THE INHERITANCE OF RESISTANCE TO *RHYNCHOSPORIUM SECALIS* IN BARLEY

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1. INTRODUCTION

SEVERAL studies have been carried out, mainly in the U.S.A., on the inheritance of resistance to the leaf blotch (or scald) disease of barley, caused by *Rhynchosporium secalis* (Oudem.) J. J. Davies. Mackie (1929) found that resistance in an unspecified variety was controlled by a single recessive gene. Riddle and Briggs (1950) identified a single dominant gene conferring resistance in *La Mesita*, which was also present with a recessive gene in *Trebi* and *California 1311* (= *Modoc*), and with one or more additional dominant genes in *Turk*. A designation for genes conferring resistance to *R. secalis* was first used by Bryner (1957), who allocated the symbol *Rha* (later amended to *Rh* by Robertson, 1960) to a dominant gene found in *Brier*.

Dyck and Schaller (1961a) isolated a further five dominant genes for resistance, which were designated *Rh2* (in *Atlas* and *Atlas 46*), *Rh3* (in *Atlas 46* and *Turk*), *Rh4* (in *La Mesita*, *Osiris* and *Trebi*), *Rh4²* (an allele at the *Rh4* locus, in *Modoc*) and *Rh5* (in *Turk*). These genes were shown to be specific against certain races of the pathogen. The *Rh3* and *Rh4* loci were very closely linked on chromosome 3 (Dyck and Schaller, 1961a, b), and *Rh3* was considered to be identical, allelic or closely linked to the *Rh* locus in *Brier*.

A pair of complementary recessive genes, *rh6* and *rh7*, in *Jet* and *Steudelli*, and an incompletely dominant gene, *Rh9*, in *Kitchin* and *C.I. 668*, were found by Baker and Larter (1963), who also confirmed the presence of a single dominant gene in *Turk*. Wells and Skoropad (1963) identified a recessive gene for resistance in *Nigrinudum*, designated *rh8*, and a dominant gene, considered to be *Rh3*, on chromosome 3 of *Turk*; this same gene, or one closely linked to it, was also found in *Bey*, *Rivale*, *36Ab 1991*, *C.I. 3515*, *C.I. 8256* and *Osiris*. Frecha (1967) found that the varieties *Osiris*, *Psaknon* and *Atrada* × *Atlas* (= *Atlas 46*) had a dominant gene for resistance in common, and that the latter variety also had an additional dominant gene.

The results of a study on 23 varieties by Starling *et al.* (1971) suggested that *Atlas 46*, *Turk*, *Brier*, *La Mesita*, *Modoc*, *Gembloux 14*, *Alaska* (*C.I. 534* and *C.I. 4106*), *Tennessee Winter* (*C.I. 876*), *Kentucky No. 36*, *Olympia*, *Tschermak*, *Hudson*, *Carstens 2-row*, *C.I. 3515*, *C.I. 8071*, *C.I. 8101*, *C.I. 8256*, *C.I. 9042* and *C.I. 10176* all had a single gene or gene locus in common. *Atlas 46* had an additional gene which was also found in *Atlas*, and *C.I. 3515* also had an additional dominant gene, different to that found in *Atlas*. *C.I. 8618* was found to contain a dominant gene not found in any other variety tested.

In Britain, Evans (1969) detected a single dominant gene for resistance in *Atlas 46*, *Turk* and *Osiris*; since these genes were either allelic or closely linked, they were presumed to be *Rh3* in the first two varieties and *Rh4* in the last.

In most of the studies cited above, no recombination was observed in F_2 or F_3 progeny from crosses between two resistant varieties. The use of small test populations and/or single isolates of *R. secalis* frequently made it impossible to verify whether the genes in the two varieties were allelic, closely linked or identical. Consequently, the genetic diversity of host resistance, and hence the potential of the available resistance sources for breeding purposes, cannot be determined.

2. MATERIALS AND METHODS

The 18 barley varieties used in this experiment gave the reactions shown in table 1 when inoculated with three isolates of *R. secalis* provided by Dr H.

TABLE 1
The reactions of 18 barley varieties to three isolates of R. secalis

Variety			Isolates		
Cb*	Name	C.I.†	1394	1435	1436
763	<i>Proctor</i>	—	S	S	S
1098	<i>Maris Otter</i>	—	S	S	S
1084	<i>Wisc. W. × Glabron</i>	8162	R	R	S
957	<i>Turk</i>	5611-2	R	R	S
952	<i>Atlas 46</i>	7323	R	R	S
1080	<i>Brier</i>	7157	R	R	S
1081	<i>Hudson</i>	8067	R	R	S
1092	<i>Dea</i>	—	R	R	S
1085	<i>Modoc</i>	7566	R	S	S
956	<i>Osiris</i>	1622	R	R	R
1086	<i>La Mesita</i>	7565	R	R	R
1105		3515	R	R	R
1106		8256	R	R	R
1107	<i>Gembloux 14</i>	8286	R	R	R
1087	<i>Jet</i>	967	R	R	S
1149	<i>Nigrinudum</i>	11797	R	R	S
1089		4364	R	R	S
1090		4368	R	R	S

* Accession Number Arable Crop Breeding Department, Welsh Plant Breeding Station, Aberystwyth.

