

Physiologic Differentiation of Wheat Stem Rust on Rye

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Abstract

Five self-fertile lines of *Secale cereale* possessing single genes conferring resistance to stem rust were successfully used to differentiate between strains of *Puccinia graminis tritici*. In an earlier study four of these lines were shown to differentiate between Australian cultures of *P. graminis secalis*. Gene H transferred from Wrens-5 rye to one of the five lines appears to be identical with gene *Sr27* in Acosta's wheat-Imperial rye translocation stock WRT 238.5.

Since, in the past, the genes for resistance in the rye lines have not been closely associated with *P. graminis tritici* and thus could not have exerted a selection pressure on the fungus, the present results imply that differentiation of a pathogen on host genotypes is, in the first instance, not due to specialization but is an intrinsic phenomenon of host-pathogen relationships. The results further suggest that fungal virulence *per se* is not associated with reduced fitness.

Introduction

The concept of 'strong and weak genes' for resistance to fungal pathogens (Van der Plank 1963) and the many theories based on this hypothesis all postulate that as fungal strains acquire genes for virulence, they lose fitness. Since parasitic fungi, which have the ability to multiply rapidly, have coexisted with their hosts over long periods of time, it has been argued that the presence of effective genes for resistance in the host is proof for an accompanied loss of such fitness in a fungus possessing the corresponding genes for virulence (Robinson 1976). This view was supported by findings which suggested that wild pathogen populations, i.e. those that were present before the release of resistant cultivars, lacked unnecessary genes for virulence (Watson 1970). If this is a general feature of fungal pathogens under field conditions, then it should be expected that a forma specialis of a pathogen adapted to one host, e.g. wheat, would give only avirulent interactions when tested on lines of another related species, e.g. rye, carrying single genes conferring resistance to its own forma specialis.

The main objective of the present study was to test this prediction by determining whether strains of wheat stem rust (*Puccinia graminis* Pers. f. sp. *tritici* Eriks. & E. Henn.) give differential interactions when inoculated on to five selected lines of rye (*Secale*). It had been shown earlier that four of these rye lines differentiate between Australian cultures of *P. graminis secalis* and that they possess different genes conferring resistance to formae speciales *tritici* and *secalis* of *P. graminis* (Tan *et al.* 1975, 1976, 1977).

Materials and Methods

Differential Hosts

The five self-fertile lines of *S. cereale* used in this study comprise progenies of F_3 lines from crosses between five resistant and two susceptible varieties. Four of the lines, namely those carrying genes provisionally designated *SrA*, *SrC*, *SrD* and *SrH*, have already been referred to above. The fifth line comes from the cross Wrens-6 \times Imperial rye and carries a factor(s) for resistance derived from the latter.

Cultures of P. graminis tritici

The five differential hosts were tested with 69 cultures; of these only 16 are described. They were selected from (1) Australian field strains, (2) exotic strains present in our rust collection, and (3) somatic hybrids between strains belonging to (2). The strains together with their culture numbers are listed below.

126-6,7,11 (culture 334). The predominant strain in Australia during the years 1927 to 1955. It differs from the post-1954 strains on the standard differentials by producing necrotic ('X') infection types on the five durumms.

21-0 (57043). Origin unknown; it appeared in 1954 in southern New South Wales. The majority of present Australian field strains have evolved from this original strain (Luig 1977).

194-1,2,3,5,6 (69642). Probably an introduction to Australia from Africa (Luig 1977).

32-2,11 (73560). The first strain belonging to standard race 34 which appeared in 1957. Probably a somatic hybrid between standard races 21 and 126 (Luig and Watson 1977).

194-2,3,7,8,9 (68642). It appeared first in 1968 when it attacked the common cultivar Festiguay which carries the Webster-type resistance. Possibly an introduction to Australia (Luig and Watson 1970).

57096. An unusual strain detected in 1957. It resembles standard race 34 but produces distinctly lower infection types on Marquis, Arnautka, Mindum and Spelmar.

60-L-4. Previous designation M2 (Watson and Luig 1962). A somatic hybrid between NR-2 orange and 80-E-0 grey-brown.

80-E-0 grey-brown. Obtained from St Paul, Minnesota, U.S.A. Because of its colour it is essentially a laboratory culture (Watson and Luig 1968).

NR-2 orange. A U.S.A. culture used in several studies to obtain somatic recombinants (Watson and Luig 1958, 1962).

