry the *Ch*1 gene. The F_1 hybrid plants between Sonalika (*Ne*2*Ch*2-carrier) and four *T. dicoccum* varieties, namely, HW 43, HW 1018, HW 1046 and Khapli-53 Yellow expressed the symptoms of both necrosis and chlorosis simultaneously, indicating that these varieties also carry the gene *Ne*1. The variety HW 178-A produced normal and necrotic F_1 offspring when crossed to C 306 and Sonalika respectively. Results indicate that the variety is non-carrier for chlorosis gene while it carries the gene *Ne*1. Normal F_1 hybrids were obtained in the crosses of HW 489

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with both the testers indicating that this variety is a non-carrier for both necrosis and chlorosis genes (*ne*1*ne*2*ch*1). The F_1 hybrids between Sonalika, a *Ne*2 *Ch*2^m-carrier (Kochumadhavan et al 1984), and three *T. dicoccum* varieties, viz., HW 1016, HW 1017 and Sangli 2-2, exhibited only chlorosis and it is likely that chlorosis may obscure the expression of necrosis. Consequently, this type of epistatic gene action caused difficulty in identifying the genotype of these varieties of *T. dicoccum* as to have *Ne*1 or *ne*1. When both the complementary gene systems for hybrid necrosis and hybrid chlorosis operate in F_1 hybrids, the phenotypic expression of one hybrid weakness over the other or the simultaneous occurrence of both kinds of hybrid weakness depends on the relative strength of alleles existing at *Ne*- and *Ch*-loci. Therefore, the genes with respect to necrosis in the varieties, HW 1016, HW 1017 and Sangli 2-2, could not be determined for want of a single-gene tester (*ne*2*Ne*2*ch*1*ch*2) for necrosis or a two-gene tester (*ne*1*Ne*2*Ch*1*ch*2) for necrosis and chlorosis with the authors. While the single-gene tester is extremely rare, the two-gene tester has not yet been established in polyploid wheats.

Varieties of *T. dicoccum*, like other tetraploid species of wheat, are either *Ne*1-carriers or non-carriers (Nishikawa 1967; Tsunewaki 1969; Tomar et al unpublished). *Ne*2-carriers have not yet been reported in 4x wheats despite the location of this gene in B genome. Therefore, the *Ne*2 gene, found restricted to the western 6x wheats is presumed to have originated by mutation at the hexaploid level in Europe (Tsunewaki and Kihara 1962).

Table 1. Genotype of *T. dicoccum* varieties with respect to the genes for necrosis and chlorosis

Variety	Tester		Genotype of the variety tested
	C 306 (*Ne*1*ne*2*ch*1*Ch*2)	Sonalika (*ne*1*Ne*2*ch*1*Ch*2)	
HW43	c	nc	*Ne*1*ne*2*Ch*1
HW178-A	–	n	*Ne*1*ne*2*ch*1
HW 1016	c	c	? *ne*2*Ch*1
HW 1017	c	c	? *ne*2*Ch*1
HW 1018	c	nc	*Ne*1*ne*2*Ch*1
HW 1046	c	nc	*Ne*1*ne*2*Ch*1
Khapli-53 Yellow	c	nc	*Ne*1*ne*2*Ch*1
Sangli 2-2	c	c	? *ne*2*Ch*1
HW 489	–	–	*ne*1*ne*2*ch*1

– = normal; n = necrotic; c = chlorotic; ? = gene could not be determined.

Hermsen (1966) reported that the Indian emmer wheat Khapli carried the *Ch*1 gene. The present study reveals a high prevalence of *Ch*1-carriers in *T. dicoccum* varieties and this further corroborates the observation of Kochumadhavan et al (1984) that the gene *Ch*1 widely occur amongst the Indian varieties of *T. dicoccum*.

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Use of weak-necrotic mutant variety PNC 306 to preclude hybrid lethality in wheat crosses

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Hybrid lethality reported in F_1's is one of the major barriers (Pandey and Rao 1979) encountered by many wheat breeders for combining desirable genes from different varieties into hybrid combinations. Some of the promising indigenous varieties like C 286, C 306, NP 839 have been found to produce strong lethality in crosses with the Mexican *Ne2*^{s} carriers, Sonalika, Chotti Lerma, Safed Lerma and Kalyansona (Narula et al 1970; Gill et al 1969, 1972; Anand et al 1969; Kochumadhavan et al 1980). As a consequence of hybrid lethality many F_1's die at prematurity, imposing a serious limitation on the choice of desirable recombinants. The present paper reports the results to elicit informations for the use of weak-necrotic mutant variety, PNC 306, for obtaining desired combinations with *Ne2*^{s} carriers.

Three varieties, S 308, Kalyansona and HP 1303, all *Ne2*^{s} carriers, were crossed with a mutated variety PNC 306. The parents, S 308, Kalyansona, HP 1303 and PNC 306 as well as the F_1 plants by the crosses of S 308, Kalyansona and HP 1303 with PNC 306 were space-planted in the field along with the F_1's seeds of S 308, Kalyansona and HP 1303 with C 306 as control during the *rabi* season 1982-83. A variety C 306 is a carrier of *Ne1*^{s}.

It is apparent from the results that all the F_1's, S 308 × PNC 306, Kalyansona × PNC 306 and HP 1303 × PNC 306 grew normally and reached full maturity (Table 1), whereas F_1's of the control crosses of S 308, Kalyansona and HP 1303 with C 306, died at 2-3 leaf seedling stage without forming ears and seeds. The hybrid nature of the F_1 plants was identified by glume pubescence. The stem length of normal F_1's ranged from 72.0 to 78.6cm with normal leaf sector extending upto the flag leaf stage which had normal head and varying degree of seed setting. The seed was quite normal, with thousand grain weight ranging from 40.6 to 45.6g. The number of ears per plant ranged from 9.0 to 10.6 with good seed set and a fairly large quantity of F_2 seeds was obtained from these plants. The F_1 plants showed slight yellowing of leaves and manifested low degrees of necrosis (grade, 0-1), as suggested by Hermsen (1963). The result suggests that the variety PNC 306 carried a weaker allele of *Ne1*. As it is suggested from the results in Table 1 that the *Ne1*^{s} (strong) dominant allele present in the variety C 306 has been mutated to a weaker allele in the variety PNC 306 as it is not a completely effective non-carrier in crosses with *Ne2*^{s} carriers. Similarly, Pukhalskii (1972) showed that the durum wheat mutants Senator Sr Sv 2, Sr 48, Sr V 132, Sr V 144 which were obtained by irradiation with neutron and x-rays were ineffective for the preclusion of *Ne1*^{s} gene for hybrid necrosis.

Induction of weak or non-necrotic mutation in carriers of strong necrotic alleles with the desirable characteristics is important if breeders are to utilize them frequently in crossing

programme. Preclusion of hybrid necrosis might be possible by using weak-necrotic mutant variety PNC 306.

The author wishes to express his sincere gratitude to Dr. M. V. Rao, IARI, New Delhi for supplying the seeds of wheat strain PNC 306 used in the present study.

Table 1. Morphological characteristics of different F_1 hybrids crossed with weak-necrotic mutant variety PNC 306

Crosses	Height of main stem (cm)	Number of tillers	1000 grain weight (g)	Necrosis grade	Phenotypic expression
S 308 × PNC 306	72.0	10.6	45.6	0-1	Weakly necrotic
Kalyansona × PNC 306	78.6	9.0	41.6	0-1	Weakly necrotic
HP 1303 × PNC 306	75.6	10.0	40.6	0-1	Weakly necrotic

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Seedling studies on dwarf mutants of tall varieties of rainfed wheat

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Summary

The study on the seedlings of three rainfed wheat varieties and their dwarf mutants indicated that in spite of a positive correlation between the coleoptile length and plant height, it is possible to select genotypes having long coleoptile and well developed root system with dwarf plant type suitable for sowing under rainfed conditions. The study also revealed that coleoptile length is not under pleiotropic control of dwarfing gene. Embryonic root number and length are two independent characters.

Introduction

Traditionally tall wheat cultivars, matchless for quality characteristics have remained unexploited for intensive cultivation in agronomically rich environments, obviously due to tendency for lodging. However, under rainfed agriculture they find no substitute from dwarf category. Under moisture stress conditions, seeds are deeply placed in the moist zone to insure maximum germination. In deep sowings recently-developed dwarf wheat varieties have poor seedling emergence due to short coleoptile length, which is known to show significant positive correlation with the plant height (Allan et al 1961). There is thus, a need for identification/induction of new and effective culm shortening genes for non-lodging with standard coleoptile length for better field emergence. Good root system is another important characteristic of rainfed wheats (Rozentreter 1950; Vedrob 1971). Rajendra Kumar (1984) induced dwarfness through experimental mutagenesis in three tall rainfed wheat cultivars. The present attempt is to evaluate the seedling characters of these dwarf mutants to assess their suitability under rainfed conditions.

Materials and Methods

The experimental material for present study comprised of three rainfed wheat varieties C 306, K 65 and NI 5439 and their ten dwarf mutants in M_5 generation developed through experimental mutagenesis. Ten dwarf mutants along with three parental lines were sown in three replications in random block design at IARI Experimental Farm, New Delhi. The average height of ten plants per replication was recorded from ground surface to the tallest

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tiller's earhead excluding awns at maturity. One thousand seeds were counted from the cleaned seed and the weight was recorded for each plot. Twenty seeds/replication of each line were kept for germination in template stands in a seed germinator at 20° C under moist conditions for seven days as per ISTA rules (Anonymos 1976). The Laboratory experiment was conducted in three replications. Observations on coleoptile length, embryonic root number and maximum root length were recorded on ten seedlings per replication. Random block design analysis were done for calculating critical difference (C. D.) for each character.

Results and Discussion

The plant height, seed weight, coleoptile length, root number and maximum root length of parental lines and their mutants have been presented in Table 1. All the mutants were significantly shorter in plant height than their respective parents. The maximum reduction in plant height compared to parental line was recorded in M-19 (65.5cm) of wheat variety NI 5439 (106.3cm). Reduction in plant height of mutants was associated with the reduction in seed size in all the mutants except three, M-53, M-256, and M-257 of wheat variety K 65.

Seven mutant lines showed significant reduction in coleoptile length as compared to parental lines. Three mutants, M-10 of wheat variety C 306, M-53 of wheat variety K 65 and M-252 of wheat variety NI 5439, did not show any significant difference in coleoptile length as compared to respective parental lines.

The embryonic root number in 7 days old seedlings of seven mutant lines reduced significantly as compared to the parental lines. In the rest three mutant lines, M-53 and M-256 of wheat variety K 65 and M-250 of wheat variety NI 5439, there was significant increase in the root number as compared to parental lines.

The root length of four mutant lines significantly decreased than the parent varieties. Mutant 250 of wheat variety NI 5439 showed significant increase in root length as compared to the parent variety. In the rest five mutant lines the root length remained unaffected.

All the three attributes of rainfed wheats i.e. coleoptile length, root length and root number were combined in only one mutant (M-53) which did not show any reduction in grain weight also. Two mutants (M-10 and M-252) showed long coleoptile (more than 6cm) as well as seminal roots (more than 13cm) but this increase was associated with the reduction in root number. Mutant 250 had increased root number as well as root length but with reduced coleoptile length. Mutant 306 had increased root number only, the coleoptile as well as root length were significantly reduced. In rest of the five mutants the reduction in plant height was associated with the reduction in coleoptile and root number and also with root length in M-56, M-257 and M-253.

Yadav and Sharma (1982) reported a positive association of embryonic root number with root length and suggested selection for root length on the basis of root number. In the present study no such association was apparent since mutant 252 showed increased root length with decreased root number and mutant 256 had decreased root length with increased

Table 1. Plant height, grain weight, coleoptile length, root number and root length in three wheat varieties and their ten dwarf mutants

Parent variety	Mutant	Plant height cm.			1000 grain weight			Coleoptile length			Root number			Root length cm.		
		Parent	Mutant	Diff.	Parent	Mutant	Diff.	Parent	Mutant	Diff.	Parent	Mutant	Diff.	Parent	Mutant	Diff.
C 306	10	116.6	89.2	27.4*	41.0	35.0	6.0*	7.48	6.91	0.57	4.70	3.93	0.77*	13.08	13.45	0.37
K 65	48	115.3	91.8	23.5*	41.0	27.5	13.5*	7.49	5.76	1.73*	3.36	3.00	0.36*	11.68	11.24	0.44
K 65	53	115.3	82.4	32.9*	41.0	39.0	2.0	7.49	7.25	0.24	3.36	3.73	0.37*	11.68	11.37	0.31
K 65	56	115.3	90.0	25.3*	41.0	35.0	6.0*	7.49	6.60	0.89*	3.36	3.03	0.33*	11.68	10.24	1.44*
K 65	256	115.3	89.3	26.0*	41.0	41.0	0.0	7.49	5.89	1.60*	3.36	3.66	0.30*	11.68	9.79	1.89*
K 65	257	115.3	96.3	19.0*	41.0	44.0	3.0	7.49	5.77	1.72*	3.36	3.16	0.20*	11.68	9.44	2.24*
NI 5439	19	106.3	65.5	40.8*	34.0	29.0	5.0*	6.49	3.50	2.99*	3.26	3.03	0.23*	12.87	12.41	0.46
NI 5439	250	106.3	86.3	20.0*	34.0	23.0	11.0*	6.49	4.61	1.88*	3.26	3.56	0.30*	12.87	14.61	1.74*
NI 5439	252	106.3	90.0	16.3*	34.0	30.0	4.0*	6.49	6.45	0.04	3.26	3.00	0.26*	12.87	13.19	0.32
NI 5439	253	106.3	83.4	22.9*	34.0	30.0	4.0*	6.49	4.63	1.86*	3.26	2.96	0.70*	12.87	7.31	5.56*
Mean	–	111.8	86.4	25.4	38.2	33.3	5.4	7.09	5.74	1.35	3.45	3.31	0.38	12.30	11.31	1.48
C. D. (P=0.05)				8.23			3.10			0.70			0.20			1.05

*: Significantly different at the 5% level between parent variety and mutant.

root number.

The faster early growth measured in terms of long coleoptile which is an essential requirement for varieties under rainfed conditions. The study indicated that in spite of a positive correlation between the length of coleoptile and plant height it is possible to select lines having long coleoptile with semi-dwarf plant stature. The study also indicated that embryonic root length is not associated with root number. Such studies may help in selecting suitable genotypes for rainfed conditions. It is also evident from the study that the coleoptile length is not under pleiotropic control of dwarfing genes as reported by Woo and Konzak (1969).

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Ditelosomic analysis of some morphological characters in wheat, *Triticum aestivum* cv. 'Chinese Spring'

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Introduction

Aneuploidy, which is the loss or increase of chromosomes has been widely studied in hexaploid wheat, *Triticum aestivum* (2n = 6x = 42). Aneuploids can be classified as primary or secondary, the former being a loss or increase of complete chromosomes, the latter involve different dosages of chromosomes derived through the misdivision of the centromere.

Due to the availability of the aneuploid series in 'Chinese Spring', which was developed by Sears (1954), the complexity of genetic analysis in hexaploid wheat has been greatly reduced. All the 42 possible telocentrics have been obtained (Sears and Sears 1978). Out of these, 35 are maintained as ditelosomics. They have been obtained by selfing either monotelosomics or monotelodisomics (Sears and Sears 1978). Ditelosomics, in which each line is deficient in one pair of chromosome arms, breed true and are usually of high fertility (Law et al 1987). These are most frequently used to study the role of individual chromosomes, as differences for a particular character can be directly attributed to the effect of the missing arm.

Days to heading, whose nature of dominance is not clear yet, is a complex character. Date of sowing, day length, latitude and day and night temperatures are some of the factors affecting days to heading. Spike length is a genic character. Genes controlling spike length have an additive effect. Leaf size has a direct relationship with biomass and yield of the plant. As compared to other leaves, the flag-leaf makes an important contribution of photosynthates especially at the stage of grain-filling. Hence elucidation of the role various chromosomes play in determining flag-leaf size is important.

In this paper, the contribution of chromosome arms in the performance of some agronomic characters, viz., days to heading, spike length, flag-leaf length and width was estimated by comparing ditelosomics with normal disomics in *T. aestivum* cv. 'Chinese Spring'.

Materials and Methods

The material for the present study comprised of 32 ditelosomics, which could be maintained and confirmed cytologically, and disomics of *T. aestivum* cv. 'Chinese Spring'. The seeds of the ditelosomics were kindly provided by Dr. E. R. Sears, University of Missouri, Columbia, U.S.A., and maintained at the Department of Genetics, University of Karachi, Karachi, Pakistan.

The seeds were placed on moist filter papers in Petri dishes and kept at 20℃. Root tips were collected after 3 days for cytological confirmation. The seeds were resown in small pots and kept at 20℃. After 15 days, the plants were transferred to large pots and kept in the screen house, i.e. in the open, under natural conditions.

The day on which the spike emerged from the boot leaf was taken as date of heading from which days to heading was calculated. Observations on spike length, and flag-leaf length and width were recorded at maturity. Mean and standard error for these variables in each line were calculated. The statistical analysis was carried out using the statistical package SPSS/PC+ and utilizing the command MANOVA. The analysis of variance was done by using the Completely Randomized Design (CRD). Several types of contrasts are available for different types of parameter estimates. The contrast used in our analysis was 'SIMPLE' in which one variable (in this case the disomic) was compared to the rest (the ditelosomics). This type of analysis is not orthogonal (Norusis 1988).

Results and Discussion

The plants were grown in the open, under natural conditions. They were kept in the screen house to protect them from rodents and birds. The plantation was done in Karachi during the period December 1988 to March 1989. Karachi lies at a latitude of 24.51°N and a longitude of 67.02°E. During the plantation period, the maximum monthly temperature ranged from 26.4° to 31.6℃, the mean temperature being 28.4℃ and the minimum from 12.7° to 17.8℃, the mean temperature being 14.4℃. The range of relative humidity during this period was 58.6% to 74.6%. The mean relative humidity was 66.3%. The monthly rainfall ranged from 1.0 to 8.0mm, they mean rainfall being 3.5mm (Source: Regional Daily Weather Report for Sindh and Baluchistan, Regional Meteorological Centre, Karachi Airport, Karachi).

Many genes have been identified in wheat but most of the characters with which the plant breeder is concerned show quantitative variation. In recent years, procedures have been developed which enable biometrical analysis of quantitative characters. To examine the inheritance of such characters, statistical analysis has to be carried out and interpretations made through means, variances and covariances.

The progress of spike development can be measured by the time taken to achieve defined developmental stages (Kirby and Appleyard, 1987). It is possible to quantify development by analysing the rate of initiation of heading. When the ditelosomics were compared to the disomics, Ditelo-1AL, 2AS, 4AL, 5AL, 6AL, 7AL, 3BL, 5BL, 6BS, 3DL, 5DL, 7DS, and 7DL showed highly significant deviations as far as days to heading was concerned. All of these were early as compared to the control population, their mean days to heading being less than that of the disomics (Table 1). Literature available showed the effect of all these chromosomes on heading time. Bhat and Goud (1979), working on monosomic F_2 population derived from the cross of monosomic lines of Pb. C591 with UP

301, reported the effect of chromosomes 5A, 7A, 3D and 7D. Out of these four, monosomic populations 7A, 3D and 7D were early while monosomic population 5A showed delayed heading. Yoshida and Kawaguchi (1984) worked on the monosomics and ditelosomics of 'Chinese Spring'. They grew the monosomics for 3 weeks in the greenhouse and then transferred them outdoors. According to them monosomic 3D and telosomic 1AL were early and monosomics 2B, 3B, 6B, 7B and 6D and telosomics 2AS, 6AS, 5BL, 6BL, 2DS, and 5DL were late. In addition, the effect of chromosomes 1A, 4A, 5A, 3B, 2D and 7D of the hexaploid wheat variety DWR. 39 of *T. aestivum* on heading time was reported by Goud and Sridevi (1988). Of these the populations of Mono 6A and 7D were early while the rest were late as compared to the control population. They, in another report, (Sridevi and Goud 1988) found the influence of chromosomes 4A, 5A, 2B, and 6B in the trisomics of *Triticum durum* cv. HD 4502. All the populations which were trisomic for these chromosomes were late. This indicates the presence of genes imparting lateness on these chromosomes. The other lines which, according to our analysis, differed highly significantly from the disomics were 3AL (F value=42.25), 1BL (47.90), 4BS (20.70), 7BS (50.23), 7BL (14.22), 1DS (10.17), 1DL (60.35), 4DS (63.38) and 4DL (20.47)(Table 2). All of these were early as compared to the disomics.

The length of the spike in wheat is predominantly controlled by polygenes with an additive gene action (Bhat and Goud, 1979). A comparison of the spike lengths of the ditelosomics with the disomic population indicated that Ditelo-3AS, 4AL, 5AL, 1BS, 3BL, 4BS, 5BL, 6BS, 4DS, 4DL, 5DL and 7DS differed substantially from the disomics. Out of these the length of the spike was increased only in Ditelo-5BL, 5DL and 7DS, while in the rest it was reduced (Table 1). The involvement of these chromosomes, as far as the spike length is concerned, is in confirmation with the work done by Bhat and Goud (1979), Yoshida and Kawaguchi (1984), Goud and Sridevi (1988) and Sridevi and Goud (1988). Chromosomes 3A, 4A, 1B, 2B, 3B, 5B, 3D, and 7D were reported to carry genes for spike length by Bhat and Goud (1979). Of these, monosomic populations 4A and 2B had longer spikes when compared to the disomics. Monosomic populations 3A, 1B, 3B, 5B, 3D and 7D were found to have reduced spike length. Yoshida and Kawaguchi (1984) reported the effect of chromosomes 3A and 5A on spike length. According to them, Mono 5A and Monotelo-disomic 5AS had longer spike length (speltoidy) and Telo 3AS had shorter spike length. Goud and Sridevi (1988) found the influence of chromosomes 4A, 5A, 6A, 7A, 3B, 4B, 5B, 6B and 7D on the length of the spike. The families derived from Mono 1B increased the mean spike length whereas the rest reduced it. Sridevi and Goud (1988) reported the effect of chromosomes 4A, 5A, 2B, and 7B. Of these, populations derived from trisomics 5A decreased the mean spike length while the rest increased it. The other ditelosomics which, according to our study, influenced the spike length were 1AS (F value=4.00), 2AS (11.35), 1DS (10.90), 1DL (14.94) and 2DL (12.64)(Table 2). Of these only 1DS increased the length while the rest of the ditelos reduced it.