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Owen, University of Reading. These isolates were selected as being representative of the pathogenic variation available in Britain. On the basis of these results, a hybridisation programme was devised from which F_1 , F_2 or F_3 progeny were obtained for testing. The F_1 hybrids were grown in the glasshouse and, with the exception of the crosses between *Turk*, *Atlas 46* and *Osiris*, all F_1 hybrid plants were protected at anthesis to ensure that no outcrossing occurred. The F_3 families were obtained from spaced F_2 plants grown in the field, which were not protected against outcrossing.

The reactions of progeny to *R. secalis* were determined by tests on seedlings. These were sown in rows 40 mm. apart in shallow boxes of sterile compost, in a glasshouse maintained at 10° to 20° C. The parent and F_2 plants were spaced about 20 mm. apart, F_1 hybrids 35 mm. apart, and each F_3 family consisted of 20-25 plants sown about 10 mm. apart. These plants

were inoculated 14 days after sowing, when the second leaf had partially emerged.

Inoculum was prepared by growing the cultures of *R. secalis* on 2 per cent. lima bean agar (Schein and Kerelo, 1956) in 250 ml. medical flat bottles to which 10 ml. sterile distilled water was added at the time of seeding (Sarasola and Campi, 1947). After incubation at 18° C. for 10 days, the mycelial mat was scraped off, macerated in water, and filtered through muslin to remove large mycelial fragments. The conidial concentration was adjusted to 800,000-1,000,000 per ml., and the suspension sprayed on to the plants. In one cross (*Osiris* × *Proctor*), F_2 plants were tested against two isolates simultaneously, by rubbing a conidial suspension of one isolate on to the second leaf and the other on to the third leaf.

Following inoculation, the boxes were placed in a shallow trough of water under a polythene-covered frame for 48 hours to maintain high humidity, and then allowed to dry. Symptoms generally appeared 10-14 days after inoculation, and were scored 4 days after their first appearance. In some cases, a second assessment was made 28 days after inoculation.

Although the reactions of the parent varieties fitted the categories described by Dyck and Schaller (1961*a*), it was found that in most cases the progeny could be more simply classified as resistant or susceptible on the basis of the first leaf reaction. At the conidial concentrations used in this study, the first leaves of susceptible segregates wilted completely, whilst the resistant ones were unaffected. However, in a few instances, the tips of the first leaves and small areas of the second leaves showed symptoms; this is referred to as a "type 2" reaction, in contrast to the "type 4" of complete susceptibility and "type 0" of complete resistance.

Crosses between two resistant varieties frequently produced F_2 or F_3 populations which were completely resistant. In such cases, the genes in the two varieties could either be allelic or linked too closely for a recombinant to be detected in the population tested. The maximum recombination frequency which could have passed undetected (at $P = 0.05$) was calculated from the formula outlined by Hanson (1959), viz.:

$$P_{rc} = 1 - \sqrt[n]{0.05}$$

where n is the number of F_2 plants or F_3 families tested, and P_{rc} is the proportion of detectable recombinants. With two genes in repulsion, this is equal to $p^2/4$ in the F_2 generation and $(4p - p^2)/4$ in the F_3 generation. The same formula was also used when F_2 plants were tested simultaneously against two isolates of the pathogen, to determine whether one or two genes were involved. For two genes in coupling, P_{rc} is equal to $(2p - p^2)/2$.

The location of the resistance genes in *Turk* and *Osiris* was determined from crosses between these varieties and stocks homozygous for the chromosome 3 recessive marker genes "streaked" (*Stst*) which produces streaked seedlings, and "uzu" (*Uzuz*), which produces dwarf plants recognisable in the seedling stage, and the chromosome interchange stock T2-3d. These were tested for resistance and the marker character or semi-sterility in F_2 or F_3 in the field or glasshouse, as indicated in the results. Linkages between resistance genes and marker genes were calculated by the product method, using tables by Fisher and Yates (1963), and between resistance genes and the T2-3d break-point using tables given by Joachim (1947).

3. RESULTS

The F_2 progeny from a cross between *Cb 1084* (*Wisconsin Winter* $\times$ *Glabron*) and Proctor segregated to give a good fit of 3 resistant : 1 susceptible plants when assessed 14 days after inoculation with isolate 1394 of *R. secalis* (table 2).

TABLE 2

The reaction of progeny from crosses involving Cb 1084 to isolate 1394 of R. secalis

Cross	Generation	Days after inoculation	Frequency of reaction types			Expected ratio	Probability
			0	2	4		
<i>Cb 1084</i> $\times$ Proctor	F_2	14	263	0	105	3 : 0 : 1	0.1-0.2
<i>Cb 1084</i> $\times$ Proctor	F_2	28	22	35	22	1 : 2 : 1	0.5-0.7

When re-assessed 28 days after inoculation, a number of resistant plants had developed type 2 symptoms, and the final ratio did not deviate significantly from 1 (type 0) : 2 (type 2) : 1 (type 4). This suggests that the resistance of *Cb1084* is controlled by a single gene which is incompletely dominant,

TABLE 3

The reaction of F_2 and F_3 progeny from crosses involving Turk, Atlas 46, Brier, Hudson, Dea and Modoc to isolate 1394 of R. secalis

