## CYTOGENETICAL STUDIES IN WHEAT

# I. MONOSOMIC ANALYSIS OF LEAF RUST RESISTANCE IN THE CULTIVARS URUGUAY AND TRANSFER

### By R. A. McIntosh,\* E. P. BAKER,\* and C. J. DRISCOLL<sup>+</sup>

[Manuscript received April 7, 1965]

#### Summary

The dominant gene for physiological resistance in the wheat cultivar Uruguay to certain Australian strains of leaf rust was located on chromosome 5D by the  $F_2$  method of monosomic analysis. The gene responsible for resistance in the cultivar Transfer was confirmed to be on chromosome 6B. Possible mechanisms producing aberrant  $F_2$  ratios involving Transfer in crosses with certain susceptible cultivars are discussed.

### I. INTRODUCTION

Despite the considerable improvements made in bread wheat (*Triticum aestivum* L. emend. Thell. subsp. *vulgare*) by plant breeders throughout the world for agricultural and commercial purposes, comparatively little progress had been made in the genetic mapping of this species until comparatively recently. Conventional genetic analysis has proved relatively unsatisfactory since duplicate (and triplicate) factors due to polyploidy frequently mask recessive mutations. Ausemus *et al.* (1946) summarized the results of genetic analyses which indicated few instances of simple inheritance. In addition linkages are rare because of the high chromosome number.

The three genomes, each of seven chromosomes, which constitute bread wheat are designated A, B, and D. These were contributed by progenitors of the presentday diploid species T. monococum L., Aegilops speltoides Tausch, and Aeg. squarrosa L. respectively (McFadden and Sears 1944; Riley, Unrau, and Chapman 1958). These species are cytotaxonomically closely related and hybrids between any two of them are characterized by a high number of bivalent associations. Riley (1960) showed that multivalent formations at the hexaploid level are prevented by a recessive mutation in one of the chromosomes (5B) of the B genome. In addition, correspondence in genetic activity between related chromosomes in the different genomes has been shown by Sears (1952). On the basis of nullisomic-tetrasomic compensations he divided the 21 chromosomes into seven groups, each of three homoeologues. Sears (1958) suggested individual chromosome designations to indicate these relationships, and this scheme is followed in the present paper. The correct designation for each particular chromosome was set out by Okamoto (1962).

Cytogenetical techniques have facilitated the genetic analysis of this species. Because of its polyploid nature, chromosome-deficient types are viable; all 21 monosomics are fertile and can be used in cytogenetical analyses. Progress in this

\* Department of Agriculture, University of Sydney.

<sup>†</sup> Department of Agriculture, University of Sydney; present address: Department of Botany, School of Biological Sciences, University of New South Wales, Sydney.

### R. A. MCINTOSH, E. P. BAKER, AND C. J. DRISCOLL

field has been largely due to Sears (1939, 1944, 1953, 1954) who described the production and characteristics of each of the monosomics and nullisomics in the cultivar Chinese Spring and indicated their use in cytogenetical analysis with certain specific examples. Monosomic series were established subsequently in other cultivars by backcrossing to the original Chinese Spring set. The particular procedure adopted in monosomic analysis depends on the inheritance pattern of the character under investigation and its presence or otherwise in the monosomic series being utilized. Unrau (1950) located a number of genes on specific chromosomes and subsequent publications by various authors have added considerably to information in this connection.

This Department has placed emphasis on breeding for disease resistance in wheat and one aspect of the cytogenetical investigations is to locate on particular chromosomes the genes responsible for resistance in the varieties being used as parents. Information as to the location on specific chromosomes of genes for disease resistance is of assistance in breeding programmes and is expected to be of value in assessing the relative roles of the three genomes in contributing such genes.

Athwal and Watson (1957) reported that Uruguay W 1064\* possessed two dominant genes conditioning resistance to wheat leaf rust (*Puccinia recondita* Rob. ex Desm.). One  $(Ug_1)$  controlled physiological resistance to all Australian strains of rust, except one, identified at that time. The second gene  $(Cw_1)$ , allelic to a similar factor in Chinese Spring, operated in the adult plant stage only and was effective against all known Australian strains. The genes involved were independent of those operative in Mentana W 1124 and Kenya W 744.