While leaf ontogeny has an important effect on leaf size, it is distinctively modified by

Table 1. Mean and standard error of the variables studied in the ditelosomic and disomic stock of common wheat, *Triticum aestivum* L. cv. 'Chinese Spring'

Line	Days to heading		Spike length (mm)		Flag leaf			
					Length (mm)		Width (mm)	
	mean	st. err	mean	st. err	mean	st. err	mean	st. err
1AS	85.0	1.0	73.2	2.5	161.7	10.3	9.8	0.5
1AL	78.3	2.4	86.5	2.9	166.3	20.1	10.3	1.1
2AS	80.0	0.8	65.6	2.2	135.0	12.7	11.2	0.4
3AS	86.3	2.0	59.7	3.4	141.9	10.9	10.3	0.3
3AL	78.8	1.2	84.0	3.2	214.5	8.5	14.3	0.4
4AL	79.9	1.4	61.1	3.2	121.1	12.7	6.7	0.4
5AL	82.5	0.8	68.3	3.7	136.4	10.0	9.5	0.8
6AS*	81.0		71.0		194.0		10.0	
6AL	81.2	2.1	84.3	2.6	198.1	17.7	11.9	0.2
7AL	73.8	1.0	78.0	5.4	160.0	10.3	12.0	0.4
1BS	84.3	0.9	66.3	2.4	125.7	8.4	8.7	0.3
1BL	78.2	1.0	86.3	3.2	175.9	8.5	11.7	0.3
2BL*	89.0		86.0		266.0		11.0	
3BS*	90.0		71.0		137.0		10.0	
3BL	80.1	1.9	68.4	4.6	188.1	16.7	12.9	0.6
4BS	76.0	1.5	63.7	2.2	169.7	5.2	7.3	0.3
5BL	80.4	1.2	93.1	2.6	272.6	10.7	14.6	0.3
6BS	83.1	1.1	61.0	2.1	222.8	7.4	8.8	0.3
7BS	79.7	0.9	87.4	2.1	196.2	7.7	12.1	0.3
7BL	80.8	1.0	74.5	1.5	147.0	14.1	10.0	0.9
1DS	80.8	2.4	100.5	11.3	169.3	7.9	11.5	0.3
1DL	73.9	0.8	65.7	2.6	198.3	9.8	10.6	0.3
2DL	86.5	1.5	67.9	4.4	170.8	21.7	11.3	0.8
3DS	89.5	0.5	58.0	5.0	162.5	7.5	8.0	2.0
3DL	76.8	2.5	63.7	3.8	182.7	12.6	13.3	0.7
4DS	77.8	1.0	65.9	3.3	164.4	5.9	9.4	0.5
4DL	79.3	1.2	67.5	1.4	109.8	3.9	8.5	0.3
5DL	78.0	0.8	100.0	2.5	197.0	12.1	12.1	0.4
6DS	85.0	1.0	69.5	2.7	128.2	10.9	8.3	0.6
6DL	90.6	0.9	89.6	5.7	165.6	7.3	10.6	0.4
7DS	77.5	1.1	91.0	1.6	174.3	7.7	12.7	0.4
7DL	75.6	2.4	83.5	3.5	218.5	11.8	13.5	0.7
Disomic	88.3	0.9	82.4	2.0	202.4	6.3	11.8	0.5

* Only one reading available

Table 2. Comparison of the ditelosomics with the disomics, each with one degree of freedom for the variables days to heading and length of spike

	Days to heading		Length of spike	
	MS	F	MS	F
Disomic vs.				
Ditelo 1AS	57.14	2.81	434.77	4.00 *
1AL	514.29	25.27 **	88.10	0.81
2AS	304.88	14.98 **	1233.37	11.35 **
3AS	24.57	1.21	3005.71	27.66 **
3AL	859.65	42.25 **	25.65	.24
4AL	421.05	20.69 **	2638.47	24.28 **
5AL	222.73	10.95 **	1303.35	11.99 **
6AS	52.32	2.57	125.59	1.16
6AL	364.09	17.89 **	28.01	.26
7AL	1392.05	68.41 **	124.49	1.15
1BS	44.31	2.18	711.39	6.55 *
1BL	974.79	47.90 **	148.76	1.37
2BL	0.43	0.02	12.88	.12
3BS	2.70	0.13	125.59	1.16
3BL	441.01	21.67 **	1280.36	11.78 **
4BS	421.23	20.70 **	967.80	8.90 **
5BL	535.16	26.30 **	970.02	8.93 **
6BS	352.13	17.30 **	5894.17	54.23 **
7BS	1022.07	50.23 **	347.96	3.20
7BL	289.29	14.22 **	317.81	2.92
1DS	207.02	10.17 **	1184.47	10.90 **
1DL	1228.12	60.35 **	1624.03	14.94 **
2DL	22.00	1.08	1373.55	12.64 **
3DS	2.58	0.13	1124.46	10.35 **
3DL	680.14	33.42 **	1797.34	16.54 **
4DS	1289.77	63.38 **	3113.28	28.65 **
4DL	416.57	20.47 **	1135.81	10.45 **
5DL	698.91	34.35 **	2036.49	18.74 **
6DS	57.14	2.81	850.67	7.83 **
6DL	22.56	1.11	230.06	2.12
7DS	1056.25	51.91 **	671.67	6.18 *
7DL	1358.31	66.75 **	10.07	.09

* $P<0.05$

** $P<0.01$

Table 3. Comparison of the ditelosomics with the disomics, each with one degree of freedom for the variables length and width of flag-leaf

	Length of flag-leaf		Width of flag-leaf	
	MS	F	MS	F
Disomic vs.				
Ditelo 1AS	8572.40	7.17 **	20.57	6.63 *
1AL	6685.73	5.62 *	11.57	3.73
2AS	19937.25	16.77 **	1.76	.57
3AS	21473.28	18.06 **	14.04	4.52 *
3AL	1392.05	1.17	58.48	18.84 **
4AL	38684.45	32.54 **	153.57	49.47 **
5AL	28524.00	23.99 **	35.64	11.48 **
6AS	68.47	.06	3.27	1.05
6AL	131.76	.11	.02	.01
7AL	11760.99	9.89 **	.18	.06
1BS	16300.52	13.71 **	27.77	8.94 **
1BL	6689.91	5.63 *	.19	.06
2BL	3937.01	3.31	.68	.22
3BS	4160.15	3.50	3.27	1.05
3BL	1331.73	1.12	7.10	2.29
4BS	2965.14	2.49	56.08	18.06 **
5BL	38579.48	32.45 **	59.90	19.30 **
6BS	5356.46	4.51 *	118.30	38.10 **
7BS	518.43	.44	1.25	.40
7BL	15777.92	13.27 **	17.29	5.57 *
1DS	3953.47	3.33	.40	.13
1DL	98.67	.08	9.33	3.01
2DL	6552.13	5.51 *	2.23	.72
3DS	3014.76	2.54	27.84	8.97 **
3DL	2000.40	1.68	11.57	3.73
4DS	16006.77	13.46 **	66.94	21.56 **
4DL	44056.44	37.06 **	57.14	18.41 **
5DL	190.08	.16	.56	.18
6DS	28331.68	23.83 **	63.00	20.29 **
6DL	5941.85	5.00 *	6.68	2.15
7DS	7126.17	5.99 *	6.25	2.01
7DL	2174.67	1.83	22.15	7.13 **

* $P < 0.05$
** $P < 0.01$

environmental factors such as temperature, day length, irradiance etc. (Kirby and Appleyard 1987). Deviations were noticed in Ditelo-2AS, 3AS, 3AL, 1BS, 1BL, 5BL, 7BL, 2DL and 3DS, all of which decreased the flag-leaf size except Ditelo-3AL and 5BL (Table 1). This indicates the presence of positive tendency genes on 3AL and 5BL, i.e. genes that increase the size of the flag-leaf. Earlier reports on flag-leaf size revealed the effect of these chromosomes (Sridevi et al 1989). The other chromosomes which, according to our study, affected flag-leaf size were Ditelo-1AS, 1AL, 4AL, 5AL, 7AL, 4BS, 6BS, 4DS, 4DL, 6DS, 6DL, 7DS and 7DL (Table 3). All of these had a negative effect on the size of the flag leaf, i.e., the size of the flag-leaf was reduced in these ditelos. As the characters under study are quantitative, the effect of many chromosomes on these characters has been observed. Our analysis led to the location of genes governing days to heading, spike length and flag-leaf size on different chromosomes.

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Studies on varietal mixtures in wheat

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Summary

A study with varietal mixtures of most promising wheat varieties of Northern India was undertaken to assess yielding potential and rust severity. Varietal mixture of varieties of good fertility irrigated conditions, low fertility rainfed conditions and a mixture of varieties belonging to both these two crop production conditions were composed to study the above two facts. It was observed that mixtures were not superior than the best adapted genotype. However, the mean yield of mixture was higher than the component mean. Yield advantage up to 14.2 per cent was recorded in mixture composited from varieties belonging to rainfed conditions. A variety in individual stand showed high incidence of rust but the same when grown as a part of mixture the rust incidence and severity is reduced.

Introduction

Pure line varieties of wheat suffer from disease as soon as new virulences in pathogen are evolved. There is always risk in cultivation of such varieties on commercial scale. Multilineal varieties and variental blends have been advocated to minimise yield losses due to parasitic fungi. Under these approaches the pathogen is allowed to propagate on some of the components of multilines/varietal mixture. Susceptible components when grown as a part of varietal mixture suffered less from rust damage (Suneson 1960; Borlaug 1958; Jensen and Kent 1963) and there had been reported yield advantages of varietal mixture (Jensen 1965; Pandey et al 1978; Gill et al 1981). Varietal mixtures have shown stable yield than the pure varieties (Marshall and Brown 1973).

The work on varietal blends of similar height, different height and of different maturity group has resulted in considerable superiorty of mixture for yield and lowering of disease severity. In the present study varietal mixture from varieties of two different crop production conditions i.e. irrigated and rainfed were studied in respect of their yield performance and rust severity.

Materials and Methods

The materials for the present study comprised of 10 most popular wheat varieties, HD 2177, HD 2122, HD 2204, WL 711, WH 147, HD 2009, Kalyansona (irrigated conditions) WL 410, IWP 72 and HD 1981 (rainfed conditions). Three multilines and three varietal mixture composed from these varieties. These varietal mixture were consituted as follows:

Mixture-A. Constituted from 7 wheat varieties suitable for cultivation under highly fertile irrigated conditions.

Mixture-B. Constituted from 3 wheat varieties suitable for cultivation under rainfed condition.

Mixture-C. Constituted from all the ten varieties of two different crop production condition.

Yield potential of the varieties and mixtures was studied in a randomised block design for two subsequent crop seasons i.e. 1978-79 and 1979-80 at IARI, New Delhi. In these experiments there were four replications and grain yield was harvested from a plot of 5.0×0.92m. Rust data was recorded on modified Cobb's scale and the observation were taken on three different dates of one week interval. Height reading of incidence and severity of infection was utilised in making conclusion. Yield data of individual years was analysed statistically.

Results and Discussion

Analysis of variance has shown that significant difference in yield was observed in data of both the crop season (Table 1).

Mixture vs Components: Among the component of mixture A of 1979 crop season, wheat variety WL 711 yield significantly higher than mixture A, while HD 2204, WH 147 yielded similar to it. The remaining three components of this mixture i.e. HD 2177, HD 2122 and HD 2009 were inferior in yield. The result of 1980 yield test favoured the superior performance of HD 2122 while the other entries were of as good as the mixture of itself. Among the components of mixture B, none was superior in yield in both years of test. Mixture B was significantly superior in yield over IWP 72 (1979) and WL 410 (1980). Mixture 'C' which consisted of 10 components, yielded significantly inferior than HD 2204, WL 711 and HD 1981 in test of 1979 while in the following year all the components of this mixture except WL 410 were at par to mixture yield.

Mixture vs Multilines: Multiline KML 7406, and KSML 3 showed significantly lower yield than mixture A and mixture B during the test of 1979 while in the following year the yield potential of the multilines and mixtures were similar.

Mixtures vs Component means: The mean yield of mixture were compared with the mean of their respective components by working out t-value. As shown in Table 2; the mean yield of mixture A and mixture B was statistically higher than the mean yield of their respective components during the test of 1979. Yield gain up to 12 per cent was recorded in these mixtures. There was no advantages in mixture C. Mixture B and Mixture C in test of 1980, showed yield superiority as high as 14.2 per cent over component means.

It is evident from this study that mixture yield is not high than the best adapted genotype. However, yield gain over the component means is observed which is in accordance with the study of Jensen (1965), Gustafson (1953), Frey and Maldonado (1967), Clay

Table 1. Mean Yield of mixtures, multilines and varieties

Sl. No.	Varieties	Yield (Q/ha) (1979)	Yield (Q/ha) (1980)
1.	Mixture A	40.7	40.2
2.	Mixture B	37.7	42.3
3.	Mixture C	32.3	41.5
4.	MLks-11	37.1	40.2
5.	KML-7406	30.0	44.0
6.	KSML-3	29.7	37.7
7.	HD 2177	30.6	37.5
8.	HD 2122	33.6	45.6
9.	HD 2204	40.7	42.4
10.	WL 711	48.3	44.5
11.	WH 147	35.3	37.7
12.	HD 2009	32.8	39.9
13.	Kalyansona	34.6	39.4
14.	WL 410	33.4	34.7
15.	IWP 72	28.9	39.4
16.	HD 1981	38.6	37.2
	S. D.	6.3	5.1

Table 2. Comparision of yield (Q/ha) of varietal mixtures with their component mean

S. No.		(1979)	% Gains over component Mean	(1980)	% Gains over component Mean
1.	Mixture A	40.7*	11.4	40.2**	−1.9
	Component mean	36.6		41.0	
2.	Mixture B	37.1*	12.2	42.3*	14.2
	Component mean	33.6		37.1	
3.	Mixture C	32.3**	−9.2	41.5*	4.4
	Component mean	35.7		39.8	

* Significantly higher yield

** Significantly lower yield

Table 3. Stem and leaf rust severity in multilines, varietal mixtures and their components

Sl. No.	Varieties	Stem rust				Leaf rust			
		(1979)		(1980)		(1979)		(1980)	
		% of Popln infected	Severity	% of Popln infected	Severity	% of Popln infected	Severity	% of Popln infected	Severity
1.	Mixture A	2	tS	0	0	5	20S	10	10S
2.	Mixture B	5	5S	0	0	2	10S	5	5S
3.	Mixture C	10	10S	3	5S	10	30S	20	20S
4.	MLKS-11	1	tS	0	0	7	20S	30	20S
5.	KML-7406	0	0	0	0	1	tS	2	5S
6.	KSML-3	0	0	0	0	2	5S	10	20S
7.	HD 2177	1	tS	0	0	5	10S	2	tX
8.	HD 2122	0	0	0	0	5	tS	10	10S
9.	HD 2204	1	tS	0	0	0	0	1	tS
10.	WL 711	15	10S	3	10S	10	60S	100	80S
11.	WH 147	10	20S	5	10S	10	30S	90	80S
12.	HD 2009	0	0	0	0	5	10S	5	10S
13.	Kalyansona	15	10S	10	10S	90	60S	100	80S
14.	WL 410	15	40S	5	20S	5	20S	10	10S
15.	IWP 72	0	0	0	0	1	tS	2	tS
16.	HD 1981	0	0	0	0	3	20S	10	5S

Popln = Population

and Allard (1969), Shortner and Frey (1979), Shorter (1977), Browning (1957) and Pandey et al (1978). This finding suggests that varietal mixture composed from varieties belonging to particular crop production condition are consistently higher in yield than mixture composed from varieties of different crop production conditions.

Table 3 shows incidence (percentage of plants affected) and intensity of black and brown rusts in respective population of mixtures and their components. It was observed that mixtures had a clear cut advantage in both years of testing over its susceptible components viz WL 711, WH 147, Kalyansona and WL 410. The incidence of black rust in Mixture A was 0-2 per cents with intensity 0-tS as compared to the highly susceptible compoenent WH 147 with incidence of disease 5-10 per cent and intensity 10-20S. Similar comparison with other susceptible components show that disease is checked in mixture. All the mixtures exhibited less disease suscepitbility than their respecitive susceptible components. This trend is distinctly discernible for brown rust as well. Susceptible varieties when mixed in a mixture population the rust build up is very much reduced in comparison to their pure stand. Most of the inoculum that falls on the resistant varieties is further reduced. The reduced rust severity as observed here are similar to those study of Wolfe and Barrett (1977) with powdery mildew of barley and wheat mixture studies of Borlaug and Gibler (1953), Kassem et al (1975) and Barrett (1978).

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Scope of some physiological characters in improvement of durum wheat under stress condition

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Introduction

Physiological characters viz., free proline accumulation, relative water content and chlorophyll content are considered to be the important potential metabolic measures for drought resistance in cereals (Walldren et al 1974 in Sorghum; Hanson et al 1979 in barley; Tregubenke 1979 in corn; Dorofeev 1979, Parshina et al 1979, Morey 1980 and Tandon and Halloran 1982 in wheat). However, no systematic approach has been attempted to know the genetic mechanisms involved in these three physiological characters in durum wheat. The importance of such metabolic traits in breeding wheat for stress condition is obvious. Besides, durums are known for their intrinsic tolerance to drought (Talwar 1979). Hence, the present genetic study was designed to explore the utility of such traits in improvement of durum wheat under stress condition.

Materials and Methods

Forty hybrids derived from line × tester pattern were sown in a randomised block design with three replications during *rabi* season at College of Agriculture, Dharwad. The sowing was done by dibbling the seeds at a distance of 30cm rows on 20 meter long, spaced 15cm apart. The leaf samples were collected from five plants selected at random in each replication to estimate the following physiological characters. Free proline content (FPC) was estimated as per the method suggested by Bates et al (1973); Relative water content (RWC) was determined according to the procedure indicated by Bayers and Weatherly (1968); and Total chlorophyll content (TCC) was estimated according to Arnon (1949). The biometricol approach suggested by Kempthorne (1957) was used for obtaining estimates of combining ability effects and variances.

Results and Discussion

Analysis of variance due to crosses showed the significant differences among the hybrids attributable to the differences existing among the lines and testers (Table 1). The variances

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due to lines and testers were also significant for three physiological characters. The significant differences of lines × testers was also observed for all the traits studied.

The ratio of general combining ability (gca) variance to sca variance effects indicated that the nature of gene action was predominately additive type for relative water content while non-additive gene action was noticed for proline content and total chlorophyll content (Table 1). These findings indicated that both allelic and non-allelic interactions are operating for the expression of these characters.

The general combining ability effects of the parents for proline content indicated that varieties, JNK 6-1-184, A 9-30-1, NP-200 among testers and only MACS-9 among lines, possess favourable genes (Table 2). And for relative water content three testers (JNK 6-1-184, A 9-30-1 and NP 406) and two lines (HD 4502 and Meghadoot) showed desirable gca effects. Similarly, for total chlorophyll content, two lines (NP 406 and N-59) and two testers (HD 4513 and MACS-9) indicated significant additive effects. The parents involved in this study have been bred particularly, for drought prone and rainfed areas of India. Hence many parents have indicated fixable type gene action for all the physiological characters. Since the gca effects are of practical use to the breeder, these nine parents could be utilized for *inter* mating population involving all possible crosses among them and

Table 1. Analysis of variance for combining ability and variance estimates for three physiological characters in durum wheat

Source of variation	D. F.	Mean sum of squares		
		Proline content	Relative water content	Total chlorephyll content
Crosses	39	25238.69**	299.52**	3.28**
Lines	7	6214.16**	308.48**	3.07**
Testers	4	15550.75**	1175.48**	11.09**
Lines × Testes	28	7041.50**	172.14**	2.23**
Error	104	736.10**	8.42	0.12
		Estimates of components of variances		
$\hat{\sigma}^2_l$	7(28)	−55.16**	9.08**	0.06
$\hat{\sigma}^2_t$	4 (28)	354.55	41.80**	0.40**
$\hat{\sigma}^2_{lt}$	− −	2268.46	54.57	0.70
$\hat{\sigma}^2_{gca}$	−28	196.97 (6)	29.22** (6)	0.25* (6)
$\hat{\sigma}^2_{sca}$	28, 104	3223.66**	9.74**	1.86**
$\hat{\sigma}^2_{sca}/\mu^2_{sca}$	− −	0.06	3.00	0.13

* and ** Significant at 5% and 1% level respectively.

adopting breeding methods like biparental matings as well as mating among the selected plants in early segregating generations. Such population would serve the maximum scope for selection of stress tolerance/resistance types.

Seventeen cross combinations indicated significant sca effects for one or two physiological characters (Table 3). These crosses were also in the combination of high × high (H × H), high × low (H × L), low × high (L × H) and low × low (L × L) combining lines. The cross of H × H combination involves genetic interactions of additive × additive types which were fixable in nature and also less prone to adverse environmental conditions. Therefore, desirable segregants with fixable types were expected from such cross combinations. As observed in our study, several crosses would throw some desirable segregants; MACS-9

Table 2. Estimates of GCA effects of the paprents for three Physiological characters in *durum* wheat

Entries	Proline content	Relative water content	Total chlorophyll content
Testers			
Bijaga yellow	14.15**	−8.5 **	−0.72**
HD 4502	−21.85**	6.4 **	−0.23*
HD 4513	33.20**	−3.7 **	0.56**
Meghadoot	−18.61**	8.02**	−0.41**
MACS-9	21.42**	−2.24**	0.82**
Lines			
JNK-6-1-184	23.97**	3.17**	−0.06
Khorchia-65	−40.83**	−1.43	−0.36**
A-9-30-1	16.35**	5.64**	−0.23*
MACS-45	−01.62*	0.83	−0.67**
A-206	−12.61**	−5.02**	−0.19
NP-406	−4.55	5.37**	0.70**
N-59	3.16	−6.37**	0.30**
NP-200	16.02**	−2.18**	0.130
S.E. ($\hat{gi}$) lines	3.96	0.75	0.10
S.E. ($\hat{gj}$) testers	3.13	0.59	0.10
S.E. ($\hat{gi}$-$\hat{gj}$) lines	5.61	1.05	0.14
S.E. ($\hat{gi}$-$\hat{gj}$) testers	4.49	0.83	0.10

* and ** significant at 5% and 1% level respectively.

Table 3. Estimates of specific combining ability of some desirable crosses in *durum* wheat

Crosses	Proline content	Relative water content	Total chlorophyll content
Bijaga yellow × JNK 6-1-184	−48.39** (L×H)	7.83** (L×H)	0.34 (L×L)
Bijaga yellow × NP 200	17.55** (L×H)	−16.95** (L×H)	−0.07 (L×L)
Bijaga yellow × A 9-30-1	22.03* (L×L)	4.43* (L×L)	0.02 (L×L)
Bijaga yellow × NP 406	−14.32 (L×L)	10.64** (L×H)	−1.37 (L×H)
HD 4502 × NP 200	70.48** (L×H)	−11.76** (H×L)	−1.40** (L×L)
HD 4502 × N-59	79.85** (L×L)	5.51** (H×L)	−0.37 (L×H)
HD 4502 × NP 406	−40.11** (L×L)	1.73 (L×L)	2.52** (L×H)
HD 4513 × A 9-30-1	36.76** (L×H)	−6.34** (L×H)	−0.26 (H×L)
HD 4513 × NP 200	33.67** (L×H)	1.58 (L×L)	0.82** (H×L)
HD 4513 × N-59	65.23** (L×L)	3.13 (L×L)	0.69** (L×L)
HD 4513 × A 206	−25.13** (L×L)	−1.64 (L×L)	−0.43** (H×L)
Meghadoot × JNK 6-1-184	33.8** (L×H)	16.94** (H×H)	−0.07 (L×L)
Meghadoot × A 9-30-1	−39.62** (L×H)	11.71** (H×H)	1.01** (L×L)
MACS-9 × A 9-30-1	60.55** (H×H)	3.53* (L×H)	0.11 (H×L)
MACS-9 × NP 200	92.03** (H×H)	−7.01** (L×L)	0.76** (H×L)
MACS-9 × NP 406	28.13** (H×L)	6.60** (L×H)	−0.44** (H×H)
MACS-9 × A 206	−8.61 (H×L)	11.49** (L×L)	0.62** (H×L)
S. E. $(\hat{sij}\text{-}\hat{sji})$	±12.54	±2.36	±0.28

* and ** significant at 5% and 1% respectively.

NOTE : Letters in the parenthesis are the general combining ability effects of the parents in High × High, (H×H) High × Low (H×L), Low × High (L×H) and Low × Low (L×L) combinations.

× A 9-30-1 and MACS-9 × NP200 for proline content and Meghadoot × JNK 6-1-184 and Meghadoot × A 9-30-1 for relative water content.

The crosses involving in high × low and low × low combinations were considered to be in dominant × additive and dominant × dominant types in nature, respectively. Since the latter combination was unpredictable because of non-allelic interactions, the possibility of selecting the desirable characters in the subsequent generations will be very rare. However, the former combinations (high × low or low × high) might throw some desirable transgressive segregants, if the additive genetic system present in one of the parents complements the epistatic effect present in the cross. As noticed in our study, nine crosses for proline content, five crosses for relative water content and five crosses for total chlorophyll content could be exploited extensively for increasing the gene frequencies of desirable alleles.

Finally, ten hybrids, showed desirable performance for physiological characters in combinations like proline content with relative water content and total chlorophyll content as well as relative water content with total chlorophyll content. They were obtained by crosses Bijaga Yellow × A 9-30-1, HD 4502 × N-59, HD4513 × NP 200, HD 4513 × N-59, Meghadoot × JNK 6-1-184, Meghadoot × A 9-30-1, MACS-9 × A 9-30-1, MACS-9 × NP200, MACS-9 × NP 406, and MACS-9 × A 206. Therefore *inter se* crossing of these ten hybrids for multiple parent input into a gene pool, would throw the desirable physiologically efficient recombinants by breaking tight linkage.

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Stability parameters of grain yield and its components in durum wheat*

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Summary

Stability parameters of 40 varieties of durum wheat (*Triticum durum* Desf.) alongwith 5 commercial bread wheat varieties were studied. Highly significant genotypic differences were obtained for all the characters studied (plant height, tillers per plant, biological yield, grain yield, harvest index, grains per spike, spikelets per spike and 1000-grain weight). The linear component of G×E interaction was significant for plant height, tiller number, biological yield, grain yield and harvest index. Variety WH 822 appeared to have high mean, unit regression and non-significant non-linear component for all the traits, except plant height. Variety HI 8078 was good for four and WH 823, CPAN 1478 and Sapi's for three characters. These varieties were also better/average for other traits. The blends did not give any trend in terms of their stability parameters compared to their base varieties. Different mechanisms were operative in different varieties as regards the contribution of yield components to the stability of grain yield.

Introduction

Durum wheat represents the second major species of the genus *Triticum* being grown in the world. India is one of the largest durum wheat producing countries and has largest area under durum wheat (2m. ha.). Due to higher yield jump in bread wheat and high susceptibility of durum wheats to rusts particularly yellow rust in North India in recent past, there is shifting of wheat growers from durum to bread wheat.

In any breeding programme it is necessary to screen and identify phenotypically stable genotypes which could perform uniformly under different environmental conditions. Such a breeding effort requires basic information on Genotype × Environment (G × E) interaction of grain yield alongwith other traits. Grafius (1959) emphasized that the study of individual yield component can lead to simplificatin in genetic explanation of yield stability and hence is valuable to breeders in prediction and determination of the effect of environment.

Durum wheats generally lack stability of performance and depict in-consistant behaviour when grown over wide range of environments. To make them competitive to *aestivum*, it is essential to develope possessing stable performance. The present study was taken up to study the stability parameters of some durum wheat (*Triticum durum* Desf) varieties.

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Material and Method

The present study was carried out on 45 varieties consisting of 36 diverse varieties and four biblends (equal parts) of durum wheat and five commercial bread wheat varieties. All the genotypes were grown in randomized block design consisting of three replications in 3 artificially created environments as given below :

i) Normal sown, high fertility and irrigated (E1)
ii) Medium fertility and restricted irrigations (E2)
iii) Rainfed and low fertility (E3).