Cross	Generation	Days after inoculation	Observed frequency			Expected ratio	Probability
			Res.	Seg.	Sus.		
<i>Turk</i> $\times$ Proctor	F_2	14	105	—	37	3 : 1	0.7-0.8
<i>Turk</i> $\times$ Proctor	F_3f^*	14 & 28	18	52	20	1 : 2 : 1	0.3-0.5
<i>Turk</i> $\times$ Proctor	F_{3s}^*	14 & 28	832	—	295	3 : 1	0.3-0.5
<i>Atlas 46</i> $\times$ Proctor	F_2	14 & 28	111	—	29	3 : 1	0.2-0.3
<i>Atlas 46</i> $\times$ Proctor	F_3f	14 & 28	16	49	18	1 : 2 : 1	0.2-0.3
<i>Atlas 46</i> $\times$ Proctor	F_{3s}	14 & 28	815	—	294	3 : 1	0.2-0.3
<i>Brier</i> $\times$ Proctor	F_2	14 & 28	216	—	65	3 : 1	0.3-0.5
<i>Hudson</i> $\times$ Proctor	F_2	14 & 28	231	—	64	3 : 1	0.1-0.2
<i>Dea</i> $\times$ Maris Otter	F_2	14 & 28	135	—	40	3 : 1	0.2-0.3
<i>Modoc</i> $\times$ Proctor	F_2	14 & 28	172	—	63	3 : 1	0.5-0.7
<i>Atlas 46</i> $\times$ Turk	F_3f	14	187	0	0	7 : 8 : 1	<0.01
<i>Brier</i> $\times$ Turk	F_3f	14	189	0	0	7 : 8 : 1	<0.01
<i>Brier</i> $\times$ <i>Cb 1084</i>	F_3f	14	193	0	0	7 : 8 : 1	<0.01
<i>Hudson</i> $\times$ Turk	F_3f	14	44	0	0	7 : 8 : 1	<0.01
<i>Modoc</i> $\times$ Turk	F_3f	14	189	0	0	7 : 8 : 1	<0.01
<i>Modoc</i> $\times$ Brier	F_3f	14	193	0	0	7 : 8 : 1	<0.01
<i>Modoc</i> $\times$ <i>Cb 1084</i>	F_3f	14	194	0	0	7 : 8 : 1	<0.01
<i>Dea</i> $\times$ <i>Cb 1084</i>	F_2	14	615	—	0	15 : 1	<0.01

* F_3f refers to F_3 families. F_{3s} to the total number of plants within segregating F_3 families only.

allowing a type 2 reaction to develop slowly when in the heterozygous condition. Confirmation of this hypothesis is available from tests on four F_1 hybrids from the same cross, which were completely resistant 14 days after inoculation, but developed type 2 symptoms within 28 days after inoculation.

The segregation of the progeny from crosses involving *Turk*, *Atlas 46*, *Brier*, *Hudson*, *Dea* and *Modoc* in response to inoculation with isolate 1394 are presented in table 3. When crossed with the susceptible varieties

Proctor or *Maris Otter*, all these resistant varieties gave F_2 or F_3 segregations compatible with the hypothesis that resistance in each variety was conferred by a single dominant gene which was fully effective at both 14 and 28 days after inoculation.

Crosses were also made between *Cb 1084* and *Brier*, *Dea* and *Modoc*; between *Turk* and *Atlas 46*, *Brier*, *Hudson* and *Modoc*; and between *Brier* and *Modoc*. In each case, all F_2 plants or F_3 families tested were completely resistant (see table 3).

The genetic control of resistance in *Osiris* was first examined in an F_3 population from the cross *Osiris* $\times$ *Proctor*, tested with isolate 1394. When

TABLE 4
The reaction of F_2 and F_3 progeny from crosses involving Osiris to isolates 1394 and 1436 of R. secalis

Cross	Generation	Days after inoculation	Observed frequency			Expected ratio	Probability
			Res.	Seg.	Sus.		
<i>Osiris</i> $\times$ <i>Proctor</i> *	F_3f	14	48	111	52	1 : 2 : 1	0.5-0.7
<i>Osiris</i> $\times$ <i>Proctor</i> *	F_3s	14	1631	—	546	3 : 1	0.9-0.95
<i>Osiris</i> $\times$ <i>Proctor</i> *	F_3f	28	14	113	84	1 : 8 : 7	0.5-0.7
<i>Osiris</i> $\times$ <i>Proctor</i> *	$F_3s^\dagger$	28	259	—	76	3 : 1	0.3-0.5
<i>Osiris</i> $\times$ <i>Proctor</i>	F_2	14	67	—	20	3 : 1	0.5-0.7
<i>Osiris</i> $\times$ <i>Proctor</i>	F_2	28	48	—	39	9 : 7	0.7-0.8
<i>Osiris</i> $\times$ <i>Turk</i> *	F_3f	14	243	0	0	7 : 8 : 1	< 0.01
<i>Osiris</i> $\times$ <i>Brier</i> *	F_3f	14	197	0	0	7 : 8 : 1	< 0.01
<i>Osiris</i> $\times$ <i>Atlas 46</i> *	F_3f	14	178	1	0	7 : 8 : 1	< 0.01
<i>Osiris</i> $\times$ <i>Atlas 46</i> *	F_3s	14	19	—	6	3 : 1	0.9-0.95
<i>Osiris</i> $\times$ <i>Turk</i> *	F_2	28	97	—	7	15 : 1	0.8-0.9
<i>Osiris</i> $\times$ <i>Modoc</i>	F_2	28	137	—	9	15 : 1	< 0.95
<i>Osiris</i> $\times$ <i>Modoc</i> *	F_3f	14	193	0	0	7 : 8 : 1	< 0.01

* Tested against isolate 1394 only.