15-2,4,5,7 (59-L-1), 214-5,7 (61-L-3) and NR-7 are somatic hybrids between NR-2 orange and 111-E-2 red (Watson and Luig 1958).

56-E-2 yellow. Obtained from the U.S.A. A laboratory culture.

59-51A, 38-51A and 36 \times 111. All are U.S.A. laboratory cultures.

Rust inoculations were made by using a pressure atomizer to disperse uredospores suspended in mineral spirit (Mobil Oil Australia Ltd.) over seedlings at the first leaf stage. After incubation in a misting chamber for about 18 h, the pots containing the seedlings were spaced out in a well-lit glasshouse. After 2 weeks, infection types were classified according to the key prepared by Stakman *et al.* (1962).

Results

The efficacy of the five rye genotypes for differentiating 16 strains of *P. graminis tritici* is shown in Table 1. Some of the stem rust cultures (126-6,7,11 and 57096) are clearly differentiated from all others by their reaction on the rye lines R1 and R17, whereas others (e.g. NR-7 v. 15-2,4,5,7 and 21-0 v. 194-1,2,3,5,6) show only minor differences from each other, and two (NR-7 and 21-0) are not differentiated at all.

Altogether 46 field strains and 23 laboratory cultures were tested on the rye lines but, with the exception of 57096, all field strains could be assigned to five 'origin' groups. These five groups correspond with those proposed recently (see Luig 1977). The first one includes two pre-1954 field strains and two derived laboratory strains,

and is represented by 126-6,7,11. This strain appeared in 1926 and dominated during the next two decades.

The second group is represented by 21-0, the original strain of standard race 21, which was found in 1954. The bulk of the present field inoculum in Australia belongs to strains which are believed to be descendants of 21-0. From this group 25 field strains and 8 laboratory strains (mutants induced by chemical mutagens) were examined, but their infection types were identical to those shown for 21-0 in Table 1.

Table 1. Infection types produced at 15–22°C by 16 cultures of *P. graminis tritici* and one culture of *P. graminis secalis* on five rye lines

Type of culture	Rust cultures		Host lines				
	Culture or strain No.		R1 SrA	R3 SrC	R4 SrD	R17 SrH	R25 Imp. gene
Australian field strains	126-6,7,11 (group 1)		3	3	3	;	1+
	21-0 (group 2)		;	2=	3	2=	2=
	194-1,2,3,5,6 (group 3)		;	2=	3	2=	3=
	34-2,11 (group 4)		;	2=	3	;	2=
	194-2,3,7,8,9 (group 5)		;	2=	3	2=	3+
	57096		;	2=	3	3	2=
Somatic recombinants and their parents	60-L-4		;	2	3	;	1+
	80-E-0 grey-brown		;	2=	2+	;	2=
	NR-2 yellow		;	2+	3=	;	2=
	NR-7		;	2=	3	2=	2=
	15-2,4,5,7		;	2=	3	2=	2+
	214-5,7		;	3	3	;	2=
Laboratory strains	56-E-2 yellow		;	2+	2	2=	1+
	59-51A		;	3	1+	2=	2=
	38-51A		;	3+	3	;	3+
	36×111		;	3	3	;	1+
<i>P. graminis secalis</i>	57241		;	2=	3	3+	3+

Strains of group 3 (represented by 194-1,2,3,5,6) differ in their behaviour from those of group 2 only on one line (R25, see Table 1). However, evidence has been presented (I. A. Watson and C. de Sousa, unpublished data; Luig 1977) which indicated that the original two strains of group 3 have been aerially transported to Australia from central Africa. The other five strains of this group we believe to be derivatives of the two exotic ones.

The infection types of eight strains of group 4 (34-2,11) resembled those of group 2 and group 3, but minor differences were apparent on line R17. These small differences in infection types were not due to micro-environmental variation but they occurred consistently with all of the different cultures assigned to each of the two groups. Earlier, Luig and Watson (1977) compared the behaviour of 34-2,11 on 14 wheat genotypes with that of standard races 21 (21-0) and 126 (126-6,7,11), and they concluded that the former has had its origin as a somatic hybrid between the latter two standard races. The other strains of group 4 are believed to be stepwise mutants from 34-2,11.