The data were recorded for traits as shown in Table 1. Stability parameters of 45 varieties were computed using the model proposed by Eberhart and Russell (1966).

Results and Discussion

The introduction of dwarf wheat varieties led to the striking increase in the wheat yields all over the world. However, in developing countries limited input conditions put considerable constraints on the full exploitation of genetic potential of the new wheat varieties. In developing countries like India there is large area of rainfed wheat where annual fertilizer consumption per unit area is also very low. Under such situation it becomes imperative to test the wheat genotypes under rainfed, restricted irrigation/fertility conditions to identify their predictable response to inputs and irrigation levels. Hence G×E interaction would be of great interest if it is estimated under aforesaid conditions which may be more realistic to wheat growing situations. Eberhart and Russell (1966) suggested an approach to identify stable and responsive genotypes. According to their model, a stable genotype is characterized by average response (unit regression) and least deviation accompanied by high mean.

Highly significant variances due to varieties revealed the presence of genetic variability in the material included in this study for all the traits studied (Table 1). The linear component of G×E interaction was significant for plant height, tiller number, biological yield, grain yield, and harvest index. Therefore, prediction for these traits appeared to be feasible. This also shows the existence of additive type of gene-action for these traits. Significance of pooled deviation for all these traits showed that the varieties differed considerably with respect to their response to different environments for these traits. Singh and Singh (1980) and Jatasara and Paroda (1980) observed similar estimates of linear proportion of G×E interaction for grain yield in bread wheat. Kaltsikes and Larter (1970) observed the same for grain yield, plant height and days to maturity in durum wheat. Nanda et al (1983), in bread wheat, found larger predictable portion for plant height and number of spikelets par spike.

Seven best varieties based on linear (b) and non-linear ($S^{-2}d$) components of G×E interaction and high mean values are listed in Table 2. Varieties CC530, JU 72, R6009 gave high grain yield, b value above average and $S^{-2}d$ non-significant. Therefore, these varieties were suitable for favourable environment. However, WH 822, R 6011, HI 8078, CPAN6038 and R 6032 produced average grain yield, b value near unity and $S^{-2}d$ non-sig-

Table 1. Pooled stability analysis of variance for eight characters in wheat

Characters	Mean sum of squares					
	Genotypes (MS_1)	Env. + (Geno. × Env.)	Environment (Linear)	Genotype × Environment (Linear)(MS_2)	Pooled deviation (MS_3)	Pooled error
Plant height	354.05**	61.75	4015.89**	24.44**	10.35**	14.78
Tiller number	4.07**	9.38	753.95**	2.12**	0.840**	2.35
Biological yield	66.98**	382.13	30301.19**	67.31**	25.09**	61.61
Grain yield	16.24**	68.10	5229.67**	14.93**	4.39**	12.90
Harvest index	15.51**	5.87	41.67**	9.38**	3.65**	2.57
Number of grains per spike	53.34**	24.39	1751.72**	4.05	5.90**	7.30
Number of spikelets	4.29**	1.28	72.55**	0.455	0.51	0.22
1000 grain weight	22.14**	4.42	89.88**	2.07	4.82**	1.92
d. f.	44	90	1	44	45	264

*, ** Significant at 5 per cent and 1 per cent level of significance respectively.

Table 2. Seven best varieties of durum wheat based on high mean, unit regression and non-significant squarred deviation ($S^{-2}d$)

Trait/variety	$\mu+di$	bi	$S^{-2}d$	Trait/Variety	$\mu+di$	bi	$S^{-2}d$
Plant height				Tiller number			
1. CC 530	78.44	0.856	7.406	1. Meghdoot	9.60	0.937	0.794
2. H 19	74.22	0.972	-4.175	2. WH 825	7.73	1.076	-0.449
3. H 22	80.91	1.098	2.467	3. CPAN 6048	7.93	1.153	-0.420
4. HD 4502	74.13	1.019	-4.746	4. WH 822	8.06	0.957	1.091
5. R 6039	74.26	0.777	-3.068	5. CPAN 6038	7.93	1.153	-0.420
6. JU 72	78.20	1.039	-3.063	6. HI 8078	7.71	0.826	0.360
7. CPAN 1478	78.42	0.348	13.43	7. R 6039	7.77	1.113	-0.278
Biological yield				Grain yield			
1. R 911	40.46	0.923	-16.00	1. WH 822	20.87	1.015	7.280
2. R 6011	43.97	1.067	11.968	2. R 6011	19.48	1.053	3.786
3. WH 822	46.62	0.975	24.368	3. HI 8078	19.20	0.933	-2.957
4. Sapi's	42.48	1.004	-18.449	4. CPAN 6038	19.16	1.085	0.051
5. JU 72	41.35	1.291	8.519	5. R 6032	17.76	1.055	-1.400
6. R 6932	40.24	0.853	6.330	6. R 911	16.97	0.863	-0.321
7. HI 8078	42.75	0.845	-9.657	7. V 43	17.50	0.904	-3.390
Harvest Index				Number of grains per ear			
1. WH 822	44.78	0.586	0.066	1. WH 822	67.77	0.963	1.549
2. CPAN 6058	44.98	1.997	3.671	2. Sapi's	65.93	0.827	-1.800
3. H 22	43.37	1.416	-0.405	3. JU 133	63.85	1.078	-0.934
4. WH 823	44.20	1.668	-0.264	4. WH 823	68.26	1.065	-2.252
5. WH 826	43.07	1.329	-0.616	5. V 44	69.36	0.998	2.602
				6. JU 132	64.02	1.418	5.900
				7. CPAN 1478	63.04	0.944	-1.255
Number of spikelets per spike				1000 grain weight			
1. WH 823	22.03	0.930	0.162	1. WH 825	52.77	0.258	0.187
2. CPAN 6048	22.60	0.541	0.320	2. CPAN 6038	54.22	0.721	-0.272
3. WH 822	22.52	0.560	1.539	3. HI 8078	54.11	1.275	0.629
4. CPAN 1478	21.67	1.049	1.123	4. WH 822	51.88	0.925	-0.276
5. JU 133	21.43	1.120	4.736	5. V 44	51.77	1.275	0.630
6. JU 132	21.75	0.742	-0.040	6. V 40	49.77	1.579	0.299
7. Sapi's	21.48	0.528	1.423	7. Bj Yellow	50.22	1.798	0.701

nificant hence were stable and responsive. Amongst *aestivum* wheat varieties C 306 was desirable because of its average grain yield, stable and responsive behaviour. Similar results have been reported by Chowdhury and Paroda (1983) for C 306 variety. HD 2009 was also found to be average performer and stable. WH 157 was suitable for favourable environment since it had above average linear regression.

Variety HD 4502 appeared to be the dwarfest, stable and responsive and hence is desirable for breeding dwarf types. Similarly, varieties CC530, H19, H22 and R6039 had low height, b value near one and S^2d near zero. Amongst *aestivum* varieties WH 147 and WH283 were stable and responsive for plant height and their performance over environments, can be predicted. Variety Meghdoot gave maximum tillers followed by WH 825, CPAN6038, WH 822 and CPAN6039. These varieties also had unit regression and non-significant non-linear component and hence were most desirable for this trait. Varieties BJ yellow, WH820, V43, R6009 and R6029 also had high tillering, but their regression values were more than unity. Hence these were suitable for better environment. Among *aestivum* wheats WH 157, WH 147 and HD 2009 had high mean, S^2d non-significant but regression values more than unity hence were more suitable for better environment.

Maximum total biological yield was produced by genotypes V 45, CC530, BJ. yellow, V41, R6009, CPAN6038 and CPAN6048, however, their b values were above average indicating their suitability for better environment. Varieties R911, R6011, WH 822, Sapi's and JU72 also produced higher biological yield, had b value near unity and S^2d equal to zero indicating that these were the most stable and responsive varieties for biological yield. Variety C 306 produced maximum biological yield, had average linear regression and non-significant S^2d, hence was stable and responsive. Variety WH147 had high biological yield, b value less than unity and S^2d non-significant and, therefore, was more suitable for poor/medium fertility situations.

For harvest index, varieties WH 822, CPAN6058, R6011, H 22 and WH823 were found to be the most desirable genotypes because of their high mean performance, stable and responsive behaviour. Varieties WH 820, CPAN 6038, CPAN 6048, JU 133, were more suited to better environment. Among *aestivum* wheats WH 147 gave high mean, linear regression above average and S^2d near zero and hence may be taken as suitable for better environment while C 306 was more suited to poor environment. Variety WH 283 appeared to be the stable and responsive variety with high mean. For number of grains per ear, varieties WH822, Sapi's, JU133, WH 823 and V44 were desirable because of their high mean, stable and responsive behaviour. Varieties H22, Meghdoot, WH825, WH826, V41, V45, CPAN6038 were more suitable for better environment. On the other hand varieties V 43, R6011, CPAN6048 were more suitable for poor environment. In *aestivum*, WH 157, WH 147, WH 283 and C 306 had average performance, responsive and stable behaviour.

Varieties CC 530, H 22, WH 820, WH 826 and V 41 produced average number of spikelets per spike, b value above average and S^2d equal to zero indicating their suitability for better environment, while, V42, V207, R6011 suited more to poor environment. Var-

ieties WH825, CPAN 6048, WH 822, CPAN 1478 and JU 133 gave high mean, stable and responsive behaviour. Amongst *aestivum* wheat varieties WH 157, Wh 147 and C 306 were desirable since they had high mean, stable and responsive behaviour. HD2009 and WH283 were suitable for poor environment. Varieties WH 825, CPAN 6038, HI 8078, WH 822 and V 44 had high grain weight, b value near unity and $S^{-2}d$ near zero indicating that these were stable and responsive for grain weight. Varieties V 45, V 207, R 6009, R 6029, R 6039, R 6011, CPAN 6048 also produced high grain weight but were unstable hence their performance cannot be predicated over environments. Among *aestivum* wheats, WH 157, WH 283 and C 306 were stable and responsive with average mean performance. Varieties WH 147 and HD 2009 had significant $S^{-2}d$ value and hence they were not predictable for this trait.

If we review critically the overall situation from Table 2, it becomes obvious that variety WH 822 appeared to have high mean, stable and responsive behaviour for all the traits except plant height for which it had low mean, non-significant $S^{-2}d$ and low b. Variety HI 8078 was good for four characters, WH 823, CPAN 1478 and Sapi's were good for three characters. These varieties were also better/average for almost all other traits though not shown in this table.

The varietal mixtures have been reported to increase and stabilize the production in some crop plants. Khalifa and Qualset (1974) reported that the binary mixtures of wheat gave higher yield than either of the components. So a synthesis of acceptable heterogeneous population is necessary where economic advantage due to individual as well as population buffering can out weight those advantages obtainable from growing single varieties. In the present study the behaviour of mixture showned no particular trend. For example in almost all the cases where mixtures were found stable and responsive, at least one of the variety included in these mixtures was stable and responsive for that particular trait. However, one mixture WH 823 + CC 530 showed stability and responsiveness compared to its base varieties i.e. WH 823 and CC 530.

The results listed in Table 2 further suggest that the stability of grain yield was contributed by different characters in different varieties. For example, the grain yield stability of variety WH 822 is contributed by 7 characters, while of R 6039 and R 911 by 2, of R 6038 by 3 characters. Similar results have been reported by Chowdhury et al (1982) and Chowdhury and Paroda (1983).

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Genetic divergence in some wheat strains and their hybrids

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Introduction

Quite often genetic divergence among parents has been referred to as a deterministic factor of the inherent potential of a cross in terms of its *per se*, combining ability effects and segregational ability. In that context it is belived that the high genetic divergence would lead to heterotic response in F_1 and high frequency of transgressive segregants in F_2 generation. High genetic divergence was not found favourable for heterotic response rather medium divergence among parents figured to be more compatible level of divergence (Srivastava and Arunachalam 1977; Arunachalam et al 1984; Behl et al 1985). Moreover, it is not necessary that group distances measured by multivariate analysis, especially refer to actual genetic divergence among parents entering crosses (Behl et al 1985). Rather clustering pattern of F_1 hybrids was found to be influenced by the dominace relationship for few traits with major contribution to total D^2-value among parents entering crosses. Quantitative assessment of degree of divergence among genotypes entering crosses and their hybrids in thus essential. Present study was, therefore, conducted to determine genetic divergence among 36 F_1 hybrids and their 9 parents.

Materials and Methods

Nine cultivars, namely, 1. WL 711, 2. NP 846, 3. WG 377, 4. HD 1981, 5. UP 262, 6. HD 1925, 7. HD 2122, 8. Raj 821 and 9. Sonalika of wheat (*Triticum aestivun* L.), suitable for different agro-ecological conditions, were crossed in all possible combinations (excluding reciprocals) to yield 36 F_1 hybrids. The parents and F_1s were evaluated in a randomized block design with three replications in two locations with centrasting environments i.e., normal environment (irrigated) at Haryana Agricultural University, Hisar and stress environment (rainfed) at Dry Farming Research Centre (HAU) Bawal. The plot size consisted of a 3 m long single row for each parent and F_1. The intra- and inter-row plant distances being 15 cm and 30 cm, respectively. Observations on tiller number (NT), number of grains per ear (NG), 1000-grain weight (GW) and grain yield (GY) were recorded on randomly selected five plants from each entry per replications in each environment.

Following the analysis of variance and covariance, the data were subjected to multivariate analysis. Mahalanobis'D^2-statistic was used for assessing genetic divergence among genotypes. Generalized statistical distances (TOCHER Method) described by Rao (1952) were used to classify parents and F_1s into different clusters. Heterosis was estimated over mid-parent.

Results and Discussion

Analysis of variance revealed significant differences among parents and hybrids and parents vs hybrids for all the characters in both the environments except for number of grains per ear in stress environment and grain yield in both the environments. This indicated sufficient amount of genetic variability in the present material. This was further substantiated by the fact that D^2 values for 990 combinations ranged from 3.32 between HD 1925 and HD 1925 × HD 2122 to 439.91 between NP 846 and HD 1925 × Sonalika in normal environment and from 2.23 between WG 377 × Sonalika and HD 1981 × Raj 821 to 462.75 between NP 846 and Sonalika in stress environment. Such a range among parental combinations varied from 15.69 between HD 1925 and Sonalika to 428.26 between NP 846 and HD 2122 in normal environment and from 6.64 between Raj 821 and Sonalika to 462.75 between NP 846 and Sonalika in stress environment.

Table 1. Grouping pattern of 9 parents and 36 hybrids in normal and stress environments

Cluster	Number of genotypes	Genotypes (parents and hybrids)
		Normal environment
I	16	1, 3, 6, 1×3, 1×4, 1×5, 1×6, 1×7, 1×8, 1×9, 3×4, 3×7, 3×9, 4×5, 4×9, 5×7
II	9	4, 9, 4×6, 4×7, 6×7, 6×8, 6×9, 7×8, 8×9
III	8	5, 8, 3×5, 3×8, 4×8, 5×6, 5×9, 7×9
IV	4	1×2, 2×3, 2×4, 2×6
V	4	2×5, 2×7, 2×8, 2×9
VI	1	2
VII	1	3×6
VIII	1	5×8
IX	1	7
		Stress environment
I	12	1, 4, 1×3, 1×4, 1×5, 1×6, 1×7, 2×6, 2×7, 3×4, 3×5, 4×9
II	10	6, 7, 3×6, 3×7, 3×8, 4×7, 5×7, 6×7, 6×8, 7×8
III	9	8, 9, 3×9, 4×5, 4×8, 5×6, 5×8, 6×9, 8×9
IV	6	1×8, 1×9, 2×4, 2×5, 2×8, 2×9
V	4	5, 4×6, 5×9, 7×9
VI	2	1×2, 2×3
VII	1	2
VIII	1	3

Parents : 1. WL711, 2. NP 846, 3. WG 377, 4. HD 1981, 5. UP 262, 6. HD 1925, 7. HD 2122, 8. Raj 821, 9. Sonalika.

On the basis of degree of divergence (D^2), 45 populations could be grouped into 9 and 8 clusters in normal and stress environments, respectively (Table 1). Clusters I and II were the largest and included 5 parents and 20 hybrids in normal environment and 4 parents and 18 hybrids in stress environment. Rest 4 parents and 16 hybrids, and 5 parents and 18 hybrids scattered over 7 and 6 groups in normal and stress environments, respectively, mainly added to the divergence. This corroborated the findings of Srivastava and Arunachalam (1977), Behl and Singh (1986), that only few hybrids could add substantial variation in the population. Groups VI, VII, VIII and IX in normal and VII and VIII in stress environment were monogenotypic and comprised of genotypes which were extraordinary for one or more characters. In general, intracluster distances (Table 2) were almost equal and lower than the intercluster distances in both the environments. Therefore, the genotypes included within a cluster tended to diverge less from each other possibly due to large similarity in parentage or selection of genotypes. The intercluster distances varied from 6.65 between group III and VIII to 20.69 between VI and IX in normal environment and from 6.06 between III and V to 18.78 between III and VII in stress environment. On overall basis, clustering pattern over the environments showed only 45 per cent similarity. Such inconsistencies for clustering pattern in different environments have also been

Table 2. Intra- and intercluster distance ($D = \sqrt{D^2}$) in normal and stress environments

Cluster	I	II	III	IV	V	VI	VII	VIII	IX
				Normal environment					
I	6.53	8.26	7.52	11.81	12.80	16.61	8.49	9.52	9.30
II		5.36	6.96	15.00	14.66	19.91	10.91	6.69	7.89
III			5.55	11.96	11.24	16.53	7.41	6.65	9.68
IV				5.49	7.05	7.89	9.79	13.42	16.29
V					5.54	8.78	9.66	12.14	15.92
VI						0	14.58	17.98	20.69
VII							0	11.15	10.88
VIII								0	11.49
IX									0
				Stress environment					
I	6.46	9.38	9.82	8.76	8.92	12.66	12.99	10.05	
II		6.19	8.01	12.97	7.72	17.84	18.03	11.25	
III			5.24	11.67	6.06	18.06	18.78	14.71	
IV				4.95	11.25	8.34	11.69	13.87	
V					6.06	16.68	16.75	14.37	
VI						3.57	8.31	16.01	
VII							0	17.41	
VIII								0	

reported earlier (Somayajulu et al 1970; Jatasra and Paroda 1978). The clustering pattern of hybrids is known to be influenced by the parentage affinity between the parents and progeny (Chaudhary and Singh 1975). In this regard, mainly three grouping trends were evident in the present study. Out of 36 hybrids, 17 and 16 were grouped in different clusters than both the parents in normal and stress environments, respectively. Such a grouping behaviour of F_1 hybrids may be explained on the basis of genic interactions among parents. In other 17 cases in normal as well as stress environment, hybrids were found to be grouped with one of the parent which showed dominance in its favour. In rest 2 and 3 cases in normal and stress environments, respectively, F_1s and their parents were clustered together. Such cross combinations exhibited relatively low genetic divergence among parents.

Per cent heterosis (over mid-parent) ranged from -30.2 to 16.7 for NT, -13.8 to 31.2 for NG, -5.4 to 13.2 for GW and -31.9 to 36.9 for GY in normal environment. Likewise, in stress environment such a range was from -29.0 to 23.8 for NT, -26.8 to 22.1 for NG, -9.7 to 16.5 for GW and -35.4 to 56.4 for GY. Highest heterosis 36.9 and 56.4 per cent for GY was expressed by hybrids WL 711 × HD 2122 and UP 262 × HD 2122 in normal and stress environments, respectively. Out of 36 hybrids, only 3, 10, 16 and 13 in normal environment and 1, 7, 16 and 11 in stress environment showed significant heterosis for NT, NG, GW and GY, respectively. Majority of hybrids, exhibiting significant heterosis, manifested negative heterosis and narrow range for NT in both the environmets. Heterosis for GY, therefore, could be attributed mainly to GW and/or NG. Intercluster hybrids were more frequent among hybrids with significant heterosis though the expression of heterosis was better in intracluster hybrids. Lack of heterosis in highly divergent intercluster hybrids may be attributed to unfavourable gene interactions or intercancellation of favourable gene effects, whereas high heterosis in intra-/intercluster hybrids with medium to low genetic divergence may be explained on the basis of complementary gene effects.

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Scheduling irrigation in wheat through leaf water potential

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Introduction

Observations indicate that crop growth and yield are determined directly by the plant water stress and only indirectly and partially by the soil water status (Kramer 1969). Therefore, warning stress for irrigation should come from the crops themselves. Leaf water potential (LWP) represents the integrated effect of soil water deficit, atmospheric evaporative demand, rooting pattern and crop susceptibility characteristics (Cary and Wright 1971; Clark and Hiler 1973; Hiler and Clark 1971; Hiler et al 1974; Teare et al 1974). The concept of stress day index (Hiler and Clark 1971) was suggested to detect crop water stress for optimizing irrigation timings. It is numerically expressed as the product of the crop susceptibility factor (CS) and the stress day factor (SD). CS indicates the plant's susceptibility to a given water deficit at different growth stages and depends on the crop species and growth stages of the given crop. Stress day factor is a measure of the degree and duration of plant water deficit and can be characterised from the measurement of leaf water potential. The objective of this investigation was to evaluate the effectiveness of stress day criteria using LWP for scheduling irrigation to wheat (*Triticum aestivum* L.) crop.

Materials and Methods

The experiment was conducted in Haldi Loam soil series at the Crop Research Centre, Pantnagar, India. The soils are moderately well drained and water table fluctuated from 1.8 to 2.3m during the wheat season. The ground water was slowly flowing and free from hazardious chemicals (EC$<$0.4ds/m). The crop susceptibility (CS) factor for wheat (RR-21) was determined experimentally as fractional reduction in grain yield resulting from not irrigating the crop at a particular growth stage and irrigating at all other stages during the wheat growing seasons of 1980-81 and 1981-82 under field conditions in the above mentioned soil series. CS for growth stage i was expressed as (Hiler and Clark 1971).

$$CSi = (X-Xi)/X$$

Where X is the grain yield from the treatment irrigated at all growth stages and Xi is the yield in the treatment subjected to water deficit only in the growth stage i. Crop suscep-

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tibility (CS) as determined from the two years experiment was averaged for each growth stage (Table 1).

For getting a range of irrigation schedules five values of morning leaf water potential (SD^1) were chosen as 4, 6, 8, 10 and 12 bars ranging from those prevailing at Crown root initiation (CRI) to milk stages (as per previous observation). At these chosen values of SD^1 and experimentally determined values of CS, stress day factor (SD) at irrigation was determined (Hiler et al 1974). Irrigation schedules thus prepared (Table 2) were executed. The experiment was laid out in 6×5m plots with six treatments with four replicates in 1982-83. In next year (1983-84) most promising treatment of 1982-83 was taken alongwith three treatments (Table 2) based on existing growth stage concept to compare and confirm the previous years' findings.

Table 1. Crop susceptibility (CS) factor for wheat

S. NO.	Growth stages	CS factor
1.	Crown root initiation (CRI)	0.20
2.	Late tillering (LT)	0.10
3.	Late jointing (LJ)	0.10
4.	Flowering (F)	0.10
5.	Milk (M)	0.045

Table 2. Irrigation schedules based on leaf water potential concept

Treatments	SD^1 (Assumed SD) bar	Absolute value of morning LWP (bar) at different growth stages				
		CRI	LT	LJ	F	M
1982-83						
T1 (well watered)	–	Irrig.	Irrig.	Irrig.	Irrig.	Irrig.
T2	4	2.0	4.5	4.5	4.5	9.5
T3	6	3.5	6.5	6.5	6.5	14.5
T4	8	4.5	8.5	8.5	8.5	19.5
T5	10	5.5	11.0	11.0	11.0	24.0
T6	12	6.5	13.0	13.0	13.0	29.0
1983-84						
T0 (Rainfed)	–	–	–	–	–	–
T1 (well watered)	–	Irrig.	Irrig.	Irrig.	Irrig.	Irrig.
T2	–	Irrig.	–	Irrig.	Irrig.	
T3	6	3.5	6.5	6.5	6.5	14.5

These four treatments with four replication were also laid out in 6×5m plots. The SDI treatment amounted to avariable leaf water potential (LWP) characterized from the measurement of morning LWP (30 minutes after sunrise) by a pressure chamber (Scholander et al 1965). Irrigations were applied when the absolute value of morning LWP (SD) approached the specified value for that growth stage. Each irrigation amounted to 6 cm, measured with a parshall flume. Final grain yield and the total amount of water received by each treatment was determined during both the years.

Results and Discussion

During 1982-83 the specified values of morning LWP (SD) at irrigation (Table 2) approached in T2, T3 and T4 (Fig. 1a) near CRI and therefore received one irrigation alongwith T1. Following criteria, no irrigation was warned at LT and LJ stages. After CRI stage, the irrigation initiation value of LWP was observed near flowering and milkstages in T2 and near flowering in T3. During this period LWP in T4 (irrigated at CRI) was similar to those in T5 and T6. On 96th day after sowing (10 days after F stage), T4 was irrigated accidently when LWP was −9.0 bar. Similarly at milk stage T5 received irrigation alongwith T1 although it did not show the specified value of LWP. Grain yields (Table 3) were significantly higher in T3 receiving two irrigations near CRI and flowering stages. Thus, T3 performed best in terms of yield and water use efficiency. Yield reduction in T1 and T2 were due to severe lodging following the irrigation near milk stage. In fact high speed winds prevailed after this irrigation during this year and coused severe lodging in light textured soils.