The cultivar Transfer (initially referred to as T 47) carries a translocation in the Chinese Spring background of a segment of *Aeg. umbellulata* Zhuk. chromatin which conditions leaf rust resistance (Sears 1956). This was obtained by X-irradiation of an appropriate *Aeg. umbellulata* addition line. By means of monosomic analysis and by chromosome pairing relationships in hybrids between Transfer and different Chinese Spring lines into which the specific *Aeg. umbellulata* chromosome had been substituted, Sears (1961) detected that the segment had been translocated to chromosome 6B of Chinese Spring.

#### II. MATERIALS AND METHODS

Uruguay W 1064 was obtained from the New South Wales Department of Agriculture, the cultivar being originally introduced from Uruguay. Seeds of the Chinese Spring monosomic lines as well as the cultivar Transfer W 2382 were originally provided by Dr. E. R. Sears, United States Department of Agriculture, Columbia, Missouri.

 $F_1$  plants were tested in the seedling stage with strain 76-Anz-0<sup>†</sup> of leaf rust and the meiotic chromosome constitution of each was determined. The  $F_2$  seedling populations in each line were inoculated with this strain and classified for reaction type.

\* W numbers refer to the Sydney University Wheat Accession Register.

<sup>†</sup> Strain classification according to Watson and Luig (1961).

### III. RESULTS

Uruguay W 1064 exhibited a highly resistant (";")\* reaction type in the seedling stage in contrast to the susceptible ("4") type of Chinese Spring. The  $F_1$  plants were characterized by a "; 1-" reaction type indicating almost complete dominance of resistance. As shown in Table 1 the segregation ratios of all  $F_2$  families originating

#### TABLE 1

segregation of seedling reaction type to leaf Rust strain 76-Anz-0 in monosomic  $F_1$  progenies from crosses involving the chinese spring monosomics with uruguay

Chromosome	Reactio	v <sup>2</sup> Value*		
Involved	··· ·· ·· 1_·"	"4"	(3:1  ratio)	
morod	(resistant)	(susceptible)	(0.1.1.0000)	
1A	38	16	0.62	
$2\mathrm{A}$	21	8	0.10	
3A	40	7	$2 \cdot 56$	
<b>4</b> A	<b>35</b>	14	0.33	
$5\mathrm{A}$	33	6	$1 \cdot 92$	
$6\mathbf{A}$	18	7	0.12	
7A	<b>25</b>	12	$1 \cdot 09$	
1B	95	36	0.43	
$2\mathrm{B}$	34	10	$0 \cdot 12$	
$3\mathrm{B}$	30	5	$2 \cdot 14$	
$4\mathrm{B}$	30	7	0.73	
$5\mathrm{B}$	<b>26</b>	6	0.67	
6B	<b>2</b>	1	0.11	
$7\mathrm{B}$	36	12	0	
1D	30	8	0.32	
$2\mathrm{D}$	71	14	<b>3 · 3</b> 0	
3D	63	20	0.04	
<b>4</b> D	<b>26</b>	10	0.15	
$5\mathrm{D}$	133	17	$44 \cdot 17$	
6D	<b>24</b>	10	0.35	
$7\mathrm{D}$	38	8	$1 \cdot 42$	
Total (excluding				
monosomic $5D$ )	715	217	$1 \cdot 46$	
Normal $(2n = 42)$ †	175	63	$0 \cdot 27$	

\* Values for significance:  $3 \cdot 84 (P = 0 \cdot 05); 6 \cdot 64 (P = 0 \cdot 01).$ 

† Disomic sib of monosomic 5D.

from the various monosomic  $F_1$  plants, with the exception of that involving 5D, conformed to that expected on the basis of a single gene for resistance. This establishes that the gene conditioning physiological resistance in Uruguay is located on chromosome 5D.

\* Reaction types based on description by Mains and Jackson (1926).