Apart from producing susceptible infection types on line R25, the six members of group 5 (194-2,3,7,8,9) are identical in their behaviour to those in group 2. Luig and Watson (1970) have discussed the possibility that the first two strains (194-2,3,7,8,9 and 21-2,3,7,8,9) of this group which attacked the common cultivar Festiguay (*Sr30* = Webster-type resistance) are introductions to Australia. The present finding, which reveals that these two strains and all their derivatives differ from the other field strains by being virulent on line R25, supports this hypothesis.

Culture 57096 was isolated from the susceptible cultivar Bencubbin in southern New South Wales in 1957, but has not been detected hereafter. Although possessing genes for avirulence (corresponding to wheat genes *Sr10* and *Sr20*) which are not present in other Australian field strains, this culture shows increased virulence on certain rye genotypes (Table 1 and unpublished data). Furthermore, it was established that 57096 is virulent on line 177 (W2691/W.R.T. 238.5) into which gene *Sr27*, derived from Imperial rye via Acosta's wheat-rye translocation stock W.R.T. 238.5 (Acosta 1963), had been transferred (Luig and Watson 1976). *Sr27* is ineffective against Australian strains of *P. graminis secalis* Eriks. & E. Henn. (rye stem rust). It is possible, therefore, to suggest that 57096 is not a true representative of *P. graminis tritici* but has resulted from somatic hybridization between *P. graminis secalis* and *P. graminis tritici* [probably (*tritici* × *secalis*) × *tritici*].

Of the five lines the one with *SrA* was highly effective in distinguishing the pre-1954 strains (namely 126-6,7,11 and derivatives) from the post-1954 strains. *SrH* in line R17 was ineffective only against culture 57096, with the other 15 strains showing minor differences. The line with *SrD* was semi-susceptible to all Australian strains but produced resistant infection types with three laboratory cultures.

Line R25 (gene *Imp*) was susceptible to two strains, semi-resistant to one (15-2,4,5,7) and exhibited a range of resistant infection types (e.g. 3 = to 194-1,2,3,5,6; 2 = to 21-0; 1+ to 126-6,7,11) with the remaining 13 strains. The line *SrC* proved to be a very good differential, especially when the laboratory cultures and their somatic hybrids were taken into account.

It has been postulated that all present Australian field strains of *P. graminis tritici* have descended as mutations from a few 'original' strains (Watson and Luig 1966; Luig 1977). Extensive testing of the strains available from our liquid nitrogen storage on several rye lines, including those mentioned in this study, has confirmed this hypothesis, and this information will be published elsewhere. It is, therefore, not surprising that the five lines of *S. cereale* did not differentiate between strains previously assigned to one 'origin group'. This lack of differentiation also shows that the genes for avirulence or virulence in the fungus which correspond with the genes for resistance in the *S. cereale* lines are relatively stable, e.g. the many strains tested of group 2 come from collections which were made over a period of more than 20 years, but they all produce identical infections.

It is interesting to compare the efficacy of the 12 international testers versus that of the five *S. cereale* lines in differentiating between the groups of Australian strains. Strains of group 1 and culture 57096 are distinguished on both sets (see under Materials and Methods and Table 1). Although the other four groups cannot be distinguished on the international set, minor differences between strains of groups 2, 3, and 4 are exhibited on the five rye lines, and strains of group 5 are clearly distinct by the susceptible infection types which they induce on R25.

Further studies were carried out to determine whether the genes in the five rye lines are identical with genes for resistance present in wheat-rye translocation or substitution stocks. Since R25 came from a cross involving Imperial rye, two addition lines, namely 'E' [chromosome 1R (see Shepherd and Jennings 1971)] and 'G' [chromosome 3R (Lee and Larter 1969)] of Chinese Spring \times Imperial rye, were of special interest. The stem rust resistance of line E was recognized in these studies and independently by W. Q. Loegering (personal communication) in Missouri, U.S.A. Pathogenic tests were conducted and they showed that line E and Shepherd's 1R-rye \times 1D-wheat translocation line (W.R.T.-2, derived from line E), together with wheat cultivars Kavkaz, Aurora, Mildress and Skorospelka, all involving chromosomes 1R of rye and 1B of wheat (see Zeller 1973; Mettin *et al.* 1973), are not susceptible to 194-2,3,7,8,9 and derived strains which possess additional virulence on plants with *Sr5*. The latter virulence is essential when testing Russian cultivars like Aurora, Kavkaz and Skorospelka which carry this gene. Thus it can be concluded that gene *Imp* is different and distinguishable from those transferred to wheat.