As per 1983-84 schedule, T3 ($SD^1 = 6$ bar) approached the specified values of morning LWP on 27 and 93 days after sowing and therefore received two irrigations near CRI and F stages (Fig. 1b). T1 and T2 were irrigated at their respective growth stages. Grain yield

Table 3. Effect of irrigation schedules, based on LWP concept, on wheat yield and water applied

Treatments	1982-83		Treatments	1983-84	
	Yield q/ha	Irrigation + rainfall cm		Yield	Irrigation + rainfall cm
T1	51.5	42.1	T0	31.4	19.7
T2	54.5	30.1	T1	56.9	49.7
T3	57.7	24.1	T2	55.2	37.7
T4	49.3	24.1	T3	54.9	31.7
T5	43.3	18.1			
T6	37.2	12.1			
CD at 5%	2.6			3.4	

in T3 (Table 3) found statistically at par to those in T1 and T2. Results reveal that more than two irrigations (near CRI and F stages) did not show any beneficial effect on grain yield. Thus the results of 1983-84 confirm the irrigation schedule (SD^1 = 6bar) found optimum in 1982-83.

Above results reveal that for deciding SD at irrigation time SD^1 should be 6 bar for wheat crop grown in soil with water table similar to Haldi series if morning LWP is the indicator of stress day factor. It may be concluded that stress day index concept using morning leaf water potential can be used to warn the irrigation timings in wheat.

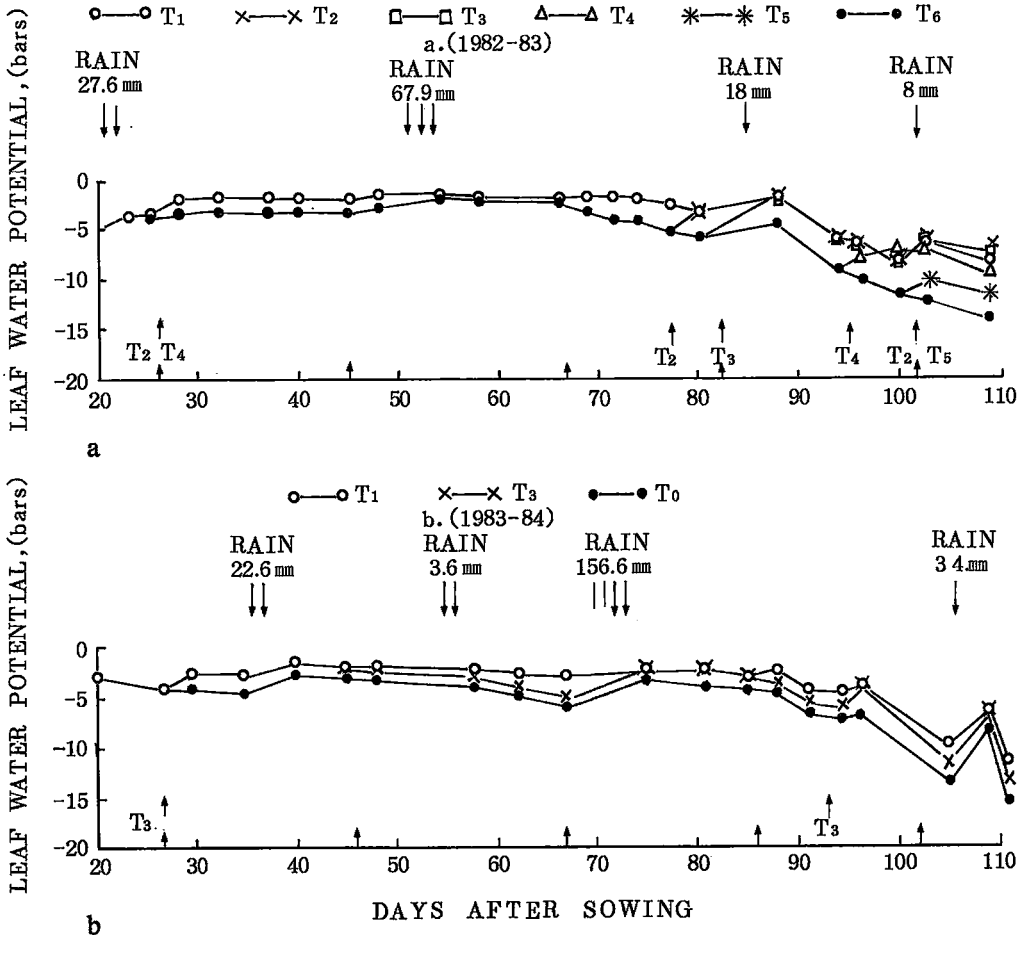

Fig. 1. Seasonal variations of leaf water potential (LWP) during 0730-0800h at different irrigation schedules. a: 1982-1983, b: 1983-1984. Arrows on the X-axis indicate irrigation dates in well watered (T_1) and above the X-axis in the treatments mentioned.

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Induced variability and selection for yield components in bread wheat

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Introduction

Mutagenesis is a potent tool for inducing variability and offers an opportunity for selection for yield contributing characters. Attempts have been made from the early days of mutation breeding to use this technique for increasing crop yield (Gustafsson 1954; Gregory 1955, 1956; Gaul 1964).

It is well known that in the case of wheat, seed number per spike and seed weight show a negative correlation (Sikka and Jain, 1958; Gandhi et al., 1964; Paroda and Joshi, 1970; Knott and Talukdar, 1971). An effective screening technique to break the negative correlation would be to select for these two components simultaneously. If an improvement in one component could be brought about without adverse effect on the other, it may be possible to improve the yield. In this paper a possible approach is suggested which may prove usefulness in the development of selection procedure.

Materials and Methods

Two cultivars, Sonalika and Arjun, were included in this study. The seeds were treated with hydroxylamine (HA), nitrosomethyl-urathane (NMU) and gamma rays as shown in Table 1. Treated seeds were sown close to each other in order to discourage side tillering. Normal looking plants were harvested in M_1.

In M_2 generation, 200 individual plant progenies were sown from each mutagenic treatment. Twenty-five plant progenies of each cultivar were sown as control. The control progenies were repeated after every 24 treated progenies. Each progeny was sown as a single row of 6m with a spacing of 30cm between rows and 10cm between seeds. For recording observations on quantitative characters, M_2 progenies with uniform stand were considered.

Table 1. Details of mutagenic treatments to seeds

Treatment	No. of seeds treated	Duration of presoaking	Conc./dose	pH
Hydroxylamine	1000	7h	0.04M	7.0
Nitrosomethyl-urathane	1000	7h	0.03%	-
Gamma rays	1000	Dry	26KR	-
Control	1000	-	-	-

Fifteen normal-looking competetive plants were selected and observations on the number of seeds per spike, 100 seed weight, number of tillers per plant and single plant yield were recorded. Family as well as population means and variances were worked out.

For screening of polygenic mutations, two characters, number of seeds per spike and 100 seed weight were, taken into consideration. The families showing significantly high variance ratio were taken as segregating. Attention was focussed on those segregating M_2 families in which some of the scored plants showed an improvement in seed weight with no adverse effect on number of seeds per spike and vice-versa. Selected plants were advanced to next generation. The process of raising and screening was continued in M_3 and M_4 generations. The selected plants within a family of M_4 generation were bulked to raise M_5. One hundred and twenty-one families of Sonalika and sixty-two of Arjun were raised as M_5 in randomized block design with three replications as a single row of 6m. In each case control was provided by non-treated seeds of parental cultivar which has continued to show high degree of uniformity. Observations on number of seeds per spike, 100 seed weight and yield of a meter length of row were recorded and data analysed.

Results and Discussion

In an autogamous crop like wheat induction of variability for yield contributing characters is very important for effecting selections. In this context mutation induction for generating variability assumes great importance. The response of two genotypes subjected to mutagenic treatments on population mean, inter-family range for mean and coefficient of variation for four quantitative characters in M_2 is shown in Table 2. It is clear that the two genotypes responded favourably by generating large variability for the quantitative characters.

In general all the mutagenic treatments showed slight reduction in mean for all the four quantitative characters, but the extent of reduction in the mean for two characters, number of tillers per plant and single plant yield, was greater. The reduction in the means may be due to greater number of mutations in negative direction (Gaul 1965; Borojevic and Borojevic 1968; Galal et al 1974).

Table 2. Inter-family range for mean in M_5

Cultivar	Family	Seeds/spike	100 seed wt. (g)	Yield of 1m row (g)
Sonalika	Mutant	32.8-45.7	4.96-6.00	99.66-146.00
Sonalika	Control	40.8	5.30	111.60
Arjun	Mutant	41.4-54.7	3.33-4.10	85.60-143.00
Arjun	Control	47.34	3.66	113.30

Although the population mean showed slight reduction, the observations on the extent of inter-family range for mean clearly reveals that some of the families exceeds the mean of the control family in positive direction. This suggests that at least in these families mutations in positive direction are more frequent. This has obviously offered an opportunity to select individual plants with increase in one of the components without adversely affecting the other for advancing the selected plants to next generation. The process of selection in this way has enabled to effect the improvement in population mean in subsequent generations. The effectiveness of selection is evident from the observations on per cent increase in mean of treated population over the control in subsequent generations (Fig. 1-4). Thus the improvement in the mean for 100 seed weight and seeds per spike can be attributed to the effect of selection of best plants for these two characters from previous generations. This observation is in confirmity with those of Scossiroli (1974) and Borojevic and Borojevic (1968).

The selected plants within an M_4 family were bulked and evaluated in M_5. The analysis of variance revealed that in both the varieties, the families differed significantly for 100 seed weight, seeds per spike and yield of one meter row. Table 3 shows inter-family range for mean in M_5. It is seen that for all the characters, the mutant families exceeded the mean of control on both the directions.

Unit one hundred seed weight, number of seeds per spike and number of effective tillers per unit area are the most important components of yield in wheat. If an increase in one of the yield components is not associated with the decrease in the other, it should lead to the improvement in yield. The present study brings forth this point very convincingly. It has been possible to improve the yield by bringing about improvement in seed weight with no adverse effect on seeds per spike and vice-versa of most of the mutant families. It is clear from Table 4 that improvement in the yield is mainly due to increase in 100 seed weight since number of seeds per spike remained unaltered. For example, the improvement in the yield of the mutant families S-5, S-17, S-19, S-29 and S-56 of the cultivar Sonalika and mutant families A-3, A-25 and A-41 of the cultivar Arjun is due to increase in 100 seed weight with no loss in seeds per spike. The improvement in the yield of the mutant family A-62 of the cultivar Arjun is due to increase in number of seeds per spike with no loss in 100 seed weight. The improvement in the yield of the mutant families not showing significant change in seed weight or seed number can perhaps be attributed to the cumulative effect of small increase in yield contributing characters. It may be mentioned that although the observations on number of tillers per m^2 were not recorded, it can be inferred that the mutant families showing improvement in seed weight with no adverse effect on seed number per spike and vice versa must have at least comparable tillering capacity to that of respective control.

Efforts have been made to bring about yield improvement by selecting for one of the components of yield, either seed weight (Knott and Talukdar 1971) or seeds per spike (Borojevic and Borojevic 1968; Szamak 1973), but this methodology did not succeed as the selection for one of the two components resulted in the reduction in the other because of

Table 3. Population mean, inter-family range for mean and coefficient of variation in M_2

Treatment	Seeds/spike			100-seed weight			Tillers/plant			Single plant yield		
	Mean	Range	C. V.	Mean	Range	C. V.	Mean	Range	C. V.	Mean	Range	C. V.
Sonalika												
HA	38.79	27.07-43.13	18.27	5.45	5.12-6.12	10.71	11.32	7.81-17.08	42.23	17.86	11.95-29.95	45.10
NMU	38.54	29.07-52.27	20.33	5.56	4.60-6.35	10.15	12.55	6.53-21.33	46.01	19.82	13.33-30.56	49.45
γ-rays	37.18	26.00-55.66	21.99	5.53	5.15-6.32	9.56	12.55	7.60-18.40	50.59	18.26	9.10-30.79	53.58
Control	38.78	33.71-44.50	15.30	5.55	5.22-5.73	9.03	13.54	11.08-16.04	40.47	20.09	16.00-23.27	42.23
Arjun												
HA	47.73	41.60-61.54	16.52	4.01	3.30-4.75	8.97	13.10	7.87-20.53	43.10	19.24	9.96-30.49	46.41
NMU	48.00	42.00-61.16	17.58	4.08	3.36-4.68	10.93	13.81	8.86-21.00	46.29	18.84	10.00-29.65	49.80
γ-rays	47.89	36.60-60.50	20.16	4.06	3.41-4.61	13.84	14.02	7.80-25.59	45.47	18.33	8.64-31.55	54.77
Control	48.02	38.93-56.53	15.24	4.03	3.36-4.35	8.43	14.58	11.67-15.71	41.80	19.32	13.13-21.00	40.22

negative correlation between these components. It is in this context the methodology of selecting simultaneously for both the components, as explained earlier, adopted in the present investigation appears to be an effective approach for breaking the negative correlation between these two components which eventually lead to improvement in the yield. The study thus reveals that induced variability can be made use for selecting the plants with

Table 4. Observations on families showing improvement in yield

Family	Seeds/spike	100 seed wt. (g)	Yield/1m row (g)
Sonalika			
S-5	40.26	5.59*	136.33**
S-17	37.53	5.66**	146.00**
S-19	38.80	5.59*	133.16**
24	38.20	5.37	144.16**
S-29	44.66	5.61*	143.50**
33	40.86	5.46	139.00**
51	43.86	5.50	129.80*
S-56	41.03	5.59*	138.50**
58	39.19	5.40	134.00**
69	37.66	5.02	133.50**
101	40.93	5.30	133.00**
Control	40.80	5.30	111.60
CD @ 5%	4.23	0.28	15.50
CD @ 1%	5.66	0.37	20.72
Arjun			
A-3	47.29	3.90**	143.50**
10	48.40	3.60	135.50*
11	49.00	3.80	137.83*
A-25	49.49	3.90**	135.33**
30	47.66	3.73	135.00*
A-41	49.26	4.13**	140.00**
62	52.40*	3.46	135.03**
Control	47.33	3.60	113.30
CD @ 5%	4.75	0.21	20.89
CD @ 1%	6.28	0.28	27.61

* Significant at 5%

** Significant at 1%

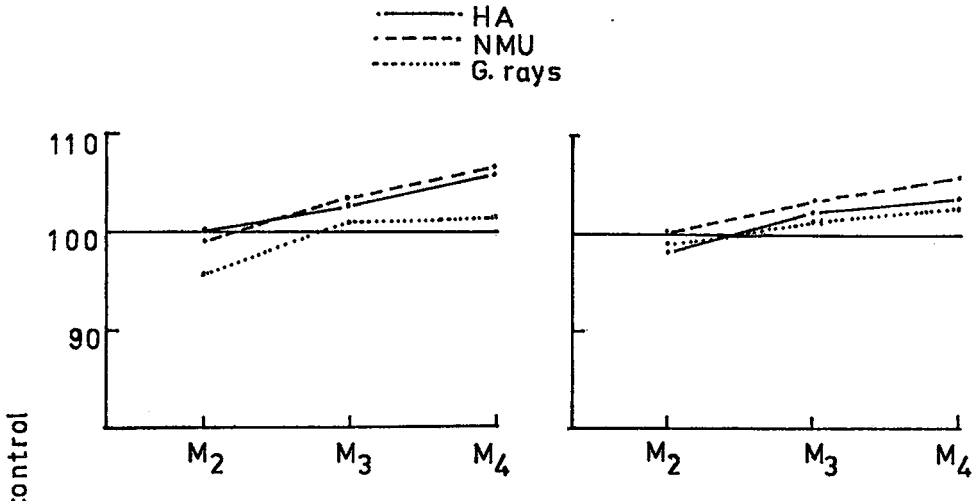

Fig. 1. No. of seeds / spike

Fig. 2. 100 Seed weight

Var. SONALIKA

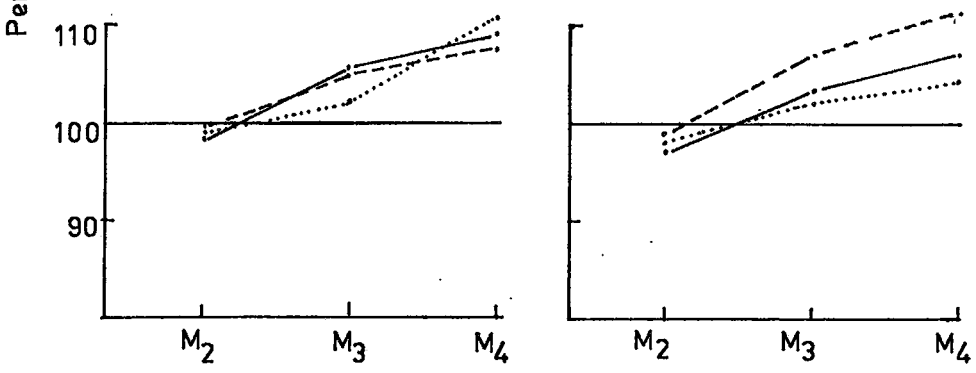

Fig. 3. No. of seeds / spike

Fig. 4. 100 seed weight

Var. ARJUN

Fig. 1-4: Mean of treated population in per cent of control in M_2-M_4

Fig. 1. No. of seeds/spike
Fig. 2. 100 Seed weight
Fig. 3. No. of seeds/spike
Fig. 4. 100 seed weight

increase in one of the components with no adverse effect on the other. Although far reaching conclusions cannot be drawn at this stage, nevertheless the study clearly suggests that the methodology of simultaneous selection for two characters is an effective way to overcome the negative correlations between seed number per spike and 100 seed weight which act as a barrier in the improvement of yield.

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II Genetic Stocks

List of *Cs1* and *Cs2* carrier in tetraploid wheats

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Cs chlorosis is found in hybrids between the Emmer and the Timopheevi group of tetraploid wheats (Tsunewaki and Hamada 1968, Tsunewaki and Nakai 1973, Kawahara 1985, 1991). The following list summarize the observations carried out by the author from 1982 to 1989. Passport data of these strains are listed in the catalogue of the Plant Germ-plasm Institute, Kyoto University (Tanaka 1983).

Triticum dicoccoides (wild Emmer)

cs1 carrier (*cs1cs2*);

108-1, 108-2, 108-3, 108-5, 109, 110, 195, 198, 1921, 1945, 1947, 1948, 1949, 1951, 1952, 1953, 1955, 1957, 1959A, 1959B, 1972B, 1974, 1976B, 1978B, 1991, 8536, 8537, 8538, 8539, 8541, 8736A, 8736B, 8737, 8804, 8805, 8806, 8807, 8808, 8809, 8810, 8811, 8812, 8814, 8815, 8816A, 8816B, 8817, 8821A, 8821C, 8915A, 8915B, 8935, 8937B, 8941, 8942, 8943.

Cultivated Emmer wheats

cs1 carrier (*cs1cs2*);

111, 112, 124, 125, 126, 127, 128-1, 134, 136, 137, 138, 139-1, 139-2, 140, 141, 142, 143, 145, 146, 147, 148, 185, 186, 187.

Cs1 carrier (*Cs1cs2*):

123, 144, 190-1.

T. araraticum (wild Timopheevi)

cs2 carrier (*cs1cs2*);

196-1, 1986, 1988, 8451, 8456, 8460, 8461, 8465, 8470, 8471, 8476, 8479, 8481, 8489, 8490, 8495, 8498, 8500, 8506, 8511, 8520, 8528A, 8543, 8546, 8549, 8551, 8561, 8564, 8570, 8593, 8600, 8602, 8617, 8623, 8625, 8632, 8640, 8646, 8656, 8658, 8668, 8674, 8675, 8678, 8682, 8685, 8691, 8718A, 8719, 8720, 8733, 8779, 8781, 8797, 8799A, 8802, 8818, 8819, 8820, 8821B, 8822, 8824A, 8825, 8826, 8828, 8831, 8857, 8866, 8867, 8869, 8874, 8877, 8880, 8882, 8884, 8887, 8890, 8892, 8912, 8917, 8924, 8928, 8933, 8934, 8938, 8939, 8940.

Cs2w carrier (*cs1Cs2w*);

1926A, 1930, 1933, 1936, 8597, 8609, 8620, 8662, 8673, 8697, 8700, 8702, 8707, 8710, 8723, 8735, 8740, 8742, 8758, 8761, 8763, 8784, 8793, 8907, 8944, 8945, 8946, 8947, 8948.

Cs2m carrier (*cs1Cs2m*);

196-2, 1901, 1902, 1903, 1904, 1905, 1906, 1911, 1913, 1914, 1923, 1927, 1938, 1943, 1944, 1960, 1964, 1967, 1972, 1976A, 1978, 1980A, 1982, 1984A, 1992, 8713, 8715, 8725, 8727, 8732, 8766, 8908.

Cs2s carrier (*cs1Cs2s*);

1907A, 1907B, 1908A, 1908B, 1909A, 1909B, 1909C.

T. timopheevi (cultivated Timopheevi)

Cs2w carrier (*cs1Cs2w*);

107-1, 107-3, 107-4, 1818, 1819, 1820, 1821.

Cs2m carrier (*cs1Cs2m*);

107-2, 107-5.

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Number 72: 60 – 63 (1991)

III Records

Proceedings of the 22th Wheat Genetics Symposium of Japan

The 22th wheat Genetics Symposium of Japan was held on December 15 and 16, 1990 at Nara, Japan, chaired by Dr. T. R. Endo. The followings are the proceedings of the presentations. Addition to the reports on wheat studies, Dr. M. Hori and Dr. H. Ogura presented recent topics of human genetics and rice breeding, respectively.

Chromosome mapping by use of aneuploids in wheat

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The term, chromosome map includes genetic map and cytological or cytogenetical map. Cytological mapping which is referred to as physical mapping in the broad sense is the subject of the Dr. Y. Mukai, the second speaker in this symposium. Dr. K. Tsunewaki, the third speaker will talk about RFLP mapping. So I will confine myself to the method of genetic mapping of morphological and isozyme markers by use of aneuploids and discussion of some problems accompanied. McIntosh (1987) described principle and method for gene location and gene mapping in hexaploid wheat. McIntosh and Cusick (1987) presented linkage map of hexaploid wheat. Nomenclature for description of wheat aneuploids was presented by Kimber and Sears (1968). With the rather few good markers available, genetic mapping in wheat has not been well developed. So some data reported in barley and cotton will be cited for supplement.

1. Hyperaneuploids

a) Trisomic: Primary trisomic had been used to determine the gene-chromosome association. Trisomic analysis is generally less efficient than monosomic analysis, and largely applied to diploid plants, which is hard to withstand hypoaneuploidy. Determination of genotypes of F_2 plants with dominant character makes it possible to estimate the recombination value, but this is not realistic, because of too much time and labor to be required.
b) Monotelotrisomic (telotrisomic): Instead of primary trisomic, monotelotrisomic has been used for determining the centromere position in diploid plants such as barley. In Fig. 1, the locus A nearby and the locus B far from the centromere on the arm homologous to telocentric would show chromosome segregation and random chromatid segregation depending on respective distance from the centromere, while the locus C on the opposite arm would show disomic segregation. Thus by monotelotrisomic analysis, centromere can be

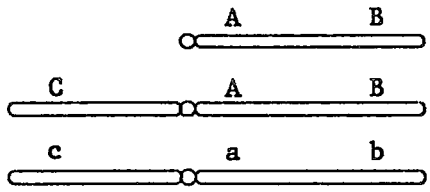

Fig. 1. Monotelotrisomic with three marker genes, A, B and C (Tsuchiya and Singh 1982, modified)

positioned between markers when a considerable number of markers have been mapped on a give chromosome. This had been done by Tsuchiya and his coworkers in barley (Tsuchiya and Singh 1982). Similarly, monoacrotrisomic (acrotrisomic) analysis can provide information on the position of break point of the chromosome concerned (Tsuchiya *et al.* 1984).
c) Monoisodisomic: Monoisodisomic carries three doses of loci locating on the isosome. In cotton recombination value of 16.2% between centromere and a marker, *M1* was obtained in monotelodisomic for chromosome 4S, while the value of 21.7% in monoisodisomic 4S (Endrizzi and Bray 1980). The significantly higher value in monoisodisomic was attributed to chiasma between two isoarms followed by a chiasma formation between a complete chromosome and one arm of isosome at the proximal region.

2. Hypoaneuploid

a) Monotelodisomic: Sears (1966) first reported gene-centromere distance with the aid of telocentrics in wheat. Neatby's virescent gene (*v*) on the short arm of chromosome 3B was located at 0.28 crossover unit from the centromere. And the loci on the long arm of 6B, *Sr11*, *Ki*, and *B2* were 45.1%, 41.3% and 0.44% apart from the centromere, respectively. A dominant marker on the monosomic arm of complete chromosome in montelodisomic, *Co* in the case of Dr. Sears, marks transmission of the complete chromosome to the offspring and can more or less save cytological determination of chromosome complement. In comparison with disomic or conventional analysis, reduction of amout of crossing over in the centromere region is a problem that deserves careful attention. Sears (1972) investigated crossover values in the heterozygote both for the markers (*Sr11* and *B2*) on the long arm and for *umbellulata* segment substituted for most of the short arm of 6B, but the remaining segment being still enough to make the pairing conditions different from a monotelodisomic for the long arm of 6B. The recombination value of 3.5% was obtained in the region from *B2* to the proximal end of *umbellulata* segment across the centromere, which indicates four fold increase in frequency of crossing-over in comparison with those obtained by using the respective telosomes. Endrizzi and Kohel (1966) had reported decrease in recombination value comparable to Sears as well as compensatory increase in recombination value in monotelodisomic cotton.