† Considering only families resistant 14 days after inoculation.

assessed 14 days after inoculation, the segregation of this material gave a good fit to a ratio of 1 resistant : 2 segregating : 1 susceptible families, with a ratio of 3 resistant : 1 susceptible plants in the segregating families (table 4). Thus, at this stage, resistance was controlled by a single dominant gene. However, a re-assessment 28 days after inoculation showed that a number of plants which were resistant after 14 days had developed lesions; considering these as susceptible (although they varied in reaction from type 2 to type 3), the segregation then fitted a ratio of 1 resistant : 8 segregating : 7 susceptible families. Within the families resistant at 14 but segregating at 28 days after inoculation, the final ratio of resistant to susceptible plants did not differ significantly from 3 : 1. Seven F_1 hybrids were also tested, and were completely resistant at both assessment times.

These results suggest that the resistance of *Osiris* is controlled by two complementary genes. The first of these is incompletely dominant, being fully effective 14 days after inoculation, but subsequently permitting a type 2 reaction to develop when in the homozygous condition, and a type 3 reaction when heterozygous. The second gene is completely dominant, but operates only in conjunction with the first. Thus its presence was not

apparent at the first assessment, but it prevented the later development of symptoms, when either homozygous or heterozygous.

Some F_2 plants from the cross *Osiris* $\times$ *Proctor* were tested simultaneously against isolates 1394 and 1436, by inoculation of the second leaf with one and the third leaf with the other isolate. The segregations obtained with both isolates at 14 and 28 days after inoculation supported the conclusions drawn from the F_3 data, and each plant showed identical reaction to both isolates. Thus if two different gene pairs conferred resistance to the two isolates, they would have to be less than 3.3 cross-over units apart in each case.

TABLE 5

The reaction of F_2 and F_3 progeny from crosses involving La Mesita, C.I. 3515, C.I. 8256 and Gembloux 14 to isolate 1394 of R. secalis

Cross	Generation	Days after inoculation	Observed frequency			Expected ratio	Probability
			Res.	Seg.	Sus.		
<i>La Mesita</i> $\times$ <i>Proctor</i>	F_2	14	139	—	58	3 : 1	0.1-0.2
<i>La Mesita</i> $\times$ <i>Proctor</i>	F_2	28	115	—	32	9 : 7	0.5-0.7
<i>C.I. 3515</i> $\times$ <i>Proctor</i>	F_2	14	262	—	97	3 : 1	0.3-0.5
<i>C.I. 3515</i> $\times$ <i>Proctor</i>	F_2	28	84	—	72	9 : 7	0.5-0.7
<i>C.I. 8256</i> $\times$ <i>Proctor</i>	F_2	14	350	—	120	3 : 1	0.7-0.8
<i>C.I. 8256</i> $\times$ <i>Proctor</i>	F_2	28	78	—	64	9 : 7	0.7-0.8
<i>Gembloux 14</i> $\times$ <i>Proctor</i>	F_2	14	254	—	84	3 : 1	0.9-0.95
<i>Gembloux 14</i> $\times$ <i>Proctor</i>	F_2	28	44	—	36	9 : 7	0.8-0.9
<i>La Mesita</i> $\times$ <i>Osiris</i>	F_3f	14 & 28	197	0	0	7 : 8 : 1	<0.01
<i>C.I. 3515</i> $\times$ <i>Osiris</i>	F_3f	14 & 28	171	0	0	7 : 8 : 1	<0.01
<i>C.I. 8256</i> $\times$ <i>Osiris</i>	F_3f	14 & 28	194	0	0	7 : 8 : 1	<0.01
<i>Gembloux 14</i> $\times$ <i>Osiris</i>	F_3f	14 & 28	25	0	0	7 : 8 : 1	<0.01

F_3 populations from the crosses *Osiris* $\times$ *Turk*, *Osiris* $\times$ *Brier* and *Osiris* $\times$ *Modoc* were all completely resistant 14 days after inoculation with isolate 1394, but in the cross *Osiris* $\times$ *Atlas 46*, one family out of 179 was found to be segregating in a ratio of 3 resistant : 1 susceptible plants (see table 4). Thus the gene in *Osiris* which is active up to 14 days after inoculation is either closely linked to that in *Atlas 46*, and the segregating family arose as a result of genetic recombination, or the genes are allelic, and the segregating family arose as a result of an outcross to a susceptible variety; this was one of the crosses which were not protected at anthesis in the F_1 generation. F_2 progeny from crosses between *Osiris* and *Turk* and *Modoc* were assessed 28 days after inoculation, when a segregation not significantly different from 15 : 1 indicated that neither *Turk* nor *Modoc* carried the complementary gene present in *Osiris*.