Transfer exhibited the highly resistant reaction type "0;".  $F_1$  seedlings of crosses with Chinese Spring similarly showed this reaction type, indicating complete dominance of resistance. As shown in Table 2 a number of  $F_2$  progenies of the various

	Reactio		
Chromosome		$\chi^2$ Value*	
Involved	۰۰ ٫۰	"4"	(3 : 1 ratio)
	(resistant)	(susceptible)	
1A	70	21	0.18
2A	167	63	0.70
3A	172	86	$9 \cdot 56$
<b>4</b> A	192	91	7.73
$5\mathrm{A}$	195	49	$3 \cdot 15$
6A	103	34	0.00
7A	88	36	$1 \cdot 08$
1B	220	78	$0 \cdot 22$
$2\mathrm{B}$	Not tested <sup>†</sup>	Not tested <sup>†</sup>	
$3\mathrm{B}$	197	79	$1 \cdot 93$
4B	105	38	0.19
$5\mathrm{B}$	202	87	$4 \cdot 01$
6B	122	3	$34 \cdot 05$
$7\mathrm{B}$	178	100	$17 \cdot 85$
1D	128	47	0.32
$2\mathbf{D}$	19	10	$1 \cdot 39$
3D	165	61	0.48
4D	182	75	$2 \cdot 40$
$5\mathrm{D}$	50	23	$1 \cdot 65$
6D	60	20	0
$7\mathrm{D}$	110	54	$5 \cdot 50$
Fotal (excluding monosomic 6B)	2603	1052	27.89
Normal $(2n = 42)$ <sup>†</sup>	105	58	9.74

TABLE 2

SEGREGATION OF SEEDLING REACTION TYPE TO LEAF RUST STRAIN 76-Anz-0 in monosomic  $F_1$  progenies from crosses involving the chinese spring monosomics with transfer

\* Values for significance:  $3 \cdot 84$   $(P = 0 \cdot 05)$ ;  $6 \cdot 64$  $(P = 0 \cdot 01)$ .  $\chi_{18}^2$  heterogeneity (contingency table) =  $27 \cdot 85$  $(P = 0 \cdot 10 - 0 \cdot 05)$  excluding monosomic 6B and normal 2n = 42. † A line used as monosomic 2B was subsequently shown to

be monosomic 6B, see text.

‡ Disomic sib of monosomic 6B.

monosomic  $F_1$  plants did not segregate in conformity with a resistant : susceptible ratio of 3:1. The total also significantly deviated from this ratio. By inspection it is obvious that the  $F_2$  family involving chromosome 6B segregated differently from the others, the low number of susceptible segregants confirming 6B as the bearer of the gene for resistance. A Chinese Spring line, believed to have been monosomic 2B and which gave a critical  $F_2$  ratio (resistant to susceptible of 350:5) with Transfer, was subsequently shown to be monosomic 6B on the basis of a critical ratio (in other crosses) for the stem rust resistance gene Sr 11 already known to be on 6B. Person (1956) has pointed out difficulties in maintaining monosomic lines due to univalent shift.

					TABLE	3					
SEGREGATION	FOR	SEEDLING	LEAF	RUST	REACTION	IN	${\bf F_1}$	PROGENIES	FROM	CROSSES	INVOLVING
TRANSFER WITH FEDERATION, YALTA, AND GABO RESPECTIVELY											

Cross	Cross F <sub>1</sub> Plant Number		Susceptible	$\chi^2$ Value* (3:1 ratio)	Heterogeneity (from contingency table)		
Transfer $\times$	II 62·461·1	414	98	9.38	h		
Federation	$\cdot 2$	202	57	$1 \cdot 24$	2 4.47		
	•3	466	90	$23 \cdot 03$	$\begin{cases} \chi_3 = 4 \cdot 47 \\ R = 0 \cdot 2 \cdot 0 \cdot 2 \end{cases}$		
	•4	122	25	$5 \cdot 01$	I = 0.3 - 0.2		
Total		1204	270	$35 \cdot 11$	J		
Transfer $\times$	$II 62 \cdot 463 \cdot 1$	201	34	13.90	L L		
Yalta	$\cdot 2$	134	22	9.88			
	•3	118	29	$2 \cdot 18$			
	$\cdot 4$	178	26	16.34	$\chi_6^2 = 8 \cdot 18$		
	.5	136	22	10.34	$\vec{P} = 0 \cdot 3 - 0 \cdot 2$		
	·6	87	9	$12 \cdot 50$			
	.7	34	10	0.12			
Total		888	152	$50 \cdot 82$	J		
Transfer $\times$	II 62·460·1	258	59	6.90			
Gabo	•2	125	48	0.70			
	·5	206	70	0.02	$\chi_4^2 = 7.74$		
	•6	145	36	$2 \cdot 52$	$P = 0 \cdot 2 - 0 \cdot 1$		
	•7	140	48	0.03			
$\mathbf{Total}$		874	261	$2 \cdot 43$	IJ		

\* Values for significance: 3.84 (P = 0.05); 6.64 (P = 0.01).