Nishikawa et al (1974) reported recombination values of two complementary genes for progressive necrosis with the centromere, 10.5% for *Ne1* and 9.4% for *Ne2*, obtained by the telocentric method. The Distance from the centromere of α-amylase isozyme loci on the long arm of three chromosomes of homoeologous gourp 6 were determined as shown in Fig. 2 (Nishikawa et al unpublished). There is no information on shift of recombination frequency in these cases.

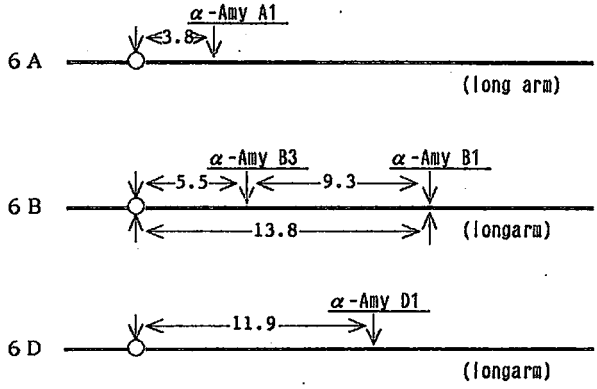

Fig. 2. Gene-centromere distance of α-amylase genes on three chromosomes of homoeologous group 6

b) Doubletelotrisomic: Doubletelotrisomic is as vigorous as disomic and can be used for genetic analysis in diploid as well as polyploid plants. An albino plant (ditelo2BL-monotelo2BS) of *durum* LD 222 occurred from the cross of a green plant of ditelo2BL-monotelo2BS with heterozygous (A/a) doubleditelosomic for 2B, indicates that the albino gene is located on short arm of 2B. Recombination value of 6.0% with the centromere was obtained from progenies of selfed heterozygous (A/a) doubletelotrisomic. Though there seems to be few, if any, paper reporting it, doubletelotrisomic analysis should be recommended for genetic mapping, because it is not only applicable to diploid and polyploid plants, but makes it possible to estimate the recombination value and position of centromere, if the both arms of a chromosome involved are properly marked.

Mase et al (1989) investigated F_2 segregation of the hybrid of doubleditelosomic for chromosome 1 with a multiple marker line (BGN 1008: *1k2, n* on the long arm and *f8* on the short arm, Tsuchiya 1985). The recombination values obtained are shown in Fig. 3 together with those from conventional method. It is obvious that recombination in *n-f8* region is reduced, and that in *1k2-n* region further apart from centromere is increased in the telocentric method as compared with the conventional method. This seems to be a phenomenon that commonly occurs in use of the telocentrics. In contrast to Tsuchiya (1985) the recombination values indicate that gene *f8* is located on the long arm.

Summary

As well known in Chinese Spring wheat all the possible telocentric chromosomes except 7DS

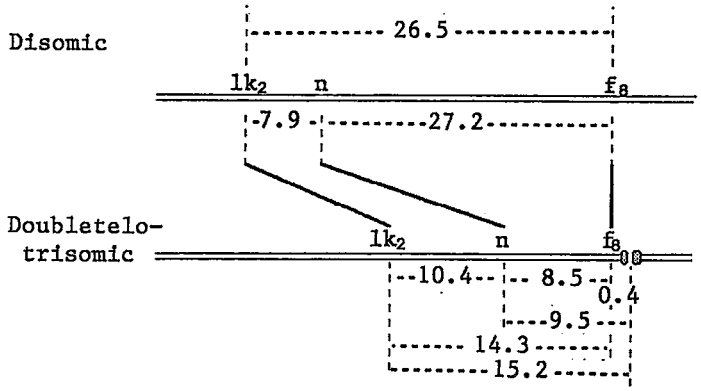

Fig. 3. Genetic map of barley chromosome 1 depicted based on disomic and doubletelotrisomic analysis

(isolated in cv. Canthatch by Dr. Kerber) have been isolated and are ready to use for cytogenetical study. In addition, 14 lines of doubleditelosomic and reasonable number of lines of ditelomonotelosomic are now available in tetraploid wheat (Joppa 1987, Nishikawa unpublished). By use of these aneuploids monotelodisomic analysis and doubletelotrisomic analysis can be easily and efficiently made, which provide the following informations; (1) Gene-chromosome arm association, (2) recombination value between marker and centromere, (3) gene order, and (4) centromere position.

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Physical mapping of wheat genes using *in situ* hybridization method and deletions

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Physical map reflects the accurate location of genes. A cytogenetic map is also a low resolution physical map. Genetic map is derived from the frequency of recombination during meiosis. There are many reports on differences between genetic and physical maps for wheat chromosomes. There are two methods for physical mapping in wheat. One is the use of deletions induced genetically by the gametocidal chromosomes, which was developed in *Triticum aestivum* cv. Chinese Spring by Endo (1988, 1990). If partial chromosome deletions between the centromere and telomere become available, some genes could directly be mapped to the deleted regions. The other is the use of *in situ* hybridization (ISH) technique with DNA probes. While ISH studies have been generally restricted to highly repeated sequences in wheat, in recent years it has been possible to localize single-copy sequences on metaphase chromosomes of human cells (Lichter 1990). In this paper, I will review some recent work in our research group on physical mapping of wheat genes. The present results were carried out with the collaboration of T. Endo, B. S. Gill and M. Yamamoto. This work was financially supported by a grant from the Japan Society for the Promotion of Sciences for the Japan-U.S. Cooperative Science Program.

1. Gross morphology --- *Q*

Using deletions induced by a gametocidal chromosome from *Aegilops longissima,* the speltoid-suppression gene *Q* was first mapped in the distal 46% of the long arm of chromosome 5A (Endo and Mukai 1988). This locus was further narrowed down to the distal 13% of the 5A long arm (Tsujimoto and Noda 1990).

2. Male sterility --- *ms1*

A male sterile mutant of common wheat, "Cornerstone" is known to be located on the short arm of chromosome 4B (Discoll 1975). The ISH analysis using repeated DNA sequences probe (pSc119) revealed that this cultivar lost half of the terminal ISH site on the short arm of chromosome 4B. A terminal deletion in the short arm of chromosome 4B of Chinese Spring which lacked the distal 13% of the short arm including a terminal large ISH site, was completely male sterile in the homozygote (Endo et al 1991). Thus, a gene controlling male fertility is located in the deleted terminal region.

3. Meiotic characters (Pairing homoeologous) --- *Ph1*

ISH analysis of a Ph mutant of Chinese Spring and duplication/deficiency stocks of chromosome 5B of *durum* demonstrated that the *Ph* gene was found to be tightly linked to the ISH site. The site was physically mapped at positon 0.39FL (fraction length, the frac-

tion of the total arm length from the centromere) on the long arm of chromosome 5B.

4. Nucleolus organizer (18S-5.8S-26SrRNA) - - - *Nor*

Direct evidence on the physical location of rRNA within *Nor* has been obtained from ISH experiments. Appeles et al (1980) found 18S-26SrRNA sites on chromosomes 1B (*Nor-B1*), 6B (*Nor-B2*) and 5D (*Nor-D3*). Recently, Mukai et al (1991) reported a new locus at position 0.76FL of the chromosome 7D long arm (*Nor-D4*), and confirmed the *Nor* locus in the short arm of chromosome 1A at the telomeric end (*Nor-A1*).

5. Pathogenetic disease/pest reaction

Reaction to *Mayetiola destructor* - - - *H*

ISH using total donor species genomic DNA probes can be used as a generalized method for the detection of donor chromatin including a target marker gene in genomes of recipient species. A tiny segment of rye chromatin in wheat specifying resistance to Hessian fly was inserted into the proximal region of the long arm of chromosome 4A (Mukai et al submitted).

6. Enzymes - - - *α-Amy*

A genomic α-amylase DNA clone cross-hybridized to several chromosomes of Chinese Spring wheat under chromosomal *in situ* suppression (CISS) hybridization conditions by using competitor DNA (total wheat DNA). The *α-Amy* loci were physically mapped at two interstitial locations in the long arm of chromosome 6B (0.21FL and 0.69FL). These loci may correspond to *α-Amy3* and *α-AmyB1*. The *α-Amy3* and *α-AmyB1* genes on chromosome 6BL were mapped genetically by Nishikawa (personal communication) to 5.5 and 13.7 crossing-over units from the centromere, respectively.

7. Endosperm strage proteins - - - *Gli*

A genomic clone containing a wheat γ-gliadin gene was used as a probe. Hybridization sites were detected on chromosomes 1B, 1D, 3B, 5B and 6B of Chinese Spring wheat. Chromosone pairs 1B and 1D had terminal singals in the short arm. However, the gliadins are well known to be controlled by genes on the short arms of the chromosomes of homoeologous groups 1 and 6.

8. Restores for cytoplasmic male sterility - - - *Rfv1*

A fertility-restoring gene, *Rfv1* against *Ae. kotschyi* cytoplasm is known to locate on the short arm of chromosome 1B of Chinese Spring (Mukai and Tsunewaki 1979). From the telocentric mapping, the distance between the *Rfv1* gene and the centromere is estimated to be 34 cM (Hamawaki and Mukai 1980). Many male sterile plants were found in the offspring of a cross, Chinese Spring with *Ae. kotschyi* cytoplasm x an alien addition line of Chinese Spring having an *Ae. cylindrica* chromosome. We isolated an array of deletions in the short arm of chromosome 1B. The *Rfv1* locus was physically mapped at position about one third of the satellite length from the centromere (Mukai et al in preparation).

9. 5S ribosomal RNA - - - *5S-Rrna*

In Chinese Spring wheat twelve 5S rRNA loci were physically mapped on chromosomes of homoeologous groups 1 and 5 (Mukai et al 1990). The FL location of 5S rRNA loci was 0.77 in 1AS, 0.96 in 1DS, 0.76 in 5AS, 0.63 in 5BS and 0.64 in 5DS. In the 1BS arm, the locus was mapped at the midpositon (0.5) of the satellite length.

10. Waxiness - - - *W1*

In *T. aestivum* strain Salmon, a segment of chromosome 2B carring a waxy locus *W1* is deleted (Tsunewaki 1964). In most strains of *T. aestivum* ISH patterns of pSc 119 probe revealed that chromosome 2B short arm contained one major terminal site, whereas Salmon lost half of a terminal ISH site. Three deletions in chromosome 2B short arm of Chinese Spring were deficient in the *W1* locus (Endo et al unpublished). Thus, we can map the *W1* locus on the distal end of chromosome 2B short arm.

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RFLP analysis of common wheat and its ancestors

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This paper reviews the results of the works on restriction fragment length polymorphism (RFLP) analysis of common wheat and its ancestors, which have been carried out last few years in our laboratory.

(1) Genomic DNA library construction and characterization of the clones (Liu et al 1990): We constructed the *Eco*RI, *Hin*dIII and *Pst*I fragment libraries of the genomic DNA of Chinese Spring wheat (CS). In all cases, fragments of 0.5 - 2.0 kbp in size were cloned using a vector pUC119. Numbers of the clones obtained were 143, 180 and 323 for the *Eco*RI, *Hin*dIII and *Pst*I fragments, respectively. Copy number per haploid nucleus was estimated for the *Eco*RI and *Hin*dIII fragment clones, of which 49%, 31%,14% and 6% were estimated to be ca. 10^1, 10^2, 10^3 and 10^4 copies, respectively. The fragments of the first class were considered the unique sequence, and used in the foregoing RFLP analysis.

(2) Genetic mapping of the RFLP sites by linkage analysis and nullitetrasomic analysis (Liu 1991): As is described in the next section, CS showed the largest RFLP to *Triticum spelta* var. *duhamelianum* (Spelta) among other seven common wheats tested. Based on this,F_2 hybrids were produced from a cross between these two wheats, and the nuclear DNAs extracted from the bulked F_3 seedlings from each of 66 F_2 plants were subjected to the linkage analysis of RFLP sites, using the unique sequence clones as probes. In total 197 RFLP loci were detected between the two parents, and their linkage relationships were analyzed. These loci and the *Q* locus were grouped into 31 linkage groups, with 10 solitary loci.

All but 20 loci showed normal Mendelian segregation. Of the 20 exceptional loci, seven showed preferential transmission of the Spelta alleles, and another seven showed preferential transmission of the CS alleles, the remaining six showed abundance (four loci) or shortage (two loci) of the heterozygote. A few representative loci of each linkage group and all solitary loci were subjected to the nullitetrasomic analysis, and they were successfully assigned to either of the 21 wheat chromosomes. In addition, 226 non-RFLP loci were located to the individual chromosomes. Thus, 424 loci in total were allocated to the 21 chromosomes. The present tatal map size amounts to 1835 cM. Number of RFLP loci located in the D genome was 28, that was much smaller than those allocated in the A and B genomes, which were 71 and 97, respectively. On the contrary, number of non-RFLP loci located in the A, B and D genomes was 61, 78 and 85, respectively, which did not differ significantly among the three genomes. Apparently, the degree of RFLP between the D genomes of CS and Spelta was extremely low, comparing to those between their A or B

genomes.

(3) RFLP analysis of eight common wheat and one emmer wheat accession (Liu et al 1990): Using the unique sequences as probes, RFLPs among eight common wheats and an emmer wheat were investigated. The proportion of the clones revealed RFLPs among the eight common wheats was about 41% in the unique sequences, whereas it was only 11% in the moderately repeated sequences (ca. 10^2-10^3 copies/haploid nucleus). No highly repeated sequences exhibited RFLP among the eight common wheats. In total, 271 probe-enzyme combinations were used, 71 combinations (27%) of which revealed RFLPs among the eight common wheats. Based on the total number of hybrid bands observed in each wheat, and the number of differential hybrid bands observed between all wheat accessions, the genetic distances between them are calculated after Nei (1987). The average distance between eight common wheats was 0.71×10^{-2} (range, $0.38-0.935 \times 10^{-2}$), whereas that between them and an emmer wheat was 2.06×10^{-2} (range, $1.86-2.44 \times 10^{-2}$), indicating great nuclear genome differentiation between emmer and common wheats as compared to the differences among common wheats. This is mainly due to the absence of the D genome in emmer wheat. The eight common wheats were classified into three groups, (1) four Asian wheats, (2) three Western wheats, and (3) Spelta. Tibetan semi-wild wheat showed the closest relation to CS, supporting an idea that the former is a recent derivative of the Chinese cultivated wheat (Tsunewaki et al. 1990). Spelta greatly differed from all other common wheats. This and the fact described in the previous section are in favor of the origin of Spelta from the hybridization between emmer and common wheat, as suggested by Schiemann (1951) and Tsunewaki (1968).

(4) RFLP analyses of wild tetraploid wheats, einkorn wheat and *Ae. squarrosa* : RFLPs between large numbers of *T. dicoccoides* and *T. araraticum* accessions, together with a limited numbers of *T. durum, T. timopheevi* and *T. aestivum,* have been carried out by Mori et al (1991). Their results demonstrated that the nuclear genomes of emmer and timopheevi groups of wheat have been greatly differentiated from each other, with no intermediate types, indicating the diphyletic origin of these two tetraploid wheat groups. RFLP analysis of einkorn wheat was carried out by Takumi et al (1991), using seven accessions of *T. boeoticum, T. urartu* and *T. monococcum,* with a single accession each of emmer and common wheat as referants. In total, 88 probe-enzyme combinations were employed. The results clearly indicated that *T. monococcum* was derived from *T. boeoticum,* whereas the A genome of emmer and common wheats was originated from *T. urartu,* fully supporting the results of Dvorak (1988) which were obtained with the repeated sequence clones as probes. RFLP analysis of five *Ae. squarrosa* accessions collected from different locations, with a common wheat as a referant, was carried out by Achiwa (1990), using 81 probe-enzyme combinations. In this case, 27 unique sequence clones of the genomic DNA of *Ae. squarrosa* were used as probes. The accessions collected from the same regions had similar nuclear genomes even though they belonged to different taxonomic

varieties, whereas those of the same variety from different regions showed differentiation of their genomes. The nuclear genomes of *Ae. squarrosa* accessions collected from south to west coastal regions of Caspian Sea showed close relation to that of common wheat, supporting the proposal of several workers (Tsunewaki 1968, and others) that this region is the birthplace of common wheat.

(5) Structure of a hypervariable sequence, TAG 546, and its use in common wheat cultivar identification (Liu and Tsunewaki 1990, Liu et al 1991): A hypervariable sequence was found among the genomic DNA clones of CS, and was designated TAG546. When this clone was used as probe, single enzyme digests of the nuclear DNAs of eight common wheats (ref. section (2)) could be distinctly discriminated from each other. Encouraged by this finding, the nuclear DNAs of 56 common wheat cultivars collected from various countries, some of which are very closely related to each other in their pedigrees, were treated with three 6-base cutters, and their fingerprints probed with TAG546 were compared. All the cultivars could be successfully identified by this fingerprinting.

Copy number of this clone sequence that is 4093 bp in size was estimated to be about ten per haploid nucleus. By nullitetrasomic analysis, these copies were located on the different chromosomes, 5A, 6A, 7A, 2B, 3B, 6B, 7B, 2D, 4D and 5D. Complete base sequence analysis of this clone was worked out, and a transposon-like structure was found within the sequence.

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Field research of wheat, barley and their wild relatives in southern Italy and Greece, 1990

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A field research work was carried out in the most southern part of Italian Peninsula and Sicily Island, Italy and two islands (Karapathos and Naxos) in Aegean Sea, the nothern part of Peloponnisos Peninsula and the area of Ioanina, northwest part of Greece. This mission was the first year's project on the comparative ecological genetics on phylogeny of wheat, barley and their wild relatives in northeastern region of the Mediterranean Sea. This project was sponsored by the Ministry of Education, Science and Culture, Japan (Grant-in-Aid for International Scientific Research Program: Field Research No. 02041037, 1990-1993).

The route and the actual schedule of the present mission is shown in Table 1 and Fig. 1. Table 2 is a summalysed list of the collection. We showed about 80 beautiful color slides showing the agriculture, the geological features, the life of people in addition to the habitats or field and some spikes or plants of wheat, barley and their relatives in the Italy and Greece. The present researchers will visit Yugoslvia as the second year's (1991) plan. After this project completely finish, the whole story will be reported elsewhere.

Table 1. The schedule and route of the present collection trip in Italy, Greece and Yugoslavia in 1990

Date	Route[1)]
Y. Furuta:	
May 19-20	Osaka(JAPAN) — Rome(ITALY)
S. Ohta:	
May 26-27	Osaka(JAPAN) — Rome(ITALY)
Y. Furuta and S. Ohta:	
May 28-29	Rome ═ Bari
29-30	Bari
31	Bari ··· Altamura ··· Sibari ··· Reggio di Calabria = Messina
June 1	Messina ··· Taormina ··· Catania
2	Catania ··· Mt. Etna ··· Adrano ··· Catania
3	Catania ··· Catenanuova ··· Piazza Armenia ··· Caltagirone ··· Siracusa
4	Siracusa ··· Ragusa ··· Licata
5	Licata ··· Canicatti ··· Caltanisseta ··· Cefalú
6	Cefalú ··· Roccapalumba ··· Cammarata
7	Cammarata ··· Agrigento ··· Ribera ··· Piana d. Albanesi
8	Piana d. Albanesi ··· Partinico ··· Trapani ··· Marsala

Table 1. (Continued)

Date	Route[1]
9	Marsala ··· Castelvetrano ··· Palermo ··· S. Stefano ··· Nicosia ··· near C. Forestale (18km N. of Cesarò)
10	near C. Forestale ··· Sant'Agata di Militello ··· Messina = Reggio di Calabria ··· Soverato
11	Soverato ··· Crotone ··· Cosenza ··· Rubo di Puglia
12	Ruvo di Puglia ··· Bari
13	Bari ··· Oppido Luccata ··· Frenza ··· Bari
14-15	Bari
16	Bari ▬ Naples
17-18	Naples
19	Naples ▬ Rome
20	Rome — Athens (GREECE)
21	Athens
22	Athens — Karpathos
23-25	Karpathos Isl.
26	Karpathos — Athens
27	Athens ··· Cape Sounion ··· Athens
28	Athens = Naxos
29-30	Naxos Isl.
30-July 1	Naxos = Athens
July 2	Athens ··· Pirgos ··· Olympia
3	Olympia ··· Tripolis ··· Argos ··· Korinthos ··· Athens
4	Athens ··· Mt.Pendeli (north of Athens) ··· Athens
Y. Furuta:	
July 5-6	Athens — Osaka(JAPAN)
S. Ohta:	
July 5-7	Athens
8	Athens — Ioannina
9	Ioannina ··· Konitsa ··· Tsotili
10	Tsotili ··· Kastoria ··· Psarades (near Lake Prespa) ··· Tsotili
11	Tsotili ··· Gravena ··· Metsovon ··· Ioannina
13	Ioannina — Athens
14	Athens
15-16	Athens ▬ Beograd (YUGOSLAVIA)
17	Beograd ▬ Novi Sad
18-19	Novi Sad
20	Novi Sad ▬ Beograd
21	Beograd ▬ Sarajevo
22	Sarajevo ▬ Mostar ▬ Sarajevo
23	Sarajevo ▬ Beograd
24	Beograd — Athens (GREECE)
25-30	Athens
31-August 1	Athens — Osaka (JAPAN)

1) — : by air, ▬ : by bus or by train, ··· : by car, and = : by ship.

Table 2. A summary of collection in Italy and Greece in 1990

Collected material	No. of samples[1]						Total
	Italy		Greece				
	Pn	Sc	Kp	Nx	Pl	Ml	
Cereals and their wild relatives:							
Triticum durum	22	103	17	2	2	11	157
T. aestivum	14	23	1	4	7	35	84
T. boeoticum						6	6
Aegilops biuncialis			12	8	5	23	48
Ae. caudata			3	9	4		16
Ae. comosa						2	2
Ae. heldreichii					4	4	8
Ae. ovata	13	37			2	6	58
Ae. triaristata	3		1		5	24	33
Ae. triuncialis	3			5	5	27	40
Ae. uniaristata						1	1
Ae. variabilis			2				2
Ae. ventricosa	1						1
natural hybrids involving *Aegilops* spp.		2[2]				1[3]	3
Hordeum vulgare	20	43	1	17	4	16	101
wild *Hordeum* spp.	14	41	4	12		7	78
Secale cereale	1			1		44	46
Triticale						1	1
Avena spp.	48	130	6	15		12	211
rice seed sample				1			1
Other wild Triticeae spp.:							
Agropyron spp.	1	2	3	2		4	12
Haynaldia villosa	10	44		3	4	22	83
Taeniatherum spp.						11	11
Other wild Gramineae spp.:							
Bromus spp.	36	84	15	25	5	34	199
Brachypodium spp.			4	1	4	7	16
Trachynia distachya	4	21	11	3	3	6	48
Lolium spp.	6	35	2	9	3	2	57
others	8	49	6	7	2		72
Other plant spp.:							
Leguminosae spp.	4	22	1	2		4	33
Caryophyllaceae spp.	1	6		3		10	20
Cruciferae spp.	4	11		6			21
others	24	36	17	19	2	7	105
Total	**237**	**689**	**106**	**154**	**61**	**327**	**1,574**

1) Pn : Southern part of Italian Peninsula, Sc:Sicily
Kp : Karpathos; Nx:Naxos; Pl:Peloponnesos; Ml:Mainland of Greece
2) F_1 hybrids between *Aegilops ovata* and *Triticum durum*
3) F_1 hybrid between *Aegilops triaristata* and *Ae. triuncialis*

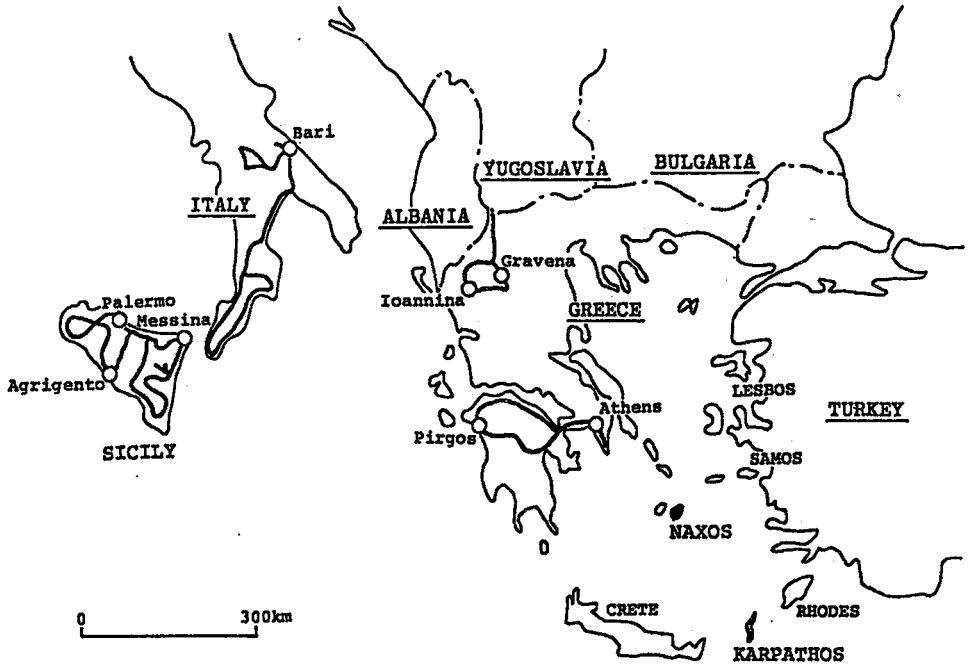

Fig. 1 The route of the mission (Furuta and Ohta)

Wheat Information Service
Number 72: 74 – 75 (1991)

Construction of genomic clone bank of *Triticum monococcum* Early mutant for RFLP analysis of wheat

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Triticum monococcum is a cultivated diploid wheat and lies a basic position of diversity of cultivated wheat given the genomic formula 'AA'. In order to compare the genomic sequences that locate in the specific position of chromosomes even in partial, between diploid and polyploid wheats, genomic clone bank of *T. monococcum* — Early mutant has been constructed.