The F_2 progeny from crosses of *La Mesita*, *C.I. 3515*, *C.I. 8256* and *Gembloux 14* with *Proctor* all segregated in ratios not significantly different from the ratios expected if resistance in these varieties was controlled by a gene system similar to that found in *Osiris* (see table 5). Crosses between these varieties and *Osiris* all produced F_3 populations which were completely resistant at both 14 and 28 days after inoculation, indicating that the gene pairs in these varieties were either identical, allelic or closely linked to the pair in *Osiris*.

When inoculated with isolate 1394, the segregation of F_2 progeny from

the cross *Jet* × *Proctor* indicated that resistance in *Jet* was controlled by two complementary recessive genes (table 6). The F_3 progeny from crosses of *Jet* with *Turk*, *Osiris*, *Brier* and *Modoc* were all completely resistant 14 days after inoculation, indicating that one of the recessive genes in *Jet* is allelic or closely linked to the dominant genes in these varieties, which also carry the other recessive gene.

TABLE 6

The reaction of F_2 and F_3 progeny from crosses involving Jet to isolate 1394 of R. secalis

Cross	Generation	Days after inoculation	Observed frequency			Expected ratio	Probability
			Res.	Seg.	Sus.		
<i>Jet</i> × <i>Proctor</i>	F_2	14	19	—	289	1 : 15	<0.05
<i>Jet</i> × <i>Turk</i>	F_3f	14	93	0	0	19 : 38 : 7	<0.01
<i>Jet</i> × <i>Osiris</i>	F_3f	14	91	0	0	19 : 38 : 7	<0.01
<i>Jet</i> × <i>Brier</i>	F_3f	14	94	0	0	19 : 38 : 7	<0.01
<i>Jet</i> × <i>Modoc</i>	F_3f	14	98	0	0	19 : 38 : 7	<0.01

The resistance of *Nigrinudum* to isolate 1394 is controlled by a single recessive gene, as shown by the segregation of F_2 plants from a cross with *Proctor* (table 7). The segregation of F_2 or F_3 progeny from crosses with *Turk*, *Modoc* and *Jet* indicate that the gene in *Nigrinudum* is inherited independently of the genes in these varieties.

TABLE 7

The reaction of F_2 and F_3 progeny from crosses involving Nigrinudum, C.I. 4364 and C.I. 4368 to isolate 1394 of R. secalis

Cross	Genera- tion	Days after inocula- tion	Observed frequency			Expected ratio	Probability
			Res.	Seg.	Sus.		
<i>Nigrinudum</i> × <i>Proctor</i>	F_2	14	55	—	139	1 : 3	0.2-0.3
<i>Nigrinudum</i> × <i>Turk</i>	F_2	14	127	—	33	13 : 3	0.5-0.7
<i>Nigrinudum</i> × <i>Modoc</i>	F_2	14	91	—	27	13 : 3	0.2-0.3
<i>Nigrinudum</i> × <i>Jet</i>	F_2	14	44	—	123	19 : 45	0.3-0.5
<i>Nigrinudum</i> × <i>Jet</i>	F_3f	14	13	30	5	19 : 38 : 7	0.9-0.95
<i>C.I. 4368</i> × <i>Proctor</i>	F_2	14	79	—	219	1 : 3	0.5-0.7
<i>C.I. 4368</i> × <i>Brier</i>	F_2	14	136	—	28	13 : 3	0.5-0.7
<i>C.I. 4368</i> × <i>Jet</i>	F_2	14	46	—	89	19 : 45	0.2-0.3
<i>C.I. 4368</i> × <i>Nigrinudum</i>	F_3f	14	24	34	6	7 : 8 : 1	0.3-0.05
<i>C.I. 4364</i> × <i>Proctor</i>	F_2	14	71	—	223	1 : 3	0.7-0.08
<i>C.I. 4364</i> × <i>C.I. 4368</i>	F_3f	14	42	0	0	7 : 8 : 1	<0.01

The results obtained with F_2 progeny from crosses with *Proctor* indicated that *C.I. 4364* and *C.I. 4368* each contained a single recessive gene for resistance. An intercross between the two varieties produced an F_3 population which was completely resistant, showing that the genes were at the same locus (table 7). However, this recessive gene was independent of that found in *Nigrinudum*, since the F_3 population from the cross *C.I. 4368* × *Nigrinudum* segregated to fit a ratio of 7 resistant : 8 segregating : 1 susceptible families.

Table 8 contains the results from crosses made to determine the location of the resistance genes in *Turk* and *Osiris*. The *Turk* × “uzu” and *Osiris* × “streaked” crosses, tested as F_2 seedlings in the glasshouse for both the genetic marker and disease reaction, indicated linkage distances of 8.3 ± 1.7 cross-over units between the locus of the resistance gene and *uz* and 7.7 ± 1.5 units between the resistance gene and *st*. The *Osiris* × “uzu” cross was observed in the field as F_2 plants for the “uzu” character, and the harvested F_3 seed was tested in the glasshouse for resistance 14 days after inoculation; a linkage value of 12.1 ± 3.8 cross-over units was obtained between the gene for resistance and *uz*.