To determine possible reasons for deviations from a 3:1 ratio in non-critical  $F_2$  populations, Transfer was crossed with the susceptible varieties Federation W 107, Yalta W 1373, and Gabo W 1422. As shown in Table 3, when Federation and Yalta were involved the number of resistant  $F_2$  plants was significantly greater than that expected for single-gene segregations. A ratio of approximately  $4 \cdot 5:1$  in the case of Federation and approximately  $5 \cdot 8:1$  in the case of Yalta was obtained. The cross with Gabo did not differ significantly from conventional single-factor segregation, although the corresponding ratio was approximately  $3 \cdot 3:1$ .  $\chi^2$  values for heterogeneity in Tables 2 and 3 were not significant.

## IV. DISCUSSION

Soliman, Heyne, and Johnston (1964) reported that a gene  $(Lr_1)$  in Malakoff, responsible for physiological resistance to races 1 and 15 of leaf rust, was on chromosome 1B. In the current investigations 374 F<sub>2</sub> seedlings from a cross between Uruguay and Malakoff W 970 were all resistant to strain 162-Anz-1,2,3,4, suggesting allelism or close linkage. There was also a parallel behaviour of the two varieties to various leaf rust strains, those attacking Uruguay seedlings being virulent in each case on Malakoff, further suggesting that the genes are identical. There is no evidence to indicate that the genotypes of Malakoff used locally and by Soliman, Heyne, and Johnston are different. A genetic system in Malakoff whereby a different gene is operative against American and Australian strains is unlikely. Incorrect chromosome designation due to univalent shift in monosomic stocks or intravarietal translocations are possible reasons for the discrepancy. The location of the factor in Uruguay on chromosome 5D in the present study was confirmed using Redman monosomic 5D as the susceptible parent in a cross with Uruguay. The progenies of monosomic 5D F<sub>1</sub> plants segregated 59 resistant : 1 susceptible in contrast to a ratio of 55 resistant : 13 susceptible obtained with a 42 chromosome sib.

Chromosome 6B was found to bear the segment responsible for seedling resistance in Transfer, thus confirming the conclusions of Sears (1961). In crosses with Chinese Spring, the ratio of resistant: susceptible  $F_2$  plants was approximately  $2 \cdot 5 : 1$ . Luig (1960) and Sears and Loegering (1961) have reported instances of abnormal segregation in intervarietal wheat crosses. Sears and Loegering suggested that a pollenkilling gene (Ki) on chromosome 6B in Chinese Spring was responsible, whilst Luig claimed that various factors, including meiotic abnormalities, were responsible. Since Chinese Spring and Transfer are presumably identical except for the translocated segment carried by the latter, such mechanisms cannot explain the present aberrant results. A more likely explanation is that male gametes bearing the *Aegilops* segment compete unfavourably with the normal type (as also suggested by Sears, personal communication). By contrast, in crosses with Federation and with Yalta, the transmission of the chromosome from Transfer carrying the segment for resistance was favoured.

Luig (1964) has shown that chromosome 6B from Chinese Spring has enhanced transmission in crosses with Yalta and with Gabo bearing the stem rust resistance gene Sr 11 on this chromosome. In the present studies a Federation × Gabo F<sub>2</sub> population segregated in conformance with a 3 : 1 ratio when tested with stem rust strain 126-Anz-6, 208 seedlings having a "; 1 =" highly resistant reaction type characteristic of Gabo, and 64 being susceptible. The absence of a distorted ratio suggests that enhanced transmission of chromosome 6B from Chinese Spring (or Transfer) would be observed in crosses of Federation with Chinese Spring (or Transfer). The confirmation of this prediction in the present instance, together with the F<sub>2</sub> segregation ratio in the cross Transfer × Yalta, indicate that reduced transmission of chromosome 6B from Transfer due to the incorporated *Aeg. umbellulata* segment was outweighed by its enhancement due to mechanism(s) cited by Sears and Loegering and Luig.