On the other hand, the resistance in R17 (gene *SrH*) could be the same as the one transferred from line G to hexaploid wheat and recently designated *Sr27* (see above). Line 177 (*Sr27*) and R17 behave similarly not only to strains of *P. graminis tritici* and *P. graminis secalis* but also when tested with cultures which originated on rough wheat grass, *Agropyron scabrum* Beauv. The pathogenic characteristics of such cultures suggest that they may be hybrids between *P. graminis tritici* and *P. graminis secalis* (Luig and Watson 1972).

Discussion

Most species of pathogenic fungi comprise 'physiologic strains' which can be distinguished from each other by physiologic characteristics, mainly pathogenicity. Strains belonging to the formae speciales of *P. graminis* can also be differentiated on host genotypes of species other than the one on which they have specialized.

As early as 1929 Waterhouse selected single plants of March and Petkus rye on the basis of their different reactions to race 34 of *P. graminis tritici*. For several generations progenies of such plants were tested, raised and selfed. Waterhouse and Watson (1941) utilized 31 of these inbred lines to compare Australian race 34 with a U.S.A. culture of 34, and they noted different reactions on 17 lines. The present study goes further by demonstrating unequivocally that rye genotypes can successfully substitute for wheat differentials. Since the genes for resistance present in these rye genotypes have had no close association with *P. graminis tritici* and thus could not have exerted a selection pressure on the pathogen, the differential behaviour of strains of the latter must be due to an intrinsic occurrence in host-pathogen relationships. These findings have great significance for all modern concepts of disease resistance by implying that virulence *per se* is not associated with decreased fungal fitness.

The above conclusions are supported by an earlier study (Luig and Watson 1972) which showed that a parallel situation exists in *P. graminis secalis*. Cultures of rye stem rust collected in New South Wales proved virulent on the well known wheat gene *Sr11* when present in the highly susceptible wheat line W3498. This gene has played a prominent role in the evolution of *P. graminis tritici* in North America, Australia and other wheat growing regions. Initially *Sr11* provided comprehensive

protection, but when cultivars with this gene were extensively cultivated strains virulent on plants with *Sr11* became established. Van der Plank (1963) classified this gene as a moderately strong gene but in Australia for nearly a decade all prevalent strains of *P. graminis tritici* have shown virulence on plants with *Sr11*. Since many leading Australian cultivars (e.g. Pinnacle, Gamenya, Olympic, Insignia and Falcon) do not possess *Sr11* it appears that now in the wheat stem rust fungus too the capacity to attack plants with *Sr11* is no longer associated with loss of aggressiveness. In Europe, by contrast, *Sr11* still confers effective resistance.

The discovery that certain genes of *Secale* are effective in differentiating strains of wheat stem rust has also great importance in regard to the classification of strains of this forma specialis. Hitherto pathological tests were carried out utilizing the 12 standard differentials and several supplementals. The inadequacy of the former has been pointed out above, and most supplementals possess single genes which have been used in the breeding program. Since mutations to virulence on plants with these genes occur frequently, the value of these genes in discriminating between strains of different origin is greatly reduced. This does not apply to the rye lines as heretofore our studies have not revealed a single case where the same factor for resistance is shared by the genera *Triticum* and *Secale*. However, if resgenes from *Secale* transferred to wheat were utilized in breeding programs, their effectiveness in distinguishing between inherently different strains of *P. graminis tritici* will be negated.

As mentioned above, line 177 with *Sr27* proved susceptible to culture 57096. Previously this gene was thought to confer 'universal resistance' to *P. graminis tritici* (Luig and Watson 1976) but the present result suggests that *Sr27* is unlike *Sr26*. The latter gene acts alone in three popular Australian cultivars (Eagle, Kite, and Jabiru), but hitherto all isolates of stem rust have proved avirulent on plants with it. This failure to detect virulent mutants may be due to a genetic make-up of the pathogen which never gives rise to a viable mutation or the performance of *Sr26* may constitute an extreme case of durability of a gene for resistance.