TABLE 8

The reactions of F_2 progeny from crosses between the resistant varieties Turk and Osiris and stocks containing the genes uz, st and the interchange T2-3d

(a) Marker genes

Cross	Observed frequencies				Probability (9 : 3 : 3 : 1)	Probabilities	
	Normal		Mutant			Res. : Sus. (3 : 1)	Nor. : Mut. (3 : 1)
	Res.	Susc.	Res.	Susc.			
<i>Turk</i> × <i>uzuz</i>	201	12	12	68	< 0.01	0.3-0.5	0.3-0.5
<i>Osiris</i> × <i>uzuz</i>	71	5	5	14	< 0.01	0.2-0.3	0.2-0.3
<i>Osiris</i> × <i>stst</i>	363	25	15	123	< 0.01	0.05-0.1	0.5-0.7

(b) Interchange stock

Cross	Observed frequencies				Probability (3 : 1 : 3 : 1)	Probabilities	
	Fertile		Semi-sterile			Res. : Sus. (3 : 1)	Fer. : Ster. (1 : 1)
	Res.	Susc.	Res.	Susc.			
<i>Turk</i> × T2-3d	27	23	48	9	< 0.01	0.2-0.3	0.3-0.5
<i>Osiris</i> × T2-3d	51	41	82	14	< 0.01	0.1-0.2	0.7-0.8

F_2 plants from the crosses involving the chromosome interchange stock T2-3d were observed in the field for semi-sterility; the *Turk* × T2-3d progeny were also tested for resistance in the field, whilst the *Osiris* × T2-3d progeny were tested in the glasshouse for resistance as F_3 seedlings. The break-point in T2-3d was found to be linked at 13.9 ± 5.1 units from the resistance gene in *Turk*, and 13.4 ± 3.8 units from the gene in *Osiris* whose resistance is expressed 14 days after inoculation.

4. DISCUSSION

The results obtained from crosses involving *Turk*, *Atlas 46*, *Brier*, *Modoc* and *Cb 1084* suggest that the resistance genes in these varieties are all situated at the same locus, although the same results could be obtained if there were separate loci up to 1.7 cross-over units apart. It is also possible that *Hudson* and *Dea* contain genes at this locus, but the evidence for these varieties is less conclusive, since in the *Hudson* × *Turk* and *Dea* × *Cb 1084* (*Wisconsin Winter* × *Glabron*) crosses, recombination rates of up to 6.5 and 13.8 per cent. respectively could have passed undetected in the populations studied.

The gene in *Modoc* can be differentiated from the others at the same

locus by its ineffectiveness against isolate 1435, and that in *Cb 1084* by its incomplete dominance. The other five varieties, *Turk*, *Atlas 46*, *Brier*, *Hudson* and *Dea*, all exhibit identical patterns in both inheritance of resistance and reaction to the test isolates, which suggests that the resistance genes in these varieties are identical.

In the case of *Atlas 46*, this suggestion is further substantiated by its ancestry; the resistance was derived from *Turk* through a back-crossing programme (Schaller, 1951). Dyck and Schaller (1961a) and Starling *et al.* (1969) found two dominant genes in *Atlas 46*, only one of which was also found in *Turk*, the other being found in *Atlas*. This resistance gene was not detected in our studies since *Atlas* was susceptible to the three isolates of *R. secalis* tested. Frecha (1967) was also unable to identify more than one resistance gene in *Atlas 46*.

A single dominant gene for resistance, *Rh*, was found in *Brier* by Bryner (1957). Dyck and Schaller (1961a) also found a single dominant gene in *Brier*, which was identical, allelic or closely linked to one of two dominant genes in *Turk*. The genes in *Turk* were designated *Rh3* and *Rh5*, *Rh3* conferring resistance indistinguishable from that of *Brier* and *Hudson* in their tests with four physiologic races of *R. secalis*. The location of *Rh3*, relative to certain genetic marker loci (Dyck and Schaller, 1961b), was compatible with the relationships established for the gene found in *Turk* in this study.

Although Riddle and Briggs (1950) discovered more than one resistance gene in *Turk*, the additional gene was clearly not effective against isolate 1394, since no evidence of its presence was found in our tests. Wells and Skoropad (1963), Baker and Larter (1963) and Evans (1969) were likewise able to detect only one resistance gene in *Turk*. Starling *et al.* (1971) detected only one gene in *Turk*, which was identical, allelic or closely linked to the genes found in *Brier* and *Hudson*.

All this work supports our suggestion that the resistance genes we detected in *Turk*, *Atlas 46*, *Brier* and *Hudson* are identical. (*Dea* has not been studied genetically by other workers, but Williams (1969) was unable to differentiate *Dea* from *Turk*, *Atlas 46*, or *Brier* in a physiologic race survey in Britain.) However, physiologic race studies carried out by Schein (1958) do not support this suggestion. He used these varieties in a differential set, and found they gave different reactions to some of the seven U.S. races of *R. secalis* identified. However, Schein's tests were carried out under conditions which induced symptoms dissimilar to those obtained here and by Dyck and Schaller (1961a). Therefore, it is possible that the expression of resistance conferred by these genes can be influenced by the environment, the method of handling, or the genetic background in which the genes are located.