Total DNA was isolated from etiolated seedlings according to the "CTAB" method. Extracted DNA was digested with *Pst*I and cloned into the *Pst*I site of plasmid, pUC18 by the 'shot gun' method. Recombinant DNAs were amplified by the polymerase chain reaction (PCR) methods. PCR has recently become a powerful tool for amplification of the desired DNA segment. Transformed bacteria, *i.e., Escherichia coli* str. DH5α were heated to 100℃ in 100μl of 0.1% Triton x-100 and 0.1mM Na_2EDTA, and then centrifuged at 15000 rpm for 2 min. After centrifugation, supernatants were used for PCR reaction. The condition for PCR was followed by the protocol recommended by the supplier, Takara Shuzo, Co. Ltd. Adjacent to the multiple cloning site of pUC18, the specific primers for DNA sequencing are commercially available. Using these oligonucleotides as forward and reverse primers, inserts were amplified by the PCR method.

It has been checked whether the PCR products were consistent with the real inserts or not. Ten plasmid DNAs were randomly picked up from the genomic library to check the inserts. These plasmid DNAs were extracted according to the ordinary alkaline method, and checked the DNA sizes with agarose gel electrophoresis. Additionally, these isolated plasmed DNAs were used for the templates of PCR reaction. Consequently, three kinds of inserts having possibly the same size should be obtained. The resultant DNAs were compared with agarose gel electrophoresis. The sizes of these inserts among three amplification methods were indentical and the sequences of them were comfirmed by Southern hybridization, indicating that the simple amplification method by PCR was efficient and reliable.

So, genomic library was amplified by the simple PCR method. Total 184 clones were amplified up to now. Size distribution of these clones is shown in Fig. 1. It is striking that an insert having 8.6kbp had been obtained by the PCR amplification, and inserts having less than 100bp were scarcely obtained. Average of inserted fragments was 1.6kbp. We can conclude from these data that the PCR method is enough efficient to amplify the inserts for probes of RFLP analysis of wheat.

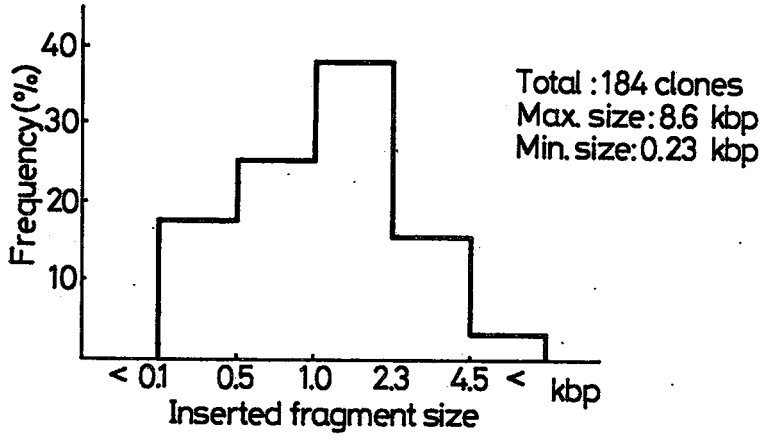

Fig. 1 Size distribution of genomic clone of *T. monococcum* amplified by the PCR method

Wheat Information Service
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Genetic differentiation between two wild tetraploid wheats, *Triticum dicoccoides* and *T. araraticum* as revealed by RFLP analysis of organellar and nuclear DNA

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Two wild tetraploid wheats, *Triticum dicoccoides* Körn. (2n = 28, genome constitution AABB), and *T. araraticum* Jakubz. (2n = 28, AAGG) are generally accepted as the ancestral species of the cultivated forms of the emmer and timopheevi groups, respectively. They are the first polyploid species evolved in *Triticum*. So, it is important to investigate the intra- as well as interspecific variation of these two species in order to clarify the origin of emmer and timopheevi wheats and to understand phylogeny of polyploid species in *Triticum*. We have studied the intra- and interspecific variations on organellar and nuclear DNAs of these two species by restriction fragment length polymorphism (RFLP) analyses. A large number of accessions of both species, of which collection sites extend across their entire natural distribution areas, were studied (Fig. 1). From these investigations the following points became clear: (1) The chloroplast genome is highly conserved in each species; When ctDNAs were treated with each of four 6-bp-cutters and electrophoresed, only two variant restriction patterns were found among 27 accessions of *T. dicoccoides*. No ctDNA variant was found among 27 accessions of *T. araraticum*, when studied in the same manners. (2) Contrary to the rare intraspecific variation in each species, the two species showed clear and distinct differences in their ctDNAs (Mori et al 1988), being identified to belong to Type 7 and Type

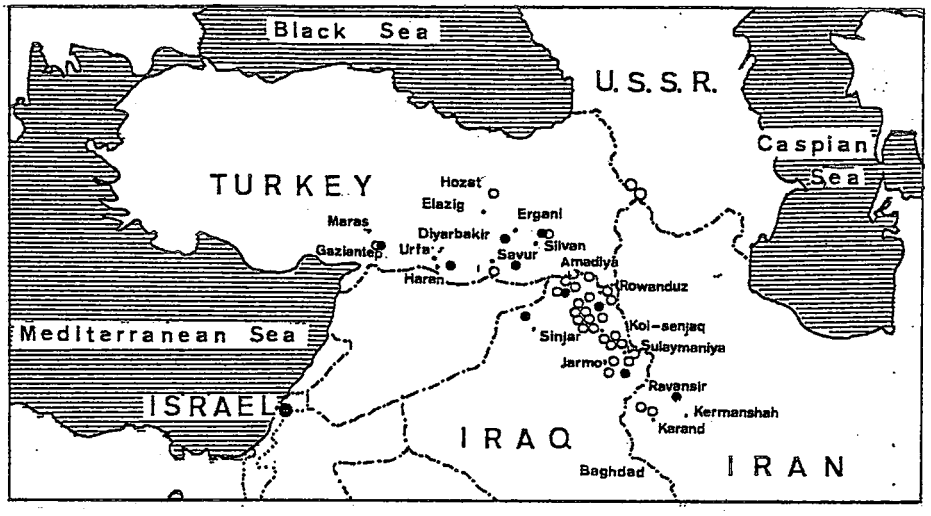

Fig. 1. Collection sites of *T. dicoccoides* (●) and *T. araraticum* accession (○).

5 chloroplast genomes (Ogihara and Tsunewaki 1988). (3) The mitochondrial genomes of the two species are also clearly distinguished from each other by both the restriction endonuclease analysis and Southern hybridization analysis of their mtDNAs; The percentage of common restriction fragments between the mtDNAs of the two species was 69.2% (Table 1). When hybridized with the two specific mtDNA clones as probes, RFLPs were detected between mtDNAs of the two species in their all digests. The average percentage of the common hybrid fragments was 49.1% (Table 2). (4) The interspecific variation of nuclear DNAs between the two species was much greater than the intraspecific varitation observed in each species; Total DNAs from 32 accessions of *T. dicoccoides* and 24 accessions of *T. araraticum* were treated with each of two 6-bp-cutters and probed to 28 nuclear DNA (or cDNA) clones. In total, 115 hybrid bands were observed per accession and more than two Southern patterns were found in all probe-enzyme combinations. The genetic distannces (d) estimated after Nei (1987) was 0.0128 on the average of 496 comparisons (range, 0.0034 - 0.0173) between *T. dicoccoides* accessions, whereas it was 0.0065 on the average of 276 comparisons (0.0009 - 0.0083) between *T. araraticum* accessios. The intraspecific variation of nuclear DNA in the former is about twice as large as that of the latter. On the contrary, the average genetic distance between the two species was 0.05 (0.0424 - 0.0549). Thus, it is clear that the interspecific variation between the two wild tetraploids is much greater than

Table 1. Number of the restriction fragments shared in common between mtDNAs of *T. dicoccoides* and *T. araraticum*

Enzyme	No. of fragments			%Common frag.
	dicoccoides (A)	*araraticum* (B)	Common frag. (C)	2C/(A+B)
*Bam*HI	55	53	37	68.5
*Hin*dIII	57	57	39	68.4
*Pst*I	54	50	37	71.2
*Pvu*II	59	58	44	75.2
*Xho*I	54	52	33	62.3
Mean	55.8	54	38	69.2

Table 2. Number of the hybrid bands observed in the mtDNA digests of *T. dicoccoides* and *T. araraticum* probed to two mtDNA clones. Number of the bands shared in common between the two species is given in parentheses

Probe	Enzyme					Total	%Common frag.
	*Bam*HI	*Hin*dIII	*Pst*I	*Pvu*II	*Xho*I	A(B)	2B/(A+B)
atpA	5(2)	3(0)	2(0)	3(0)	2(0)	15(2)	23.5
rRNA	3(1)	3(1)	5(3)	9(3)	5(3)	25(11)	61.1
Total	8(1)	6(1)	7(3)	12(3)	7(3)	40(13)	49.1

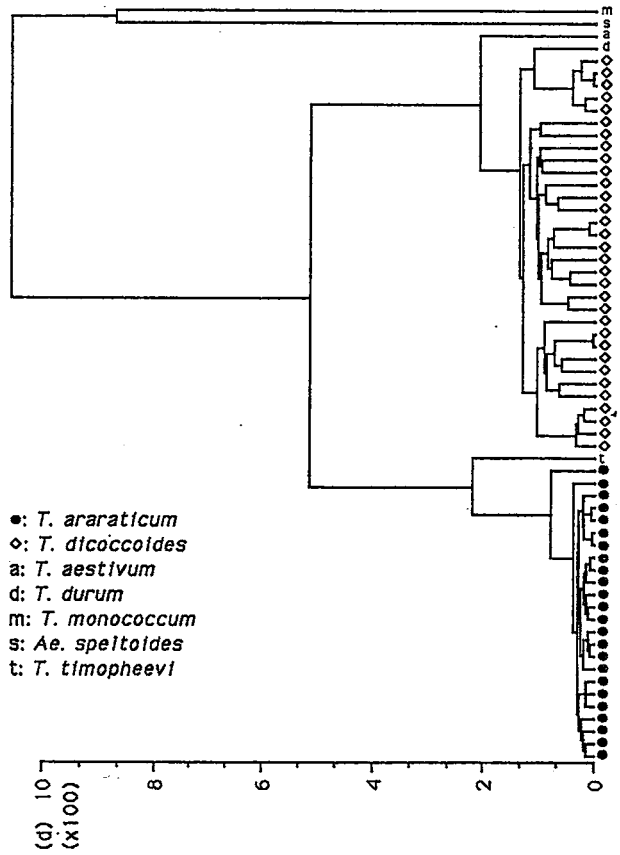

Fig. 2 A dendrogram showing genetic relationships among 61 polyploid wheat accessions, constructed after Sokal and Michener (1958) using the Nei's genetic distances (Nei 1987)

the intraspecific variation in each. A dendrogram constructed based on their genetic distances (Fig. 2) shows that all 32 accessions of *T. dicoccoides* and a *T. durum* accession form a cluster and, similarly, all 24 accessions of *T. araraticum* and a *T. timopheevi* accession form another cluster. These two clusters are distantly related to each other, indicating clear genetic differentiation between the emmer and timopheevi groups of wheat.

All these facts are in favor of the diphyletic origin of the two wild tetraploid species, and accordingly, of emmer and timopheevi groups of wheat.

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Wheat Information Service
Number 72: 79 – 82 (1991)

Two *Sec 3* loci of HMW secalin on the long arm of chromosome 1R in rye *(Secale cereale* L.)

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High molecular weight (HMW) secalin genes of endosperm proteins in rye are useful molecular makers for the long arm of chromosome 1R, together with a target of bread making quality improvement. The locus of HMW secalin was mapped and it was designated *Sec 3* by Shewry et al (1984a) and *Glu-R1* by Singh and Shepherd (1984).

We found two different loci of *Sec 3* on the 1RL by F_2 analysis using some inbred lines having different HMW secalin subunits.

Materials and Methods

Four inbred rye lines, IR27, IR48, IR51 and IR94 were chosen from The Tottori University Collections for making two cross combinations, IR51 × IR48 and IR27 × IR94. IR51 has three HMW seclain subunits which were denoted by band Nos. 1, 2 and 5 after electrophoresis fractionation and IR48 has band Nos. 2 and 6 (Fig. 1). IR27 and IR94 have band Nos. 4, 5 and Nos. 2, 4, 5', respectively, where, band No. 5' showed the same mobility but thin staining compared with band No. 5.

F_2 seeds were obtained from single F_1 plant in each cross combination. Same weight of flour samples were taken from single grains in F_2 seeds lot and the samples were fractionated by the SDS-PAGE method by Payne et al (1980) with a slight modification.

Results of F_2 analysis

1. F_2 of IR51 (Band Nos. 1, 2, 5) × IR48 (Band Nos. 2, 6): The bands 1, 5 and 6 segregated in the F_2 population (Fig 1). Results of the x^2 goodness of fit test for Mendelian segregation were shown in Table 1. The table shows that band Nos. 1, 5 and 6 were monogenic character and band No. 1 vs. No. 6 and No. 5 vs. No. 6 were allelic, but band No. 1 vs. No. 5 was nonallelic with considerable linkage. An estimation for recombination value between the two loci was made by maximum likelihood method as show in Table 2. Recombination value between the loci, 1-6 and 5-6, was calculated to be 7.0±1.0%

2. F_2 of IR27 (Band Nos. 4, 5) × IR94 (Band Nos. 2, 4, 5'): Three kinds of bands, Nos. 2, 5 and 5' segregated in the F_2 population. Results of the x^2 tests indicated that band No. 2 vs. null and No. 5 vs. No. 5' was allelic (Table 1). The recombination value between the two loci was 11.2±4.1%.

Table 1. Allelism test for HMW secalin subunit bands fractionated by SDS-PAGE of F_2 seeds of inbred rye line crosses, IR51 (band Nos. 1, 2, 5)×IR48 (band Nos. 2, 6) and IR27 (band Nos. 4, 5)×IR94 (band Nos. 2, 4, 5')

Band	No. of indiv. observed (Segregation phenotype)	Total	x^2	p
IR51×IR48				
Mendelian transmission test (3 : 1)				
	(Present : Absent)			
1-0	360 126	486	0.22	0.5-0.7
5-0	354 132	486	1.21	0.3-0.1
6-0	381 105	486	2.99	0.1-0.05
allelic (1 : 2 : 1) test				
1-6	(10 : 16 : 06)			
	105 255 126	486	3.00	0.3-0.1
5-6	(50 : 56 : 06)			
	105 249 132	486	3.30	0.3-0.1
non-allelic (dihybrid with linkage)				
1-5	(15 10 05 00)			
	340 20 14 112	486		
IR27×IR94				
allelic (3:1) test				
2-0	(2 : 0)			
	215 71	286	0.005	0.99-0.90
5-5'	(5 : 5')			
	202 84	286	2.913	0.30-0.10
dihybrid with linkage				
25'-05	(25 : 25' : 5 : 5')			
	138 77 70 1	286		

As band No. 2 and No. 4 in each F_2 were observed in all individuals examined, the bands were omitted from the segregation phenotypes.

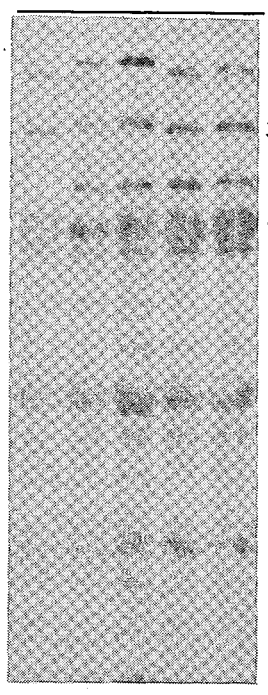

Fig. 1. Fractionation of secalins of F_2 seeds of IR51×IR48 by SDS-PAGE. The Nos. 1, 2, 5 and 6 indicate HMW secalin bands

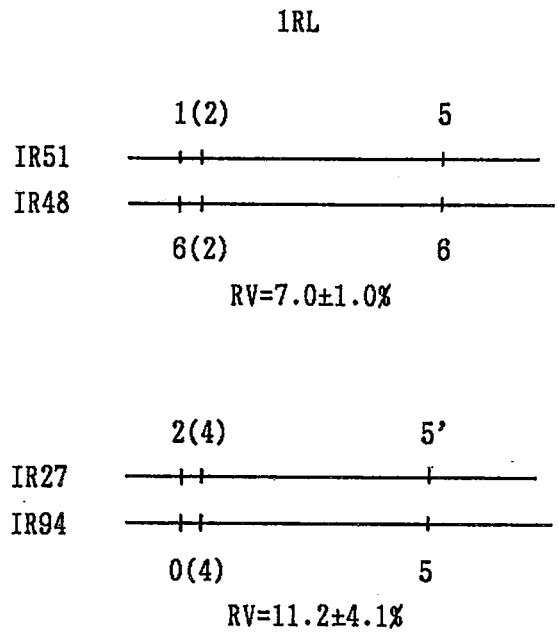

Fig. 2. Estimated positions of *Sec 3* loci on 1RL of four inbred rye lines

Table 2. Estimation for recombination value between duplicated *Sec 3* loci in F_2 of IR51×IR48

Phenotype	15	156	65	16	6
Ob. freq.	105	235	14	20	112
Exp. freq.	$N/4(1-P)^2$	$N/2$	$N/4\{1-(1-P)^2\}$	$N/4\{1-(1-P)^2\}$	$N/4(1-P)^2$
Zygotic genotype	15/15	15/16 15/65 15/66	65/65 65/66	16/16 16/66	66/66
Gametic genotype	15	15, 16, 65, 66	65, 66	16, 66	66

$P=0.070$ S.E. $=\pm 0.0098$

As the band (subunit) No. 2 was observed in all phenotypes examined, the band was omitted from all items in the Table.

Discussion

HMW secalins are related structurally to the HMW prolamins in wheat and barley (Shewry et al 1984b). HMW prolamins (glutenin) subunits of wheat are coded at each locus on the long arms of homoeologous group 1 chromosomes (Payne et al 1982). Each locus contained two HMW subunit genes, whose intra-locus recombination occurred very infrequently at *Glu-B1* locus being 0.11 ± 0.07cM and not observed at *Glu-D1* locus (Payne et al 1983).

Based on these facts, it can be assumed that there are two *Sec 3* loci at a distance of recombination value 7-11% on the long arm of chromosome 1R and each locus consists of one or two genes as shown in Fig. 2. These assumptions are supported strongly by the facts that number of HMW secalin bands is larger than those of wheat per genome and variation in the staining intensity of the same bands is observed in the F_2 populations and among inbred lines in our experiments.

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Wheat Information Service
Number 72: 83 (1991)

Further analysis of *Cs* chlorosis observed in hybrids between the Emmer and the Timopheevi group of tetraploid wheats

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Necrosis and chlorosis are hybrid weakness most commonly observed in intra- and inter-specific hybrids of wheat. In hybrids between the two groups of tetraploid wheats, the Emmer and the Timopheevi, chlorosis is reported by Tsunewaki and Hamada (1968) and by Tsunewaki and Nakai (1973). It is caused by two complementary genes; *Cs1* caried by wheats of the Emmer group and *Cs2* by the Timopheevi group. Later, Kawahara (1985) found three types of chlorosis, weak, medium and strong chlorosis, in several hybrids between wheats of the Timopheevi group and a *Cs1* tester. The present report deals with genes responsible to these three types of chlorosis. In order to study the gene(s) carried by the Timopheevi group, hybrids within Timopheevi group was pollinated by a *Cs1* tester strain, KU-123 of *Triticum dicoccum*. In total, 14 hybrid combinations were observed. Segregation of normal and chlorotic plants and of plants showing different types of chlorosis fitted well to 1 : 1 ratio. Kawahara (1985) already found that *Cs1* interact with gene(s) carried by the Timopheevi group irrespective to the degree of chlorosis. The present results indicate that the gene carried by the Timopheevi group consist of four alleles instead of the two, *Cs2* and *cs2*, reported earlier. Therefore, they were designated as *Cs2w*, *Cs2m*, *Cs2s* and *cs2*. Further, it was confirmed that *Cs2* of the earlier reports correspond to *Cs2w* in the present study. Distribution of *Cs* genes was examined by crossing the Emmer wheats with a *Cs2w* tester and the Timopheevi wheats with a *Cs1* tester. In wild Emmer, *T. dicoccoides*, 56 strains were examined but no *Cs1* was found. In cultivated Emmer wheats, *Cs1* allele was found in 3 strains and *cs1* in 24 strains. Over-all frequency of *Cs1* in the Emmer group was very low (3.6%) as was reported by Tsunewaki and Nakai (1973). In *T. araraticum*, wild Timopheevi wheat, 155 strains were examined. Of these, 87 strains (56.1%) had *cs2*, 29 (18.7%) had *Cs2w*, 32 (20.6%) had *Cs2m* and *Cs2s* was found in 7 strains (4.5%). Two alleles was found in cultivated *T. timopheevi;* 7 strains had *Cs2w* and 2 had *Cs2m*. In contrast to the Emmer group, about half (47.0%) of the strains of the Timopheevi group had either *Cs2w*, *Cs2m* or *Cs2s*.

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Wheat Information Service
Number 72: 84 (1991)

The difference of genes responsible for homoeologous pairing induction between *Aegilops speltoides* and *Ae. sharonensis* and its evolutionary significance

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The aims of the present investigaion were to analyze the genetic difference between two closely related diploid species *Aegilops speltoides* and *Ae. sharonensis* concerning homoeologous chromosome pairing induction in hybrids with tetraploid wheat and to determine whether the *Ae. speltoides* genome was homologous with the B genome of tetraploid wheat. For these purposes these two *Aegilops* species were crossed to each other. The F_1 hybrids showed nearly normal meiotic pairing and were further crossed with tetraploid wheat. The resulting triploid progeny was analyzed cytologically and morphologically. The amount of chromosome pairing segregated in a 3 : 1 ratio of a high pairing class versus a low pairing class. These two classes were corresponded to the classes caused by the parental *Aegilops* species. Intermediate classes or classes outside the range of the parents were not found. Because the present *Ae. sharonensis* strain caused little or no homoeologous pairing and its genome is not homologous with the B genome of tetraploid wheat, it was concluded that homeologous pairing induction was controlled by two duplicate and equally potent genes of *Ae. speltoides* segregation independently and that the *Ae. speltoides* genome was also not homologous with the B genome of tetraploid wheat. It was shown by morphological analysis that one of these genes was linked to a gene which controlled spike disarticulation.

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Characterization of alloplasmic common wheat with *Agropyron* cytoplasms

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Alloplasmic wheat with alien cytoplasms have provided good experimental materials for studying nuclear and cytoplasmic interactions between *Triticum* and its related genera (Tsunewaki 1988). In alloplasmic common wheat (*T. aestivum*) with *Agropyron* cytoplasms, a gross reduction in plant vigor and male sterility occurred and telocentric *Agropyron* chromosomes (telosomes) restored normal plant vigor and male fertility (Tsujimoto et al 1987, Nakamura et al 1991).

We studied four alloplasmic lines having two common wheat nuclei (cultivars Penjamo 62 and Siete Cerros 66) and two *Agropyron* cytoplasms (*trichophorum* and *glaucum*). Table 1 shows effects of *Agropyron* cytoplasms and telosomes on plant hight and selfed seed set. It is evident from the Table that *Agropyron* cytoplasms cause severe growth depression and high sterility in these alloplasmic lines, and that the presence of *Agropyron* telosomes restore normal plant vigor and fertility.