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References

- Acosta, A. C. (1963). The transfer of stem rust resistance from rye to wheat. *Diss. Abstr.* **23**, 34-5.
- Lee, Y. H., and Larter, E. N. (1969). Homologous relationship of rye chromosome VI with two homologous groups from wheat. *Can. J. Genet. Cytol.* **11**, 803-9.
- Luig, N. H. (1977). The establishment and success of exotic strains of *Puccinia graminis tritici* in Australia. *Proc. Ecol. Soc. Aust.* (In press.)
- Luig, N. H., and Watson, I. A. (1970). The effect of complex genetic resistance in wheat on the variability of *Puccinia graminis* f. sp. *tritici*. *Proc. Linn. Soc. N.S.W.* **95**, 22-45.
- Luig, N. H., and Watson, I. A. (1972). The role of wild and cultivated grasses in the hybridization of formae speciales of *Puccinia graminis*. *Aust. J. Biol. Sci.* **25**, 335-42.
- Luig, N. H., and Watson, I. A. (1976). Strains of *Puccinia graminis* virulent on wheat plants carrying gene *Sr27* derived from Imperial Rye. *Phytopathology* **66**, 664-6.
- Luig, N. H., and Watson, I. A. (1977). The role of barley, rye and grasses in the 1973-74 wheat stem rust epiphytotic in southern and eastern Australia. *Proc. Linn. Soc. N.S.W.* **101**, 65-76.

- Mettin, D., Blüthner, W. D., and Schlegel, G. (1973). Additional evidence on spontaneous IB/IR wheat-rye substitutions and translocations. Fourth Int. Wheat Genetics Symp., Columbia, Missouri, U.S.A., pp. 179-84.
- Robinson, R. A. (1976). 'Plant Pathosystems'. pp. 184. (Springer Verlag: Berlin.)
- Shepherd, K. W., and Jennings, A. C. (1971). Genetic control of rye endosperm proteins. *Experientia* **27**, 98-9.
- Stakman, E. C., Stewart, D. M., and Loegering, W. Q. (1962). Identification of physiologic races of *Puccinia graminis* var. *tritici*. Agric. Res. Sta., U.S. Dept. of Agriculture, E617. (Revised.)
- Tan, B. H. (1973). The genetics of resistance to *Puccinia graminis* Pers. in the genus *Secale*. Ph.D. Thesis, University of Sydney.
- Tan, B. H., Watson, I. A., and Luig, N. H. (1975). A study of physiologic specialization of rye stem rust in Australia. *Aust. J. Biol. Sci.* **28**, 539-43.
- Tan, B. H., Luig, N. H., and Watson, I. A. (1976). Genetic analysis of stem rust resistance in *Secale cereale*. I. Genes for resistance to *Puccinia graminis* f. sp. *secalis*. *Z. Pflanzenzücht.* **76**, 121-32.
- Tan, B. H., Luig, N. H., and Watson, I. A. (1977). Genetic analysis of stem rust resistance in *Secale cereale*. II. Genes for resistance to *Puccinia graminis* f. sp. *tritici*. *Z. Pflanzenzücht.* **79**, 299-309.
- Van der Plank, J. E. (1963). 'Plant Diseases: Epidemics and Control'. (Academic Press: New York.)
- Waterhouse, W. L., and Watson, I. A. (1941). Australian rust studies. VI. Comparative studies of biotypes of race 34 of *Puccinia graminis tritici*. *Proc. Linn. Soc. N.S.W.* **66**, 269-75.
- Watson, I. A. (1970). Changes in virulence and population shifts in plant pathogens. *Annu. Rev. Phytopath.* **8**, 209-30.
- Watson, I. A., and Luig, N. H. (1958). Somatic hybridization in *Puccinia graminis* var. *tritici*. *Proc. Linn. Soc. N.S.W.* **83**, 190-5.
- Watson, I. A., and Luig, N. H. (1962). Asexual intercrosses between somatic recombinants of *Puccinia graminis*. *Proc. Linn. Soc. N.S.W.* **87**, 99-104.
- Watson, I. A., and Luig, N. H. (1966). Sr15—a new gene for use in the classification of *Puccinia graminis* var. *tritici*. *Euphytica* **15**, 239-50.
- Watson, I. A., and Luig, N. H. (1968). Progressive increase in virulence in *Puccinia graminis* f. sp. *tritici*. *Phytopathology* **58**, 70-3.
- Zeller, F. J. (1973). IB/IR wheat-rye chromosome substitutions and translocations. Fourth Int. Wheat Genetics Symp., Columbia, Missouri, U.S.A. pp. 209-21.