The resistance of *Modoc*, which we consider to be controlled by an allele of the gene in *Turk*, *Atlas 46*, *Brier* and *Hudson*, was also differentiated from these varieties in physiologic race tests by Schein (1958) and Dyck and Schaller (1961a). However, Dyck and Schaller considered the gene in *Modoc* not to be allelic, but closely linked to that in *Turk*, on the basis of crosses involving *Modoc*, *Atlas 46* and *La Mesita* (see below). The recessive gene discovered by Riddle and Briggs (1950) in *Modoc* was not identified here or by Dyck and Schaller.

Cb 1084 has not previously been studied genetically, but the inheritance of resistance in this variety was similar to that described by Baker and Larter

(1963) for the gene *Rh9* in *Kitchin* and *C.I. 668*. These workers did not test *Rh9* for allelism with other known dominant genes.

The inheritance of resistance in *La Mesita*, *Gembloux 14*, *C.I. 3515* and *C.I. 8256* was found to be identical to that in *Osiris*. In crosses between *Osiris* and these varieties, all the F_3 progeny were completely resistant, which limits the maximum possible recombination level between two gene pairs to less than about 1.9 per cent. Since all five varieties were resistant to the three test isolates, it is highly probable that they all contain the same pair of complementary genes.

Schein (1958) reported *La Mesita* to be susceptible to race U.S.-6, whereas *Gembloux 14*, *C.I. 3515* and *C.I. 8256* were resistant to this race (Schein, 1960), and Wells and Skoropad (1963) used an isolate which was pathogenic on *La Mesita* but not on *Osiris*. These apparent reaction differences may be attributable to differences in genetic background between varieties, rather than to specific major gene differences. In our experience, *La Mesita* develops disease symptoms in field nurseries, whereas *C.I. 8256* does not. However, when the resistance genes from *C.I. 8256* were back-crossed into an adapted genotype, these plants also exhibited disease symptoms in the field. Riddle and Briggs (1950) reported that susceptibility in adult and seedling plants of *La Mesita* is associated with senescence of the leaves.

When *Osiris* was crossed with *Turk*, *Brier* and *Modoc*, all the F_3 families were completely resistant 14 days after inoculation, suggesting that the effective gene in *Osiris* is allelic to the genes in these other varieties. However, the cross *Osiris* $\times$ *Atlas 46* produced one segregating family out of 179. This we considered to be the result of an outcross to a susceptible variety, rather than a recombinant, since the F_1 hybrids from this cross were not protected at anthesis. The frequency of natural outcrossing in normally self-fertilising barley genotypes has been variously estimated as less than 0.15 per cent. (Stevenson, 1928; Robertson and Deeming, 1931) or up to 2 per cent. (Jain and Allard, 1960).

Dyck and Schaller (1961a) obtained three segregating families in an F_3 population of 317 from the cross *Atlas 46* $\times$ *La Mesita*, and considered this too many to be attributable to outcrossing. On the basis of this and other crosses and physiologic race tests, they concluded that *La Mesita* and *Osiris* contained a resistance gene, *Rh4*, located 1.0 ± 0.78 crossover units from the gene in *Atlas 46* and *Turk*, and that *Modoc* contained an allele of this gene, which they designated *Rh4*².

Assuming that it was the same genes which were studied in these varieties by Dyck and Schaller and ourselves, it is important from the view of plant breeding to determine which of the two hypotheses is correct, since non-allelic genes can be coupled in a homozygous variety, whereas allelic genes cannot be combined. If our results are interpreted in accordance with Dyck and Schaller's hypothesis, the data from several crosses can be combined, giving a "recombination" value of about 0.1 per cent. which is well outside the standard error of Dyck and Schaller's estimate. Thus we consider that the genes for resistance in *Turk*, *Atlas 46*, *Brier* (Hudson, Dea), *Wisconsin Winter* $\times$ *Glabron*, *Modoc*, *Osiris*, *La Mesita* (*Gembloux 14*), *C.I. 3515* and *C.I. 8256* are located at the same locus.

The gene we detected in *Osiris* which is complementary to the one effective 14 days after inoculation has not been previously reported, presumably because its effect is not apparent until well after the normal times of assess-

ment, when seedling material being subjected to glasshouse tests would generally have been discarded. Many workers have considered the gene in *Osiris* and allied varieties to be identical to that in *Turk* and *Atlas 46* (Riddle and Briggs, 1950; Wells and Skoropad, 1963; Frecha, 1967), but this conclusion was always based on the results of tests using only one isolate of *R. secalis*.

Jet has previously been reported to contain two complementary recessive genes, designated *rh6* and *rh7* (Baker and Larter, 1963), but these were tested for allelism only with the recessive gene in *Nigrinudum*. In this study, it was confirmed that resistance was controlled by two complementary recessive genes, but one of these was shown to be situated at the locus carrying the dominant genes in *Turk*, *Brier*, *Modoc* and *Osiris*, whilst the other gene was also present in these varieties, but not in *Proctor* or *Nigrinudum* (nor in *Montcalm*, the susceptible variety used by Baker and Larter). In varieties other than *Jet*, this complementary gene appears to be neutral, since its effect was not noted in other crosses involving *Turk*, *Brier*, *Modoc* or *Osiris*. It is unrelated to the complementary gene in *Osiris*, which has been shown to be absent from *Turk* and *Modoc*.