The present results can be adequately explained on the basis of a pollen-killing gene closely linked in coupling with Transfer leaf rust resistance. However, Luig (1964) described heterogeneity both within and between  $F_1$  plant progenies in crosses involving varieties carrying certain genes conditioning rust resistance on chromosome 6B. This implies a more complex mechanism than the action of a single gene with uniform penetrance and expressivity.

#### V. Acknowledgments

These investigations were supported by a grant from the Wheat Industry Research Council. The authors express their thanks to Mr. J. Green for technical assistance.

#### VI. References

AUSEMUS, E. R., HARRINGTON, J. B., REITZ, L. P., and WORZELLA, W. W. (1946).—A summary of genetic studies in hexploid and tetraploid wheats. J. Am. Soc. Agron. 38: 1082–99.

LUIG, N. H. (1960).-Differential transmission of gametes in wheat. Nature, Lond. 185: 636-7.

LUIG, N. H. (1964).—Heterogeneity in segregation data from wheat crosses. Nature, Lond. 204: 260-1.

MAINS, E. B., and JACKSON, H. S. (1926).—Physiologic specialization in leaf rust of wheat, Puccinia triticinia Erikss. Phytopathology 16: 89–120.

- McFADDEN, E. S., and SEARS, E. R. (1944).—The artificial synthesis of *Triticum spelta*. Genetics **30**: 14 (abstr.).
- OKAMOTO, M. (1962).—Identification of the chromosomes of common wheat belonging to the A and B genomes. Can. J. Genet. Cytol. 4: 31-7.

PERSON, C. (1956).—Some aspects of monosomic wheat breeding. Can. J. Bot. 34: 60-70.

RILEY, R. (1960).—The diploidization of polyploid wheat. Heredity 15: 407-29.

RILEY, R., UNRAU, J., and CHAPMAN, V. (1958).—Evidence on the origin of the B genome of wheat. J. Hered. 49: 91-8.

SEARS, E. R. (1939).—Cytogenetic studies with polyploid species of wheat. I. Chromosomal aberrations in the progeny of a haploid of *Triticum vulgare*. Genetics 24: 509-23.

- SEARS, E. R. (1944).—Cytogenetic studies with polyploid species of wheat. II. Additional chromosomal aberrations in *Triticum vulgare. Genetics* 29: 232–46.
- SEARS, E. R. (1952).-Homologous chromosomes in Triticum aestivum. Genetics 37: 624.

SEARS, E. R. (1953).—Nullisomic analysis of common wheat. Am. Nat. 87: 245-52.

SEARS, E. R. (1954).—The aneuploids of common wheat. Res. Bull. Mo. Agric. Exp. Stn. No. 572.

- SEARS, E. R. (1956).—The transfer of leaf rust resistance from Aegilops umbellulata to wheat. "Genetics in Plant Breeding." pp. 1–22. (Brookhaven Symposia in Biology No. 9.)
- SEARS, E. R. (1958).—The aneuploids of common wheat. In "Proceedings First International Wheat Genetics Symposium." pp. 221–9.
- SEARS, E. R. (1961).—Identification of the wheat chromosome carrying leaf rust resistance from Aegilops umbellulata. Wheat Inform. Serv. 12: 12-13.

SEARS, E. R., and LOEGERING, W. Q. (1961).—A pollen-killing gene in wheat. Genetics 46: 897 (abstr.).

SOLIMAN, A. S., HEYNE, E. G., and JOHNSTON, C. O. (1964).—Genetic analysis for leaf rust resistance in the eight differential varieties of wheat. Crop Sci. 3: 246-8.

- UNRAU, J. (1950).—The use of monosomics and nullisomics in cytogenetic studies of common wheat. Sci. Agric. 30: 68-89.
- WATSON, I. A., and LUIG, N. H. (1961).—Leaf rust on wheat in Australia: a systematic scheme for the classification of strains. *Proc. Linn. Soc. N.S.W.* 86: 241–50.

ATHWAL, D. S., and WATSON, I. A. (1957).—Inheritance studies with certain leaf rust resistant varieties of *Triticum vulgare*. Proc. Linn. Soc. N. S.W. 82: 272-84.