We reported that the alloplasmic lines showed significantly lower photosynthetic capacity than the euplasmic lines and the alloplasmic lines with telosomes and that the lower photosynthetic capacity was due to higher rates of dark respiration in green leaves (Nakamura et al 1991). To further characterize respiration in the alloplasmic lines we studied respiratory electron flows through the cytochrome and alternative paths and *in vitro* cytochrome c oxidase (cox) activity using 2-day-old seedlings and 2-week-old seedling leaves and roots. The reduction in plant vigor occurred in such early stages of development of the alloplasmic lines (Nakamura et al 1991). The respiratory parameters were measured by an oxygen electrode based on the rates of oxygen uptake in the presence and absence of respiratoty inhibitors (1 mM KCN, 5 mM salicylhydroxamic acid) and an uncoupler (2 and 10 μM carbonylcyanide m-chlorophenyl hydrazone). All the parameters differeed significantly at different ages and/or in different tissues: 2-day-old seedlings showed significantly higher activity than 2-week-old seedling leaves and roots. The activity of the cytochrome path agreed well with the *in vitro* cox activity. In 2-day-old seedlings the cox activity was higher in the alloplasmic lines with telosomes than in the euplasmic lines and the alloplasmic lines without telosomes, while in 2-week-old seedling leaves and roots the activity was higher in the alloplasmic lines without telosomes. Our results suggest that the interaction between common wheat nuclei, *Agropyron* cytoplasms and *Agropyron* telosomes is related to mitochondrial function.

The specificity of the effects of *Agropyron* telosomes on *Agropyron* cytoplasms was studied by evaluating the ability of telosomes for recovering plant vigor in the alloplasmic lines in which telosomes from one *Agropyron* species were introduced into cytoplasms of

Table 1. Growth (plant hight) and selfed seed set in alloplasmic (*Agropyron*)-*Triticum aestivum* hybrids

Cytoplasm donor *(Agropyron)*	Nucleus donor *(T. aestivum)*	Telosome*	Plant hight (cm)	Selfed seed set
	cv Penjamo 62		91.8	57.2
	cv Siete Cerros 66		99.2	94.2
glaucum	cv Penjamo 62	+	91.7	51.9
		−	28.1	0.0
	cv Siete Cerros 66	+	95.1	65.2
		−	43.6	0.5
trichophorum	cv Penjamo 62	+	103.2	36.6
		−	40.9	0.0
	cv Siete Cerros 66	+	101.5	61.6
		−	55.2	3.1

* + and − indicate the presence and absence of one or two telosomes, respectively.

Table 2. *In vitro* cox activity in (*Agropyron*)-*Triticum aestivum* hybrids

Line*	Seedling**	Root***	Leave***
	(nmol O_2/h/mg fresh weight)		
Pj, SC	190.1	54.1	68.7
(glc)-Pj, SC	148.4	115.4	152.8
(glc)-Pj, SC+t	235.0	39.2	54.2
(trc)-Pj, SC	180.0	115.3	154.4
(trc)-Pj, SC+t	232.1	54.7	82.8

* Pj: Penjamo 62, SC: Siete Cerros 66, glc: *glaucum*, trc: *trichophorum*, t: one or two telosomes,
** 2-day-old seedlings, *** 2-week-old seedling leaves and roots

Table 3. Growth (plant hight, fresh weight and dry weight) of (*Agropyron*)-*Triticum aestivum* hybrids

Line*	plant hight	Fresh weight	Dry weitht
	(cm)	(mg)	(mg)
Pj, SC	30.2	443.7	67.9
(glc)-Pj, SC	14.3	108.1	15.4
(glc)-Pj, SC+t ^{glc}	29.1	442.0	69.4
(glc)-Pj, SC+t ^{trc}	20.3	136.6	19.6
(trc)-Pj, SC	16.9	140.6	19.3
(trc)-Pj, SC+t ^{trc}	32.1	466.3	77.3
(trc)-Pj, SC+t ^{glc}	21.6	177.9	25.9

* t ^{glc} : *Ag. glaucum* telosomes, t ^{trc} : *Ag. trichophorum* telosomes, For all other designations see legends to Table 2.

the other species. Table 3 shows that *Agropyron* telosomes could recover plant vigor in the alloplasmic lines only when cytoplasms and telosomes were from the same *Agropyron* species. The result clearly shows that the effects of *Agropyron* telosomes on *Agropyron* cytoplasms are species-specific.

Cytological examinations of root tip cells showed that all *Agropyron* telosomes found in the alloplasmic lines were short-arm telosomes of *Agropyron* satellite chromosomes.

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Photosynthesis and respiration in eu- and alloplasmic common wheat with cytoplasms of *Triticum* and *Aegilops*

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Genetic diversity in cytoplasms of *Triticum* and *Aegilops* has been studied mainly using eu- and alloplasmic wheat having the same nuclear genotypes. It has become clear that cytoplasmic diversity manifests itself in various characters of the alloplasmic lines (Tsunewaki 1988). We report our results on cytoplasmic variability in photosynthesis and respiration, two of the most important cytoplasmic functions.

Photosynthesis (maximum photosynthetic capacity) and respiration of newly assimilated photosynthates (data not shown) in common wheat (*T. aestivum*) cv Chinese Spring (CS) with 17 different cytoplasms of *Triticum* and *Aegilops* (Table 1) were measured based on the rates of $^{13}CO_2$ assimilation and consumption, respectively, by far-red spectrophotometry. The maximum photosynthetic capacity in 3- and 10-week-old plants differed significantly among eu- and alloplasmic lines. Interestingly, a significant negative correlation was found between the photosynthetic capacity and growth measured based on dry matter weights in these lines. A similar negative correlation was reported in euplasmic *Triticum* and *Aegilops* species between their photosynthetic capacity and growth and/or yield (Evans and Dunstone 1970). This appareently paradoxical relationship therefore has its basis not only on nuclear genomes but also on cytoplasmic genomes. Groups of cytoplasms causing either growth depression or male sterility in CS nuclei showed higther photosynthetic capacities than euplasmic CS. No differences were found in the photosynthetic capacity between cytoplasms having H- and L- type Rubisco large subunits, which respectively show high and low *in vitro* Rubisco activity (Evans and Austin 1986).

All but C-type cytoplasms causing male sterility in CS nuclei and S group of cytoplasms showed higher rates of respiratory consumption of new photosynthates than CS, indicating that respiration in these cytoplasms depends more on newly assimilated carbons.

Respiratory electron flows were measured based on the rates of oxygen uptake using an oxygen electrode in the presence and absence of respiratory inhibitors (1 mM KCN and 5 mM salicylhydroxamic acid) and a uncoupler (2 and 10 μM carbonylcyanide m-chlorophenyl hydrazone). The *in vitro* catalytic activity of cytochrome c oxidase (cox) was measured also by an oxygen electrode. The total respiratory activity and capacity, the activity and capacity of the cytochrome and alternative paths, and the *in vitro* cox activity were all higher in 2-day-old seedlings than in 2-week-old seedling leaves and roots (Fig. 1, capacity data not shown). A significant negative correlation was found between the cox activity in roots and growth among the cytoplasms. The highest cox activity occurred in a A-type cytoplasm which caused the severest growth depression and complete male sterility.

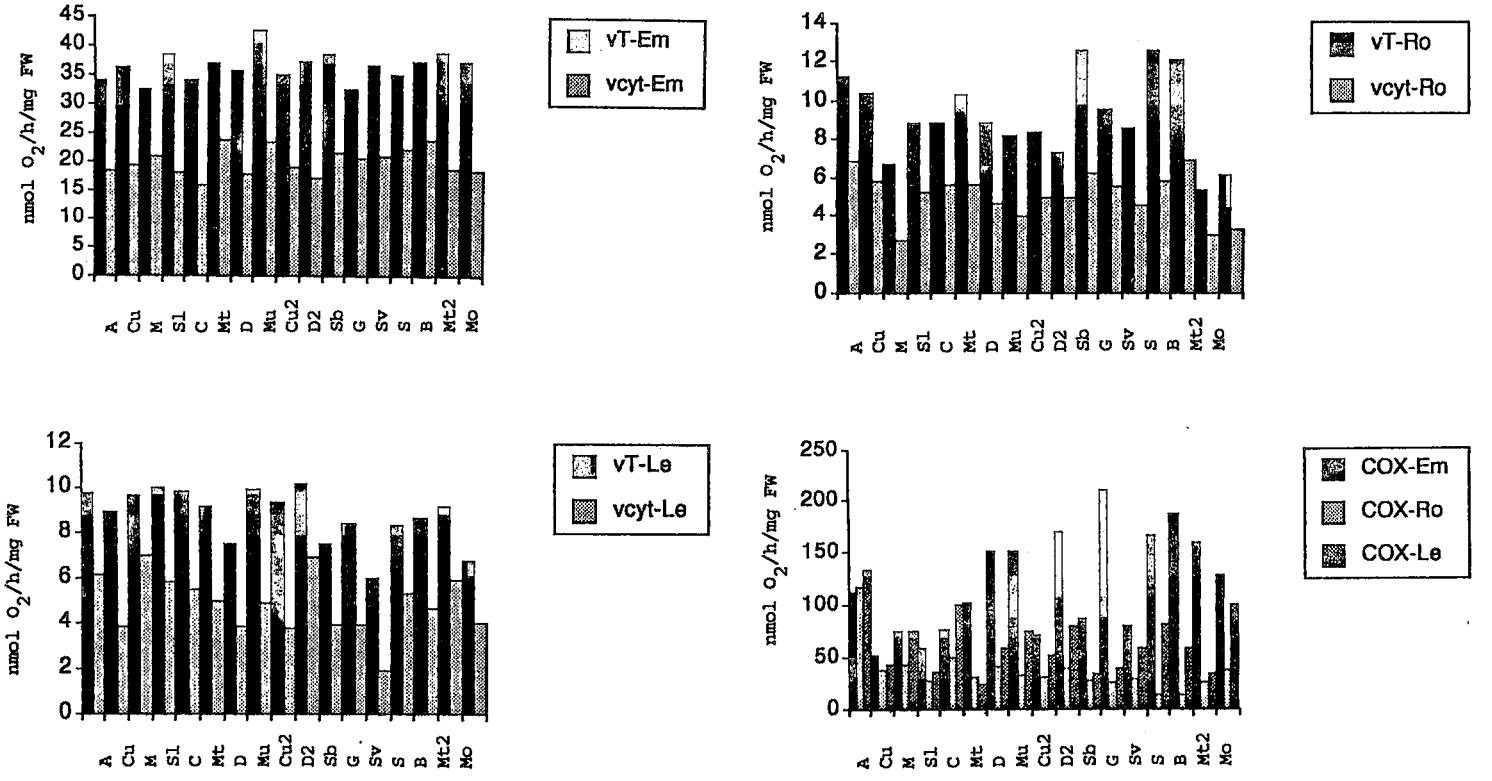

Fig. 1. Respiratory electron flows and *in vitro* cytochrome c oxidase activity in alloplasmic (*Triticum, Aegilops)-Triticum aestivum* cv CS. vT: total electron flow, vcyt: electron flow throuth the cytochrome path, Cox: cytochrome c oxidase activity, Em: 2-day-old seedlings (embryos), Le: 2-week-old seedling leaves, Ro: 2-week-old seedling roots. For designations of cytoplasms see Table 1

Table 1. Growth (dry matter weights) and maximum photosynthetic capacity in alloplasmic (*Triticum, Aegilops*)-*Triticum aestivum* cv CS

Cytoplasm*	Growth* depression	Male* sterility	Dry matter weight (mg)	Maximum photosynthetic capacity (mg C/h/g DW)
A	+	+	43.3	9.14
C^u	+		78.8	9.34
M	+	+	87.0	8.91
S^l	+		91.3	9.57
C	+	+	91.3	8.29
Mt			91.7	7.06
D			92.1	7.64
M^u			95.2	7.74
C^{u2}			97.0	7.63
D^2			97.8	8.29
S^b			99.2	8.03
G		+	99.6	7.81
S^v			102.7	7.79
S			106.7	8.15
B			109.7	6.95
Mt^2		+	119.0	8.58
M^o			131.4	7.46

* Tsunewaki K (1988)

The cox activity significantly differed depending on the ages and/or tissues: in 2-day-old seedlings the activity was higher in the euplasmic CS, whereas in 2-week-old seedling leaves and roots it was higher in the alloplasmic lines (data not shown). When growth depression-causing cytoplasms and male sterility-casuing cytoplasms were compared with the others, similar differential effects of plant ages and tissues were observed in the cox activity.

Our results thus revealed a large genetic variability in the effects of cytoplasms on photosynthesis and respiration, the parameters of which varied at different ages and in different tissues among the 17 different cytoplasms of *Triticum* and *Aegilops*.

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A new system for hybrid wheat production using *Aegilops crassa* cytoplasm

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Sasakuma and Ohtsuka (1979) observed complete male sterility in the alloplasmic line of *Triticum aestivum* cv. Norin 26 having *Aegilops crassa* cytoplasm (D^2 type cytoplasm by Tsunewaki 1980) in Hokkaido, a northern island of Japan. We showed that the male sterility was induced by long-day treatments exposed during the floral differentiation stage of the plants (Murai et al 1988). In this report, we propose a new system for hybrid wheat production using the D^2 type cytoplasm.

Materials and Methods

Alloplasmic lines, *Ae. crassa* /11* Norin 26 ((C)-N26) and *Ae. crassa* /11* Chinese Spring ((C)-CS) were used. Euplasmic and alloplasmic lines of N26 and CS were planted under various photoperiodic conditions in phytotrons as well as in the fields of the Kasai Experimental Farm of Sumitomo Chemical Co., Hyogo, and the Tanno Agricultural Cooperative, Hokkaido. Selfed seed fertility was estimated by the seed setting rate of first and second florets of all spikelets. A field trial for hybrid seed production was performed at Tanno, and yield and purity of hybrid seeds were estimated with the materials grown in the field (see Table 3).

Results and Discussion

The alloplasmic line of N26 shows almost completely male sterility under the long-day conditions of more than 15.0 hr light period, whereas N26 with normal cytoplasm is fertile under all photoperiodic conditions tested. Similarly, both the euplasmic and alloplasmic lines of CS exhibit high male fertility under the all conditions (Table 1). These results indicate that male sterility is induced by and interaction between the N26 nucleus and *Ae. crassa* cytoplasm under long-day conditions of more than 15.0 hr light period, and that the CS nucleus has some genetic factors which prevent the reduction of male fertility under long-day conditions. This male sterility will be called "photoperiod-sensitive cytoplasmic male sterility (PCMS)" to distinguish it from other types of cytoplasmic male sterility. The PCMS is caused by pistillody (Fig. 1).

The PCMS enables us to propose a new system for hybrid wheat production as shown in Fig. 2. Under the long-day conditions of more than 15.0 hr light period (condition A),

Table 1. Selfed seed fertility of euplasmic and alloplasmic lines of Norin 26 (N26) and Chinese Spring (CS) under various photoperiodic conditions

Nuclear donor	Cytoplasmic donor	Photoperiod (hr)			
		13.5	14.5	15.0	17.0
N26	(control)	82.7	60.3	71.5	45.5
〃	*crassa*	57.1	40.6	0.5	0
CS	(control)	54.6	51.6	59.9	74.1
〃	*crassa*	71.6	46.5	58.2	33.2

Note) 15° C in the dark and 18° C in the light period

Table 2. Selfed seed fertility of euplasmic and alloplasmic lines of N26 and CS under natural day length conditions at two different locations

Nuclear donor	Cytoplasmic donor	Kasai				Tanno	
		1986/87	87/88	88/89	89/90	1989	1990
N26	(control)	99.0	98.9	99.7	93.1	97.5	98.9
〃	*crassa*	56.8	53.3	72.8	60.6	1.0	6.5
CS	(control)	—	85.0	85.3	65.7	84.5	79.1
〃	*crassa*	—	70.7	69.2	69.7	66.2	61.3

Table 3. A field trial for hybrid seed production

Combination		Male sterility (%)	Out-crossing rate (%)	Hybrid purity (%)	Yeild (g/m^2)
PCMS line	Pollinator line				
(C)-N26	Ushio-komugi	86.5	40.9	75.2	93.5

Note)
Male sterility (%) = $(1-A/C)\times 100$
Outcrossing rate (%) = $(B/D-A/C)\times 100$
Hybrid purity (%) = $(B/D-A/C) / B/D\times 100$
A and B: No. of seeds/spikelet of bagged and non-bagged ear of (C)-N26, respectively.
C and D: No. of seeds/spikelet of bagged and non-bagged ear of N26, respectively.
Yield (g/m^2) = gram seeds / female plants in 1 m^2

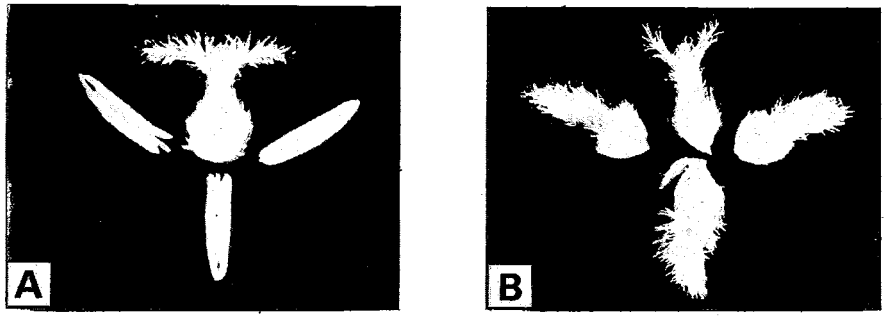

Fig. 1. Floral organs of the euplasmic and alloplasmic lines of *T. aestivum* cv. Norin 26 under a long-day condition. A: Euplasmic Norin 26, B: Alloplasmic Norin 26 with *Ae. crassa* cytoplasm

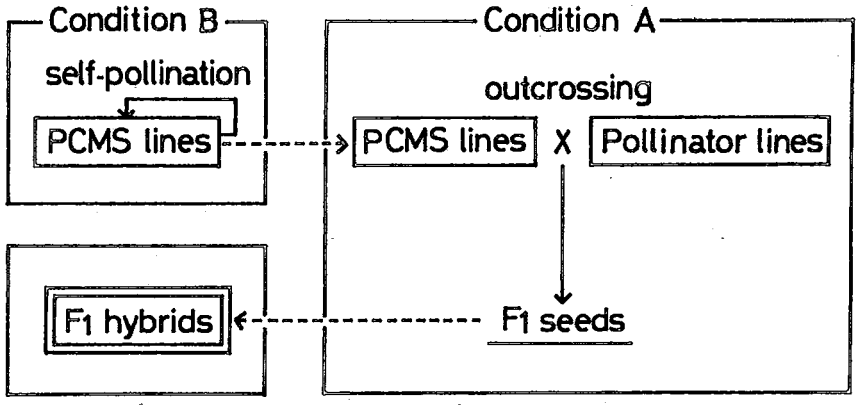

Fig. 2. A new system for hybrid wheat production using photoperiod-sensitive cytoplasmic male sterility. Condition A: Day length longer than 15 hr, Condition B: Day length shorter than 15 hr.

hybrid seeds can be produced from the outcrossing of a PCMS line with a cultivar used as the pollinator. The PCMS line can be easily maintained by self-pollination under photoperiodic condition of less than 15.0 hr light period (condition B). Hybrid wheat is grown in areas under the condition B. In contrast to the system of hybrid wheat production using *T. timopheevi* cytoplasm, the present system requires only PCMS and pollinator lines so that it is called a "two-line system".

As for in Japan, the condition A is found in Hokkaido and the condition B in southern part of Honshu, Kyushu and Shikoku islands. In fact, the alloplasmic line of N26 can be maintained by self-pollination at the Kasai Farm in Honshu and is used as PCMS line in the field at Tanno, Hokkaido, where male sterility is expressed (Table 2). In the field trial at Tanno, we could get 93.5 g/m^2 hybrid seeds of 75.2% purity (Table 3).

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Effect of culture condition on plant regeneration capacity of mature embryo derived callus in wheat (*Triticum aestivum* L.)

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Immature embryo has so far been the best explant for efficient regeneration of euploid plant in wheat. However it is required to exploit another explant, because it is usually difficult to get immature embryo throughout the year. In addition, the suitable stage of immature embryo is strictly limited (Shimada and Yamada 1979). This study was, therefore, attempted to establish an efficient culture method of callus derived from mature embryo as an alternative explant.

An Ethiopian local wheat cultivar 'IL 68' was used for embryo culture. To kow the effect of pre-treatment with 2,4-D in the course of embryo development, detached ears cultured after Kato et al (1990) were dipped in 2,4-D solution (2mg/l) for 3 minutes. This treatment was conducted at one of the seven developmental stages, that is, 8, 11, 14, 17, 20, 23 and 26 days after anthesis. Then treated ear was again cultured on liquid medium until their maturation. After ripening, caryopses were harvested and used for culture as well as control caryopses harvested from non-treated detached ear and from intact ear of field grown plant. These seeds were first soaked in hormone solution at 5° C for 3 hours. The solution contained MS inorganic salts, 30g/l sucrose and 2mg/l 2,4-D, combined with or without 1mg/l BAP as described by Sasakuma and Tei (1989). After seed sterilization, excised embryos were placed on agar medium with the scutellum uppermost. The medium contained MS inorganic salts, 30g/l sucrose and 8g/l agar, supplemented with 2,4-D and BAP as shown in Table 1. Callus induction and maintenance were conducted under a 26° C

Table 1. Culture medium and subculture interval in each culture

Medium	Culture No.		
	1	2	3
Pre-soaking	2,4-D + BAP		2,4-D
Callus induction	2,4-D + BAP 20 days		2,4-D 14 days
Subculture	2,4-D + BAP 40 days	2,4-D 100 days	2,4-D 90 days
	2,4-D 60 days		
Regeneration	free	free	free

Concentration of 2,4-D and BAP was 2mg/l and 1mg/l, respectively

and contiunous light (1500-2000 lux) regime.

Callus induction rate was not different between the two cultures, being 83.3% in culture No.1 and 85.7% in culture No. 3. It was also independent of the pre-treatment stage during detached ear culture. In culture No. 1 and No. 2, shoot bud appeared frequently at green spot region after 90 days of culture (Fig. 1). On the other hand, in culture No.3, it appeared after 130 days of culture. This result indicated that BAP added to pre-soaking medium and to callus induction medium enhanced the speed of shoot differentiation.

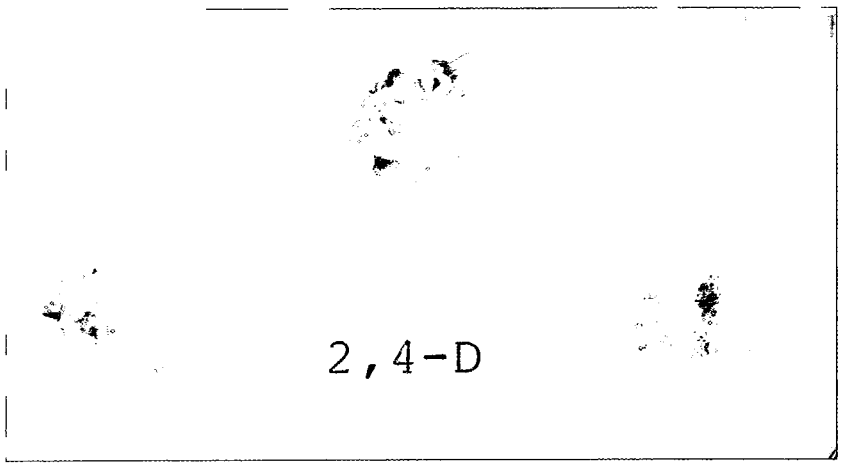

Fig. 1 Green spot formation and small shoot differentiation from mature embryo derived callus (Culture No.2, after 90 days)

Table 2. Number of plants regeneratied from mature embryo derived callus in each culture

Embryo source	No. of regenerated plants perinoculated embryo		
	Culture No.		
	1	2	3
Detached ear			
8-14 days	0.9	0.5	1.0
17-26 days	0.6	4.1	3.4
control	1.0	0.8	3.3
Intact ear	—	1.2	0.4

Culture efficiency, represented as the number of green plants per inoculated embryo, was generally lower in culture No. 1 than in the other cultures (Table 2). Embryos harvested from intact ear showed less regeneration capacity, that is, 1.2 and 0.4 plants from one inoculated embryo in culture No. 2 and No. 3, respectively. On the other hand, more than four plants were obtained in culture No.2 by culturing embryos pre-treated with 2,4-D in the later stage of detached ear culture. Similarly more than three plants were obtained in culture No. 3 by culturing the above-mentioned embryos and the control embryos harvested from detached ear. It was concluded that regeneration capacity of mature embryo derived callus decreased by the addition of BAP into subculture medium and increased by the pre-treatment with 2,4-D in the later stage of detached ear culture. Though the number of regenerated plants was still low, one original callus was usually divided into 4 pieces and the maximum number of regenerated green plants was 16 from one callus piece, indicationg that more than 60 plants could be obtained from one inoculated embryo.

In the second experiment, the effect of 2,4-D concentration in subculture medium on plant regeneration from mature embryo which was normally harvested from field grown 'IL 68'. Culture procedure was the same as in culture No.3 (Table 1), except that 1mg/l or2mg/l of 2,4-D was added to subculture medium. Though no green spot was formed by subculturing at 2mg/l of 2,4-D even after 100 days of culture, it was formed at 1mg/l of 2,4-D. The number of calli formed green spot was 0.87 per inoculated embryo. And more than two plants were obtained from on inoculated embryo. This result indicated that 2,4-D concentration in subculture medium was also the important factor for efficient plant regeneration from normal mature embryo.