The recessive gene isolated from *Nigrinudum* is probably that designated *rh8* by Wells and Skoropad (1963), and, as with these authors, our results show this gene to be inherited independently of all other genes. The recessive gene found in *C.I. 4364* and *C.I. 4368* has not been previously reported.

The results reported here indicate that the designations previously applied to certain genes are not completely valid. Since the gene designated *Rh3* in *Turk* and *Atlas 46* by Dyck and Schaller (1961a) is probably identical to the gene designated *Rh* in *Brier* by Bryner (1957), the latter symbol takes precedence. The same gene is also probably present in *Hudson* and *Dea*. It has also been shown that the *Rh4* locus of Dyck and Schaller is not separate from the *Rh* locus, so that the symbol *Rh⁴* is more suitable for the gene active 14 days after inoculation in *Osiris*, *La Mesita*, *Gembloux 14*, *C.I. 3515* and *C.I. 8256*. The gene complementary to *Rh⁴* has not previously been reported, and the symbol *Rh10* is suggested for it. Since the gene in *Modoc* has also been shown to be at the *Rh* locus, Dyck and Schaller's symbol *Rh⁴²* should be amended to *Rh²*. Also at this locus is the dominant gene in *Cb 1084*, for which the symbol *Rh³* is suggested.

One of the complementary recessive genes in *Jet* has also been shown to be situated at the *Rh* locus. Baker and Larter's (1963) designation of *rh7* could therefore be amended to *rh⁵*, whilst *rh6* is retained for the other gene, which is also present in *Turk*, *Brier*, *Modoc* and *Osiris*; consequently the genotypes of these varieties should be amended to include this gene. The designation *rh8* for the recessive gene in *Nigrinudum* should be retained, and the symbol *rh11* is suggested for the recessive gene in *C.I. 4364* and *C.I. 4368*. These changes in nomenclature for the genes for resistance to *R. secalis* are summarised in table 9.

The results of our studies show that there are five alleles at the *Rh* locus; two are dominant (*Rh* and *Rh²*), two are incompletely dominant (*Rh³* and *Rh⁴*) and one is recessive (*rh⁵*). The phenomenon of multiple allelism has been reported in several host-pathogen systems, e.g. at the *Mla* locus conditioning mildew resistance in barley (Moseman, 1966). However, the *Rp1* locus conditioning rust resistance in maize, once believed to have 13 alleles, has now been resolved into at least five separate loci with less than 0.4 per

cent. recombination between them (Saxena and Hooker, 1964, 1967). The detection of linkages of this order in barley would require the testing of at least 760 F_3 families from each cross.

Nevertheless, further investigations of this locus may prove valuable, since pseudoallelism would enable breeders to intercross resistant varieties and obtain new combinations of resistance genes, effective against wide spectra of pathogenicity.

TABLE 9

Suggested revision of the symbols allocated to barley genes conferring resistance to R. secalis, and allocation of symbols to new genes identified

Variety	Genetic Constitution	
	Previous	Amended
<i>Turk</i>	Rh3Rh3Rh5Rh5 (b)	RhRh(Rh5Rh5)rh6rh6
<i>Atlas 46</i>	Rh2Rh2Rh3Rh3 (b)	RhRh(Rh2Rh2)
<i>Brier</i>	RhRh (a)	RhRhrrh6rh6
<i>Hudson</i>	—	RhRh
<i>Dea</i>	—	RhRh
<i>Osiris</i>	Rh4Rh4 (b)	Rh ⁴ Rh ⁴ rh6rh6Rh10Rh10
<i>La Mesita</i>	Rh4Rh4 (b)	Rh ⁴ Rh ⁴ Rh10Rh10
<i>C.I. 3515</i>	Rh3Rh3 (c)	Rh ⁴ Rh ⁴ Rh10Rh10
<i>C.I. 8256</i>	Rh3Rh3 (c)	Rh ⁴ Rh ⁴ Rh10Rh10
<i>Gembloux 14</i>	—	Rh ⁴ Rh ⁴ Rh10Rh10
<i>Wisc. Winter × Glabron</i>	—	Rh ³ Rh ³
<i>Modoc</i>	Rh4 ² Rh4 ² (b)	Rh ² Rh ² rh6rh6
<i>Jet</i>	rh6rh6rh7rh7 (d)	rh ⁵ rh ⁵ rh6rh6
<i>Nigrinudum</i>	rh8rh8 (c)	rh8rh8
<i>C.I. 4364</i>	—	rh11rh11
<i>C.I. 4368</i>	—	rh11rh11

References: (a) Bryner, 1957. (b) Dyck and Schaller, 1961a. (c) Wells and Skoropad, 1963. (d) Baker and Larter, 1963.

The Rh genes referred to in parentheses signify genes identified by other workers but not detected in this study.

5. SUMMARY

1. Of the 18 barley varieties studied, 16 exhibited resistance to *R. secalis* controlled by five single genes and two pairs of complementary genes.

2. The dominance of these genes and their effectiveness against three isolates of the pathogen was established.

3. Five alleles or pseudoalleles were found at one locus on chromosome 3, whilst the remainder were inherited independently of this locus, and possibly of each other.

4. New or revised symbols are proposed for the designation of some of the resistance genes identified.

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