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Somatic genome analysis: Cloning of a C-genome specific sequence

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Triticum-Aegilops complex is a good model for studying "polyploid evolution" because the genome constitutions of all the species are already kown. To characterize the genomes at a molecular level, we have been trying to isolate repetitive sequences which are present only in special genomes. In this symposium, we report our new clone, pAcH6, that is present repeatedly in the C genome of the diploid and tetraploid species.

We have isolated DNA from the seedlings of *Ae. caudata* (2n = 2x = 14, gemome CC) and digested with a restriction endonuclease *Hin*dIII. The DNA fragments obtained were inserted in the *Hin*dIII site of a plasmid vector pUC18 and the genomic library of C genome was established. Using total DNAs of *Ae. caudata* and common wheat (*Triticum aestivum* cv. Chinese Spring) as probes of dot-blot and Southern hybridization analyses, we selected a clone, pAcH6, that showed a strong signal only with the labeled *Ae. caudata* DNA.

The pAcH6 was labeled with digoxygenized dUTP (Boehringer Non-radioactive DNA hybridization kit), and hybridized to *Hin*dIII-digested DNAs of 20 *Triticum* and *Aegilops* species with genome(s) of A, B, C, D, G, M, N, S or U. Clear and uniform hybridization patterns appeared in the lanes of *Ae. caudata, Ae. cylindrica* (2n = 4x = 28, genome CCDD) and *Ae. triuncialis* (2n = 4x = 28, CCUU). Next, this clone was labeled by biotinyl dCTP (Enzo), and hybridized *in situ* to the chromosomes of *Ae. caucata.* Hybridization signals were observed on all seven chromosomes of this species.

The fact that the AcH6 sequence is present only in the species carrying the C genome indicates that it was amplified to a detectable level after the differentiation of the C genome. Additionally, the fact that the sequence is dispersed on all seven chromosomes of the C genome may indicate that the AcH6 sequence had a transposable nature. However, it must have been stabilized before the tetraploid species was established, because the Southern hybridization patterns of the three species were the same. The present clone will be useful to identify the C-genome chromosomes in the tetraploid species and also in the addition lines.

Hitherto, genomes in the polyploid species have been analyzed mainly by observation of chromosome pairing of the F_1 hybrids. Now, we can identify a specific genome at the somatic cell level by the above described molecular biological technique using genome specific sequences such as AcH6, which we have named "somatic genome analysis".

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Chromosome mapping of ribosomal RNA genes in *Triticum spelta* by *in situ* hybridization

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Introduction

We report here on physical mapping of ribosomal RNA genes (rDNA) in *Triticum spelta,* one of hexaploid wheat (2n = 42, AABBDD) by *in situ* hybridization. The origin of *spelta* wheat is discussed in terms of *in situ* hybridization patterns of rDNA.

Materials and Methods

Three strains of *T. spelta* were used in the present study. Wheat 18S-26S rebosomal RNA gene and the repeated DNA sequences from *Ae. squarrosa* (pAs1) were used as probes. The latter probe was used for the identification of wheat chromosomes (Rayburn and Gill 1986). The details of the procedure for cytological preparations, *in situ* hybridization using biotinylated probes, and detection of hybridization sites are as described by Mukai et al (1991).

Results and Discussion

In situ hybridization analysis using biotin-labeled rDNA probe indicated that four pairs of chromosomes carried Nor loci in *T. spelta* (Fig. 1a). The identification of chromosomes 1B, 6B, 5D and 1A was made by double labeling with rDNA and pAs1 probes. The latter clone reveals a small hybridization site on the satellite and on intercalary site of hybridization on the short arm in chromosome 6B (Fig. 1b). Chromosome 5D was also observed to have four sites of hybridization with pAs1. One site was located not on the satellite but on the terminus of the short arm and three intercalary sites of the long arm (Fig. 1c). No hybridization site was observed on chromosomes 1B and 1A. Therefore, the Nor chromosomes were confirmed as 1A, 1B, 5D, and 6B. The signal intensity was $1B > 6B \fallingdotseq 5D \gg 1A$. In chromosome 1B, most of the highly condensed rDNA signal was distributed in the proximal end and the remainder in the distal end of the secondary constriction joined by a middle faint signal. In chromosome 6B, condensed rDNA was distributed evenly at each end joined by diffuse rDNA. In chromosome 5D, condensed rDNA was found on the satellite. In most cells, a part of rDNA on chromosome 5D was dispersed, showing genetic activity of ribosomal RNA genes. Chromosome 1A showed a dot-type signal at the terminal end of the short arm. None of the rDNA on chromosome 1A was

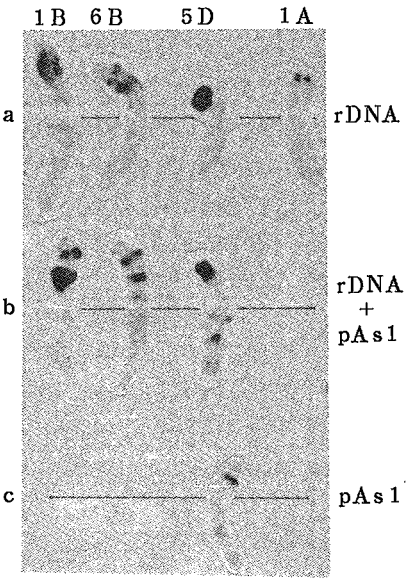

Fig. 1a. ISH of biotin-labeled rDNA probe to chromosomes 1B, 6B, 5D, and 1A of *T. spelta*

1b. ISH of both the rDNA probe and *Ae. squarrosa* repetitive DNA probe (pAs1) to chromosomes 1B, 6B, and 5D of *T. spelta*

1c. ISH of the pAs1 to chromosome 5D of *T. spelta*

found dispersed but remaind condensed.

Miller et al (1980) recognized four pairs of sites of rDNA in *T. spelta*. But, no identification of their loci was carried out except that a third site in size should be that on chromosome 1A. The presence of major rDNA on chromosomes 1A was investigated by *in situ* hybridization using plants in which chromosomes 1A from *T. spelta* have been substituted into Chinese Spring wheat. On the contrary, the present result clearly shows that the third site of rDNA is that of chromosome 5D. In many strains of *T. aestivum,* the greater most part of rDNA on chromosome 5D were deleted. The signal intensity of rDNA was 6B>1B>5D>1A. The rDNA site on chromosome 5D of *T. spelta* is almost the same as that of *Ae. squarrosa,* a diploid D-genome species, in the amount of hybridization signal.

There are two hypotheses on the origin of *T. spelta*. One hypothesis is that *T. spelta* is primitive species and *T. aestivum* was produced from *T. spelta*. The other hypothesis is that *T. spelta* is a relatively newly generated species and generated from *T. aestivum* or from a hybrid between *T. dicoccum* and *T. compactum* or *T. aestivum* (MacKey 1966, Tsunewaki 1969). Our ISH experiments fo rDNA of two genera, *Triticum* and *Aegilops* demonstrate that the size of rDNA has a tendency to diminish by terminal deletions of Sat-chromosomes in the course of evolution after amphidiploidization (Mukai et al. in preparation). The

present result supports the first hypothesis, though many studies support the second one (Liu et al 1990).

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Wheat Information Service
Number 72: 102 (1991)

Synaptonemal complex of rye

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Synaptonemal complex (SC) has been studied by thin sections, reconstructed three dimensional images from many thin sections, and/or surface spreading of meiocytes in various kind of materials. Each method has advantages and disadvantages. Well spread SCs in two dimensions are more favorable to trace whole length of the SC than the reconstructed ones. Surface spreading methods so far developed for plant materials are more or less hypotonic bursting technique with or without enzyme digestion of callose wall of pollen mother cells. Present report is a preliminary report on the surface spreading of SCs of rye. The method of Albini and Jones (1984) was used to make preparations. One of the well spread meiocytes was photographed by electron microscope (Figure 1.) and each of the SCs was carefully traced to measure the whole length of the lateral element. Total length of the seven SCs was 583 μm and relative length of each SC was calculated based upon the total length. The longest one was 0.19 (actual length, 110 μm) and the shortest was 0.09 (54 μm). This means that the longest one was twice as long as the shortest one. On the other hand somatic chromosomes at metaphase in rye range from 7 μm to 15 μm depending upon the pretreatment and the difference between the longest and the shortest is not so large as SCs. Gillies (1985) reported average length of the lateral element of SCs at zygotene and pachytene in rye. The longest one in his date was 72.2 μm (15.5% length) and the shortest was 61.8 μm (13.3%L) at pachytene of Snoopy. He also obtained similar result in R119. The difference between the present result and Gillies' data may be partly due to the difference of the method. Further study is undergone to reveal the chromosome architectrure of rye.

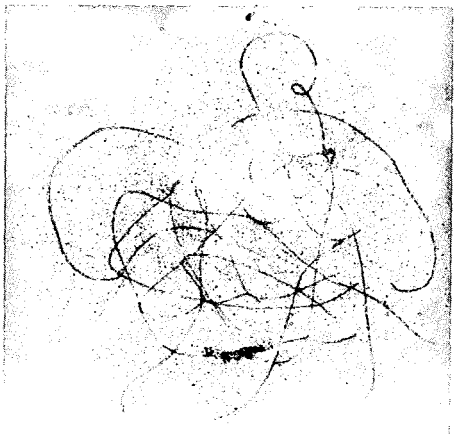

Fig. 1 Surface spreading of synaptonemal complexex of rye

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Number 72: 103 (1991)

On the origin of rye B chromosomes

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Rye B chromosomes from different regions are very similar in their morphology at somatic metaphase. The present study aimed to elucidate the relationships between rye B chromosomes in three different regions, Afghanistan, Turkey and Korea.

We carried out the following crosses, Afghan *S. segetale* (2B) × Turkish *S. cereale* (2B) and Afghan *S. segetale* (2B) × Korean *S. cereale* (2B). We selected their F_1 hybrids with four B chromosomes, which are considered to have the B chromosomes transmitted from both the pollen parent and the pistillate one, and observed the chromosome pairing of B chromosomes at diakinesis of pollen mother cells in the F_1 hybrids with four B chromosomes.

In ten F_1 hybrids with four B chromosomes obtained from Afghan *S. segetale* (2B) × Turkish *S. cereale* (2B), the frequency of the pollen mother cells showing multivalents ($1_{III} + 1_I$ and 1_{IV}) of B chromosomes varied from 12 to 38%. In 15 F_1 hybrids with four B chromosomes obtained from Afghan *S. segetale* (2B) × Korean *S. cereale* (2B), 8 to 32% of the pollen mother cells observed showed multivalents ($1_{III} + 1_I$ and 1_{IV}) of B chromosomes. Compared with the parental strains with four B chromosomes, the degree of the multivalent formation of B chromosomes in the F_1 hybrids is similar or somewhat low. These results indicate that the rye B chromosomes from Afghanistan, Turkey and Korea are homologous one another.

Therefore, we concluded that the origin of rye B chromosomes from these three different areas is most probably monophyletic. Further, we are going on to clarify the relationships of rye B chromosomes from Afghanistan to those from other countries, Iran and Japan.

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Number 72: 104 – 105 (1991)

Isozyme and chromosome polymorphisms of the genus *Avena* and its geographic distribution in Morocco

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All the biological species of *Avena* with the exception of *A. damascena* coexist in the area roughly between Southern Spain, Morocco and the Canary Islands. The most probable area of origin of the polyploids is where the putative lower ploidy ancestors and their higher ploidy descendants overlap. Clearly Southern Spain, Morocco and the Canary Islands represent the only area which meets such a demand. Four species, *A. canariensis*(2x),*A. agadiriana*(4x), *A. maroccana*(4x) and *A. longiglumis*(2x), in total 41 populations of the genus *Avena* were collected from the Canary Island and Morocco sponsored by the International Board for Plant Genetic Resources. In order to clarify genetic diversity within and between wild species of Avena, isozyme and chromosome variations were examined.

Avena canariensis is restricted to the Canary islands especially the islands of Fuerteventura and Lanzarote. I recorded and average of 53.8% variable loci, 1.76 alleles per locus and an expected heterozygosity of 0.226 in that from Fuerteventura but which values of Lanzarote were less than those of Fuerteventura. The populations from northern Lanzarote where is xeric rocky mountain area were monomorphic but had peculiar allozyme. The intraspecific variation of *A. canariensis* as indicated by morphological plasticity, variation of chromosome shape and isozyme polymorphism, has further been emphasized by a pair of satellite-deficiencies, very irregular cytomixis and a reciprocal translocation. These cause often meiotic irregularities and distort the structures or disrupt the process to bring about varying degrees of sterility or aberrant gametes. The term "meiotic drive" has been coined to characterize a preferential survival or perpetuation. *A. canariensis* has distinctive ecotypes, which are divided into early and late types. The late ecotype is having a late heading date, prostrate growth habit and pubescence whilst the early ecotype is early, erect and glabrous. These growth habits are positively correlated with isozyme and chromosome genotypes. The early type always had *Est-3S, Est-2N, Got-3M* and *Pgi-2F* alleles, whilst the late type had *Est-3F, Est-2F, Got-3S* and *Pgi-2S*. The same trend was observed in the satellite chromosome numbers. The early ecotype always had a pair of satellite chromosomes whilst the late ecotype always having two pairs of those. The early maturing types were distributed throughout the islands and were adapted mainly to disturbed habitats along ditches and roadsides. The later maturing types were collected in the southern part of Fuerteventura and the northern part of Lanzarote, mainly at high elevations in habits that were basically undisturbed by cultivation.

Avena agadiriana is the newly described tetraploid species, which is very similar to *A. canariensis*. This species were distributed from Casabranca to Tiznit along the Atlantic coast

of Morocco. Moreover, its distribution area is divided into two parts by Haut-Atlas mountains (alt. 4165 m). The northern part of the mountains is very heavy clay soil whilst the southern part is very dry and sandy soil. The genetic variability of 6 populations was as follows; an average of 46.3% variable loci, 1.50 alleles per locus and an expected heterozygosity of 0.154 were recorded by utilizing the isozyme frequency. Dendrogram resulting from a UPGMA cluster analysis showed a major dichotomy between the southern and northern part population. Chromosome variations, satellite-deficiency and translocation, were observed by hybridization between those two groups. The northern part group (3 population) is always having two pairs of satellite chromosomes and is later maturing type whilst the southern one (3 populatios) is always having three pairs of satellite chromosomes and is earlier maturing type. Intraspecific variations of isozyme and chromosome in *A. agadiriana* were very similar to *A. canariensis*. The chromosome types of *A. canariensis* were mainly in accord with the elevation of havitats but those of *A. agadiriana* were associated with specific soil characteristics and edafic factors.

Avena maroccana is a typical weedy species forming massive stands mixed with *A. sterilis* in cereal fields on heavy alluvial soil to which its large spikelet is well adapted. It has been discovered and repeatedly found only in the triangle between Tiflet-Rommani-Tedders in Morocco. It has been stated that *A. maroccana,* which is closely related to the hexaploids on the basis of morphology and meiotic affinities, appears to have a combination of the complements of *A. canariensis* and one of the C genome diploids. This species is distributed in the small area so that there is no sign of ecotypic differentiation whithin the species. However, the genetic variability of 11 populations was relatively larger than the former two species. An average of 44.3% variable loci, 1.82 alleles per locus and an expected heterozygosity of 0.182 were recorded. Dendrogram resulting from a UPGMA cluster analysis showed two clusters, which are roughly correlated with the geographic distance, and two independent populations (M7 and M9). Each population of *A. maroccana* appears to be adapted to a distinct microhabitat. Chromosome analysis on this species is now in progress.

Avena longiglumis is sporadically distributed in the Iberian peninsula, North Africa and Israel. It has very long slightly subequal glumes and very large anthers; the florets is the dispersal unit. It consists of two ecotypes, one a coastal type of robust, tall plant with large drop panicles; the other a desert type of shorter, slender plants with small panicles. Both ecotypes are specialized for sandy soils. The genetic variability of 6 populations was recorded as follows; an average of 38.3% variable loci, 1.78 alleles per locus and an expected heterozygosity of 0.160. Out of the 6 populations, one was the desert type (M61) and five were the coastal types. However, isozyme differentiation between two ecotypes was not detected. Chromosome analysis on *A. longiglumis* is also under investigation.

Wheat Information Service
Number 72: 106 – 108 (1991)

Transient expression of β-glucuronidase (GUS) gene in wheat pollen embryos via microprojectile bombardment

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1. Ishikawa Agr. College, 2. Kyoto Univ., 3. Hiroshima Univ.

Biolistic gene delivery is a useful method of introducing genes into cereal crop species, especially wheat in which neither infection of *Agrobacterium tumefaciens* nor efficient system for regeneration from protoplasts have been established (Gasser and Fraley 1989).

Cultured anthers of some wheat cultivars can produce pollen embryos at high frequencies which grow into green plantlets (Shimada and Otani, unpublished). These pollen embryos might could serve as recipients of alien genes to grow transformed wheat. In this work we demonstrated the transient expression of the β-glucuronidase (GUS) gene in wheat embryos derived from anther culture introduced by the pneumatic particle gun (Iida et al 1990). Anthers of wheat (*Triticum aestivum* cvs. Haruyutaka, BW2559, Glenson 81, Seri 82) were cultured on liquid Potato-2 medium. After a 1-month culture, pollen embryos (1 ~ 2 mm) were transferred onto regeneration medium in the petri dish (6 mm diameter). The procedure of wheat anther culture was as previously reported (Shimada and Otani 1988, 1989). The pollen embryos on the regeneration medium were bombarded with DNA-coated gold particles by a pneumatic particle gun as reported previously (Iida et al 1990).

Plasmid DNA, pBI221 which has the β-glucuronidase (GUS) gene under the control of the cauliflower mosaic virus 35S promoter and nopaline synthetase (NOS) polyadenylation signal was used.

The embryos and/or germination embryos were assayed for GUS expression using X-gluc substrate at 48 hr. and 1 week after bombardment. The expression GUS activity could be observed as blue-colored spots. GUS activity could be observed in 44 of 56 pollen embryos from 4 cultivars at 48 hr. after bombardment by DNA-coated gold particles (Fig. 1, Table 1). The number of blue spots varied from one to 15 with an average of 4.3. Two shots of bombardment increased the number of blue spots. After 1 week the bombarded embryos on the regeneration medium grew into plantlets. Young shoots and roots assayed by the X-gluc substrate showed only pale blue regions. Wheat seedlings might have endogenous GUS activity. We did not observe any dark blue spots, which would indicate a strong GUS positive response.

About 40 ~ 60% of the pollen embryos bombarded grew into normal green plants after a month (Table 2) and 10 ~ 20% of the green plants set seeds whose root tips did not express GUS activity.

Further optimization of the factors that may influence the efficiency of gene delivery, for example the introduced DNA concentration and the conformation of plasmid DNA, is required.

Table 1. Transient gene expression of β-glucuronidase (GUS) in pollen embryos following particle bombardment after 48 hr

Genotypes	Particle gun	No. of pollen embryos		Av. no. blue spots/ a embryo	(Range)
		tested	with blue spots		
BW2559	non	7	0	0	
	2 shots	8	7	6.5	(2 ~ 15)
Seri 82	non	8	0	0	
	1 shot	8	4	2.3	(1 ~ 4)
Glenson	non	8	0	0	
	1 shot	8	7	3.0	(2 ~ 7)
	2 shots	8	8	5.6	(2 ~ 13)
Haruyutaka	1 shot	8	5	2.8	(1 ~ 8)
	2 shots	8	7	4.3	(1 ~ 11)
	3 shots	8	6	4.8	(1 ~ 9)

Table 2. Effect of particle bombardment on the regeneration of pollen embryoids

Genotypes	Particle gun	No. embryos tested	No. (%) plants regenerated			
			Green plantlets		Albino plantlets	
BW2559	non	75	28	(37.3%)	1	(1.3%)
	2 shots	92	58	(63.0%)	5	(5.4%)
Seri	non	41	9	(22.0%)	3	(7.3%)
	1 shot	113	56	(50.0%)	23	(20.4%)
Glenson	non	71	30	(42.3%)	6	(8.5%)
	1 shot	54	36	(66.7%)	4	(7.4%)
	2 shots	67	35	(52.2%)	2	(3.0%)

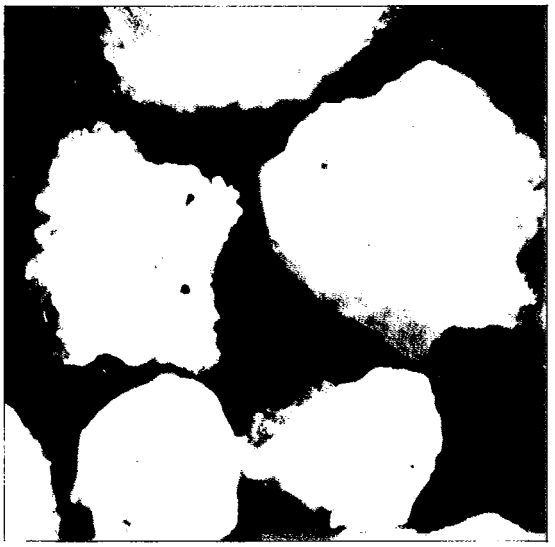

Fig. 1. Pollen embryos exhibited GUS-positive blue spots, assayed 48 hr following particle bombardment

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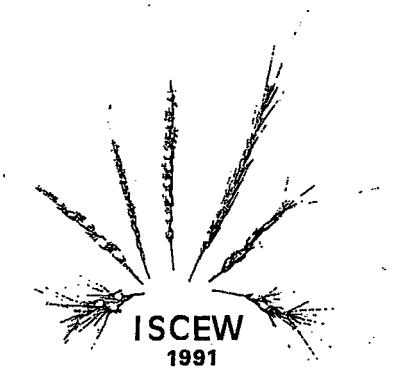
IV. Information

KIHARA MEMORIAL INTERNATIONAL SYMPOSIUM ON CYTOPLASMIC ENGINEERING IN WHEAT



Since Dr. H. Kihara started the introduction of alien cytoplasms into wheat in 1951, numerous research projects have been developed in the field of cytoplasmic inheritance in wheat and its relatives, including plasmon diversity, organelle genome analysis, molecular analysis of organelle genes, nucleo-cytoplasm interactions and practical utilization of cytoplasmic inheritance for wheat breeding. These studies in wheat have contributed to the development of other plant or crop applications as a model system for cytoplasmic engineering. Some of these studies have been conducted under internationally cooperative projects, which have been helpful for exchanging information and materials, and for cooperative research experiments. It has been proposed that we should discuss the achievements of research on cytoplasmic inheritance and enginnering.

International Symposium on Cytoplasmic Engineering in Wheat (ISCEW) is organized for the purpose of reviewing the achievements and discussing the prospect for further development in scientific studies on cytoplasms in wheat and its related studies. ISCEW will also be a good opportunity to offer international cooperation. We are very much welcome to invite you to attend the symposium.

Organizing Committee
Toshiro Kinoshita, The Cairman
Lab. Plant Bread. Hokkaido Univ.

OUTLINE OF ISCEW

DATE July 3-6, 1991.

PLACE University Conference Hall, Hokkaido University Sapporo, Hokkaido, Japan.

SESSION
1. Keynote Lectures: Achievement of Cytoplasmic Engineering
2. Organelle Genomes and Genes
3. Plasmon Diversity
4. Evolution: Cytoplasmic Donor to Polyploid Wheat
5. Nucleus-Cytoplasmic Interaction
6. Practical Use of Alloplasmon
7. Wheat Breeding: Problems and New Approarches
8. Field Trip to Experimental Stations and Biotechnology Institutes

PRESENTATION

Invitees on special topics from various countries.
General contributors (For oral or poster presentations)

LANGUAGE

Official language is English.

PROCEEDINGS

Proceedings of the symposium will be printed for publication in Seiken Ziho (special issue) soon after the symposium.

REGISTRATION

Anyone who is interested in the symposium should contact to the secretary of ISCEW. Registration fee is ¥5,000.

ORGANIZATIONS

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V. Editorial Remarks

Announcement for Future Issues

Thirty-seven years have been passed since Wheat Information Service was established in 1954, during this period 72 volumes of WIS issue have been published. Recently, it has been discussed that this journal should be improved to adapt for modern informative society, as well as overcoming the confirmed financial trouble. In order to secure the continuous publication of WIS, Kihara Memorial Foundation has dicided to support all expences of the publication and management.

From the next issue, No.73, WIS will be distributed to all the members on the mailing list. For a subscription please inform your name and mailing address to the Managin Editor.

WIS No.73 and 74 will be planned for publication in September 1991 and March 1992, respectively. Manuscripts for 73 will be accepted not later than August 15, 1991.

Acknowledgment

The cost of the publication has been defrayed by a contribution from Kihara Memorial Yokohama Foundation for the Advancement of Life Sciences. The editiors of WIS wish to express our sincere thanks to the foundation.

We also thank the reviewers listed below for their valuable efforts and contributions on behalf of this issue.

Fujigaki J, Kawahara T, Nakamura C, Nakata N, Nishikawa K, Noda K, Tosa Y, Tsunewaki K, Watanabe N, Yasumuro Y.

Explanation of the Picture on the Cover

Floral organs of the euplasmic and alloplasmic lines of *T. aestivum* cv. Norin 26 under a long-day condition. A: Euplasmic Norin 26, B: Alloplasmic Norin 26 with *Ae. crassa* cytoplasm. See the article by K. Murai et al in this volume for the details.

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